Summary of Seahorse Population and Distribution

Koh Rong Samloem
Preah Sihanouk, Cambodia

Report on seahorse demographics and habitats

Marine Conservation Cambodia
1st Quarter Report 2013 – Jan/Feb/Mar

Photo 1 –*H. Spinosissimus* on the Corral, MCC 2009

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Abstract

This is the first quarter report of 2013 for the seahorse population and habitat surveys undertaken, starting with an initial survey done in June 2011 and continuing in August 20012 through to December 2012. This report covers January, February, and March of 2013.

The aim of the ongoing survey has been to assess and monitor the changing conditions of the study site, called the Corral, off the East coast of Koh Rong Somloem, particularly in regards to the local seahorse population found there.

During the first quarter of 2013 there were a total of 48 seahorses observed over a total of 90 surveys. Of the 48 seahorses recorded at our survey site 47 were identified as *H. spinosissimus* and 1 as *H.kellogi*. Population demographics, depths, and holdfast selection were also recorded and compiled with all previous data.

Due to the ongoing and continuous nature of these surveys there is a better understanding of the local seahorse populations, their behavior, depth range, migratory patterns, yearly movements and distribution within the study site.

Through the ongoing continued research it is hoped to establish a database of the conditions of this diverse and ecologically important area over a long period of time so that the data collected can be used to protect this fragile ecosystem.

By establishing relationships between species composition and diversity, depth, preferred holdfasts and holdfast densities, habitat cover, sexual demographics and reproductive activity it is possible to more effectively design and implement an effective conservation strategy as well as monitor its success over time.

All of this will lead to a better understanding for the long term protection of this fragile species and sensitive habitat.

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Acknowledgements

Marine Conservation Cambodia (MCC) has been working on conservation and community livelihoods in collaboration with the Royal Government of Cambodia Fisheries Administration (RGC FiA), local authorities and local communities since 2008.

The Marine Monitoring and Marine Research programs around Koh Rong and Koh Rong Samloem are now well underway and are currently undertaking marine surveys around Koh Rong Samloem, this is to monitor the Seahorse populations and the coral reefs, so it is possible to assist the FiA in the creation of Marine Fisheries Management Areas (MFMAs), Cambodia's equivalent to Marine Protected Areas (MPAs).

Close collaboration with the FiA and international institutions such as the FAO Regional Fisheries Livelihoods Programme (RFLP), The Seahorse Trust (UK), Save Our Seahorses (Ireland) has proven that MCC is now a respected and credited leader in conservation and community work in Cambodia.

Special Thanks Too

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Mr. Ing Try	Deputy Director of the Fisheries Administration
Mr. Ouk Vibol	Director of Fisheries Conservation Division
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Introduction

Cambodia has a unique marine environment that has an unusual array of species and a diverse range of habitats. As part of an ongoing, long term survey of the seahorses in southern Cambodia a site was chosen within a specific area off the small island of Koh Rong Samloem Island, 28km due south west of the port of Sihanoukville. The chosen site is known locally as the Corral and the surveys were conducted throughout June/July of 2011 and again in November/December of 2011, which were followed in August/September/October/November and December 2012. This report introduces data recorded in the first quarter of 2013 (Jan., Feb., and March) and looks to integrate all of the surveys into a cohesive account.

Population assessments provide a useful tool for measuring the current condition and viability of a specific population allowing for accurate estimates of abundance and structure of organisms within a studied area. Each survey undertaken provides a static picture of the condition and abundance of organisms and bottom composition for our selected area. When done in comparison to later surveys and looking back at previous surveys on the same sites, patterns start to emerge that will be beneficial to understanding the behaviour, migration, and distribution of the seahorses.

The assessment will therefore allow the seahorse population of the Corral site to be tracked and the effects of disturbance, such as destruction from trawling boats, to be monitored over long periods of time. Other trends, such as shifts in the dynamics of the species composition and age structure can also be observed over time. By comparing the previous set of data from the previous year with the new survey data in this report it is hoped to gain insight into the changing population and distribution demographics, as well as species composition and age structures within our study site.

As more surveys are performed an accurate trend of what is really happening at the study site will begin to emerge. It is vitally important to have a clear understanding of the conditions and number of organisms throughout the study area, so that management protocols can be efficiently implemented and effective conservation and monitoring strategies designed. Furthermore, it is vital to recognize habitat degradation and consequently population decline early on, so that effective measures can be put into place to mitigate and alleviate the pressures causing it.

Study Area

Koh Rong Samloem Island is located 28 hours south-west of Sihanoukville, a port city on Cambodia's southern coast. The island's coastline is largely shallow, composed mainly of sand flats, seagrass beds and coral reef habitats. Previous studies have identified 5 geographically separated coastal areas of seahorse habitat, designating one particular area, the Corral site, as a location for targeted seahorse research, due to its large breeding populations and close proximity to Marine Conservation Cambodia (MCC) facilities.

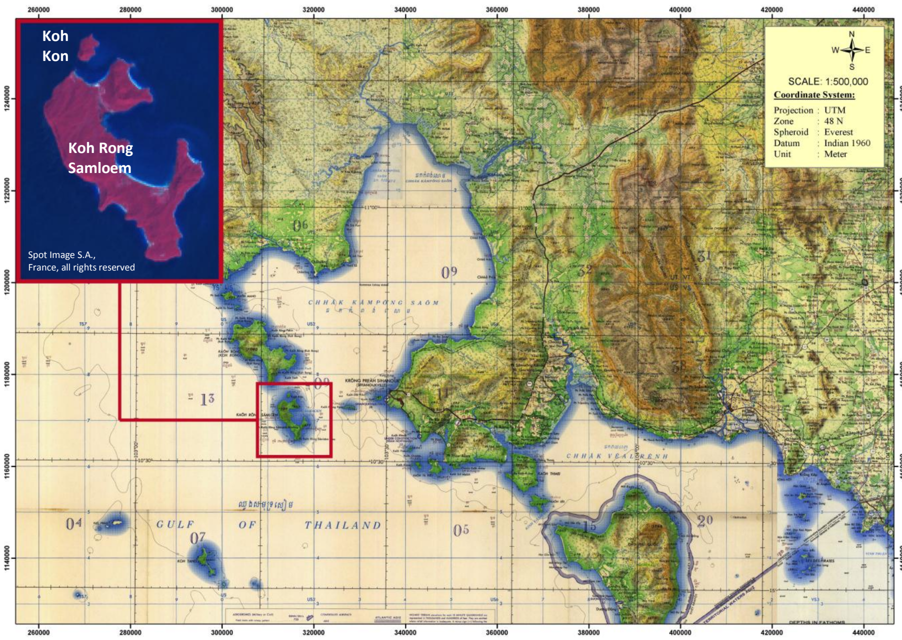

Figure 1: Map of Southern Cambodia and islands with magnification of Koh Rong Somloem

The Corral site is located to the east of Koh Koun, a small island located off the northern coast of Koh Rong Somloem. The area is dominated by sand flats, which slope gradually from the east coast of Koh Koun, with depths ranging between 5-20m. The area supports populations of bivalves, soft corals, hydrozoans and large numbers of pencil urchins (*Prionacidaris spp*), which provide valuable holdfasts for seahorses.

Species diversity of the area has been observed to be unusually high, with 6 species of seahorse identified from photographic evidence taken at the Corral site. These species are *Hippocampus spinosissimus*, *Hippocampus trimaculatus*, *Hippocampus kuda*, *Hippocampus comes*, *Hippocampus kelloggi*, *Hippocampus histrix*, (*Hippocampus barbouri* is suspected by has not been photographed) with *H. spinosissimus* heavily dominating the population particularly.

The habitat was observed to be in excellent condition in 2007, but damage from illegal trawling activity has greatly impacted the habitat since, reducing the biodiversity and productivity of the local ecosystem. Field observations from 2007 suggest that seahorse species diversity was previously higher, and has decreased over a very short period of time to strongly favor *H. spinosissimus*.

Legal protection of the habitat has been established in the form of a 300m No Take Zone (NTZ) extending from Koh Koun Island as well as a community conservation area. The 300m NTZ only covers a small percent of the study site, however the community conservation area covers far more of the area. Unfortunately protection measures are often ignored or circumvented, however, and frequent monitoring is necessary to prevent trawling activity in the area. Regularly conducted population assessments provide the consistent data necessary to measure the recovery of this area, and to make comparisons to its previously observed productivity of the ecosystem.

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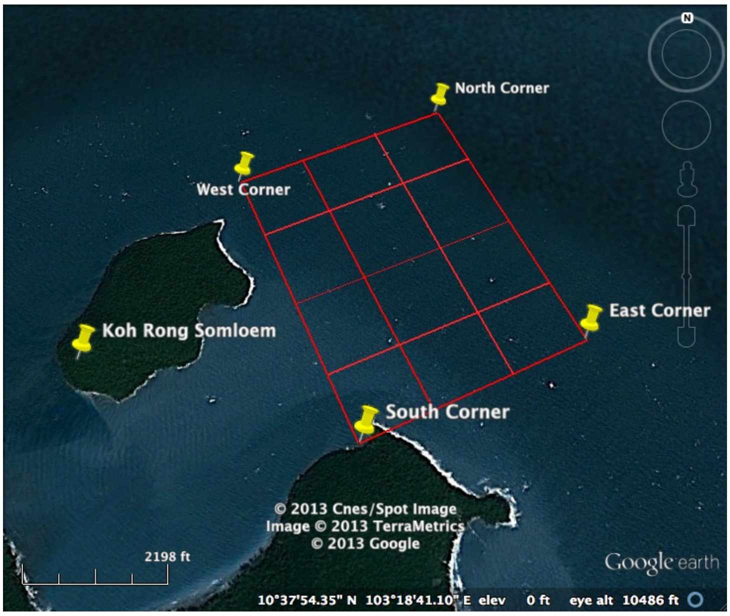
Methods



Layout of the structure of the grid pattern for surveying the Corral.

The population assessment was conducted through underwater visual transects that were semi-randomly located within the 2.52E6m area rectangle Corral study area. The starting point of each 500m2 transect is dependent upon a grid system whereby the entire seahorse area is divided evenly into 12 sections. Two GPS co-ordinates are chosen from one grid each day and surveyed. This means that each day they are randomly selected from a different grid, ensuring that each grid is selected at least once in the month before we start repeating grids. In order to have 30 sites surveyed per month inevitably some grids end up repeating more than others due to the fact there are only 12 grids. Directions are also randomize for the survey by choosing from eight options (i.e. N, NE, E, SE, S, SW, W, NW) for each survey point and making sure these are evenly distributed in any one grid. Choosing random directions from random grids ensures the same direction is not favored in any particular grid.

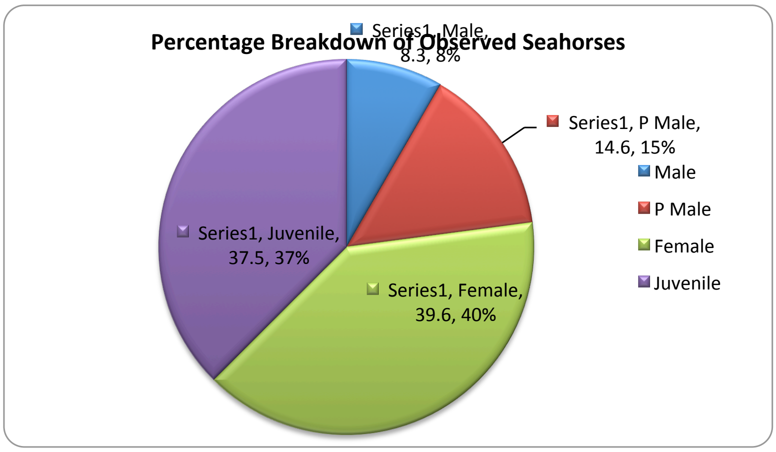
Transects were created by laying two 50m lines parallel, spaced 5m apart, projecting from the starting point in the randomly assigned direction. Two divers swim from the original side on either side of the first transect line, each surveying the 2.5m areas adjacent to the tape. At the far end of the tape, the divers would swim to the second tape and survey the 2.5m on either side going the opposite direction. The total surveyed area for each transect was 500m_2.

Seahorse species, demographic class, trunk and snout length, and associated habitat were recorded for each seahorse specimen within the transect area. Juveniles were defined as any seahorse with a trunk length under 2cm, and were not distinguished by sex due to difficulties in differentiating small individuals without fully developed sexual and species characteristics. Counts of pencil urchins, soft corals, anemones, seagrass, hydrozoans, sea pens and manmade structures were also recorded.

Estimates of the type of substrate cover were determined by swimming in a 1m circle with the centre point being along the transect line, by analysing the area it was possible to estimate the percentage of substrate area covered by benthic organisms.

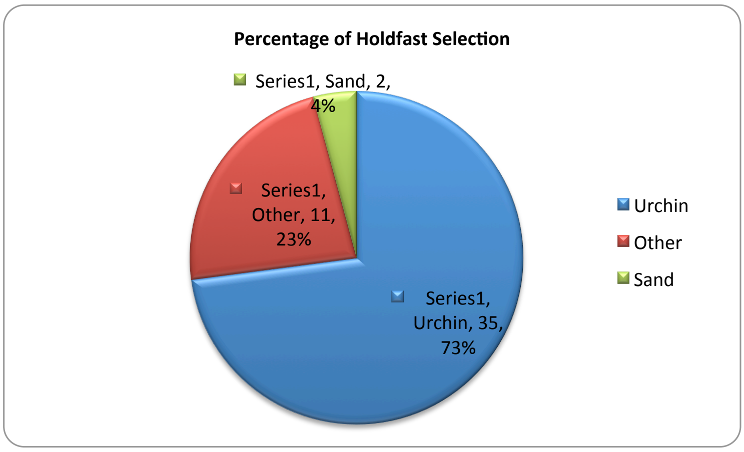
Results

During the first quarter of 2013 there were a total of 48 seahorses found over 90 surveys. Of the 48 seahorses recorded 47 were identified as H. spinosissimus and 1 was identified as H. Kellogi. The population demographic breaks down to 4 males, 7 pregnant males, 19 females, and 18 juveniles, with percentages of 8%, 15%, 40%, and 37% respectively. As is shown in Graph 1.



Graph 1: Sexual demographics of seahorses observed Jan/Feb/Mar 2013

Holdfast selection favored pencil urchins with 35 seahorses found there. Of the remaining 15, 2 were found on sand and the other 11 were observed attached to stable non-mobile holdfasts selections. These results can be seen below in Graph 2.



Graph 2: Holdfast selection for observed seahorses Jan/Feb/Mar 2013

The relationship between seahorses/pencil urchins, pencil urchins/percentage shell cover, and shell cover percent/seahorses were all found to be very significant. The values can be seen below in Table 1. The p-value refers to the probability that the results of the sample were arrived at by chance. In layman's terms, the lower the p-value, the less chance there is that the sample data is suggesting a relationship when there really is no relationship at all.

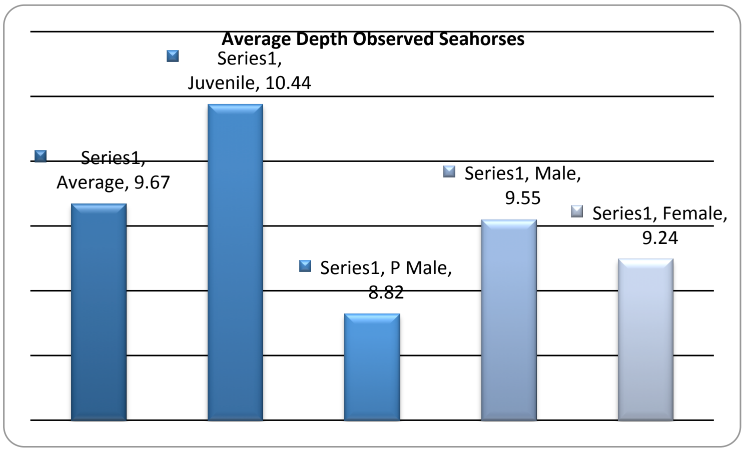
Relationship	P-value
Seahorses vs Pencil Urchins	1.53988E-09
Pencil Urchins vs Shell Cover %	4.23675E-07
Shell Cover % vs Seahorses	1.15101E-06

Table 1: P-value for relationships of seahorses, pencil urchins, and shell cover for Jan/Feb/Mar 2013 using a paired T-test

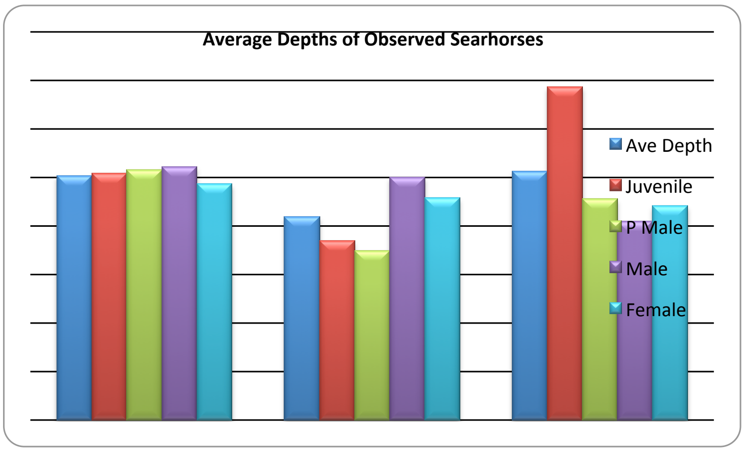
The average depth of all seahorses found observed in the first quarter of 2013 was

9.67m. With juveniles averaging 10.44, males 9.55, females 9.24, and pregnant males 8.82, as seen below in Graph 3. The averages per month of this first quarter can be seen in Graph 4.

Note: From observational data outside the study area H. kuda, H. trimiculatus, and one as of yet identified species can be found more frequently at depths between 4 and 9m.



Graph 3: Average depth of all seahorses recorded in Jan/Feb/Mar 2013



Graph 4: Average depth of seahorses recorded by month

Depths of seahorses observed from all past surveys can be seen in Graphs 11 and 12 in the Discussion section (pg 22 and 23)

Discussion

This ongoing survey is starting to reap rewards in consistent data and a pattern is now starting to emerge as to the seahorse species, their habitat preference, depth found throughout the year, and sex ratios on the site.

In this discussion we look at all of these aspects and try to understand the implications of all of these factors on the study site at the Corral.

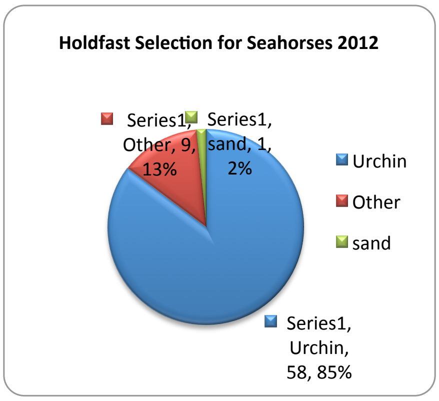
Holdfast and habitat preference

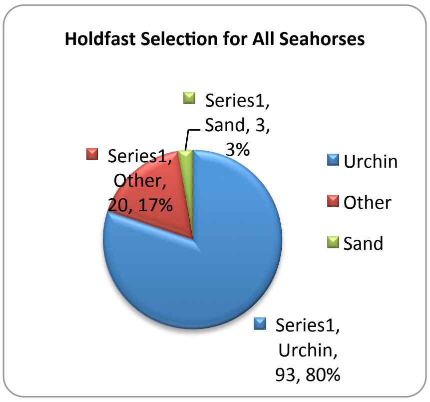
The correlation between pencil urchin population density to the percentage cover of broken shell on the seabed and seahorses found, were again all found to be significant relationships, and it is supposed that the lack of solid complete objects for the seahorses to secure themselves to, shows that the illegal trawling and bottom gill netting is having a negative effect on the seahorse populations by removing the solid holdfasts.

Photo 2: Trawling by-catch Koh Rong Somloem, Cambodia, 2008

Traditionally, based on studies and observation of other species, in a prime undamaged seahorse habitat, they would be attached to solid non-motile holdfasts such as seafans, coral, seapens, sponges and the like. However, at the Corral due to the destructive nature of the trawling, solid holdfasts are in limited supply, so the seahorses have adapted to using other items such as pencil urchins. In the 2012 surveys where these are not available they are either drifting on the seabed or have moved out of the area. There are now signs that this might be changing as described below.

The seahorse holdfast selection for Jan/Feb/Mar of 2013 was recorded to again be composed primarily of pencil urchins with 73% of seahorses observed. There were 23% found on other stable holdfasts and 4% found on sand. This is interesting when compared to last year's data. In all of 2012 the percentage of seahorse found on pencil urchins was 85% as shown in Graph 5. This is an increase in the percentage of stable holdfasts selection of 73.5% in just 3 months. It is possible that this increase in the observation of non-motile holdfasts is an indication that the habitat is recovering to its former state with stable holdfasts becoming more prevalent. The total holdfast percentages for all surveys can be seen in Graph 6. It is clear from the data collected that seahorses are being found more often on stable holdfasts. Further studies will be able to clarify whether or not this is due to an increase in the availability of stable holdfasts through decreased trawling activity and increased habitat restoration.



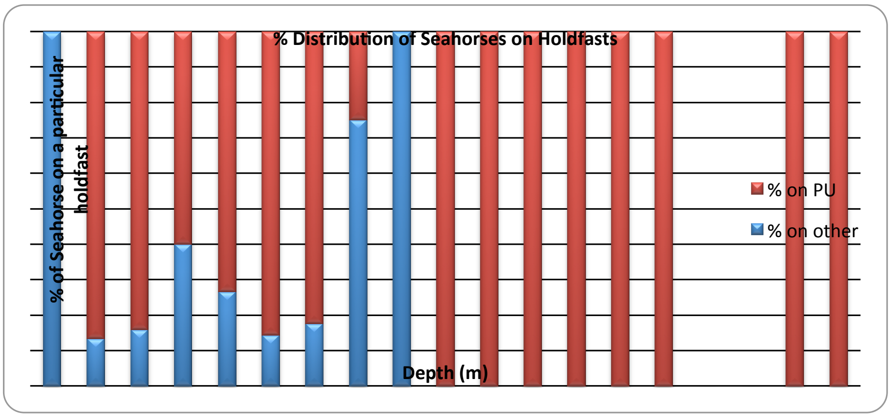


Graph 5 (Left): Holdfast selection for all seahorses observed from Aug 2012-Dec 2012

Graph 6 (Right): Holdfast selection for all seahorses observed from Aug 2012-March 2013

Note: Because pencil urchins are mobile it raises interesting questions on territoriality and migration patterns, which may disrupt their natural reproductive behavior.

In Graph 7 you can see the percent distribution of seahorses on pencil urchins vs. other holdfasts by depth. A single seahorse represents depths 6, 14, 18, 19, 20, 23, and 24, and is responsible for the 100% bars shown. Depths 15, 16, and 17 however, are represented by 15 seahorses (13% of total) all of which were located on pencil urchins. Which may indicate a boundary between areas that are still being trawled and shallower areas where trawling has decreased. This is supported when depths 7-13 are inspected, where the majority of seahorses are being observed, because it is starting to show signs of increased selection of solid holdfasts. These shallower areas are obviously closer to the island and farther into the no-take protected area. If this increased protection is the cause for the rise of available solid holdfasts (through not being trawled) and the subsequent increased frequency of selection, as well as a possible factor for the larger percentage of observed seahorses in general, will be determined with time and further observation. While Graph 7 does not offer a tremendous amount of insight at this time, in the long term it may provide information on recovery rates of the seafloor by depth through the choice of holdfast selection.



Graph 7: Percent distribution of holdfast selection by depth Aug 2012-March 2013

Picture 3: *H. spinosissimus* on pencil urchin at the Corral, Koh Rong Somleom, Cambodia – May 2009

Observational data from as early as 2007 indicates that seahorses have been using pencil urchins since the site was first being visited, prior to when the major trawling occurred. It is possible that the local populations, or at least *H. spinosissimus*, have been using these urchins as holdfasts pre-trawling simply because of there natural abundance in the area, and have only now, because of the trawling damage, become so heavily reliant on them. Future study on holdfast selection, when given options for stable objects, will be able to clarify the distinction.

Note: This relationship of seahorses using pencil urchins as holdfasts has also been observed in Thailand and Malaysia

Another interesting point of notice is that there are routinely extremely strong currents present at our study site. On several occasions it has been observed that the currents will actually take the pencil urchins with them, bouncing them along the seafloor like tumbleweed. This again raises interesting questions on holdfast selection and territoriality. How can the local population be expected to maintain a territory if the currents are far to strong to swim against and their primary holdfast is also no match? Further study on current strength and direction will hopefully provide clarification on the how the local population has adapted to these conditions.

Pre trawling it was recorded that there were 6 species of seahorse in large numbers; only the *Hippocampus spinosissimus* is present in any significant numbers during 2012 and the first quarter of 2013, leading to the conclusion that they are best adapted to this fragmented habitat. However, in December 2012 the first sighting of a species other than H. *spinosissimu*s was recorded since November 2011, with 1 H. *kuda* positively identified in our study site. In February of 2013 there was also the first sighting of H. *kellogi* made since November 2011. While it is too early to make any assumptions, it is hoped that these sightings of seahorses previously thought to have moved from the area, combined with the increase in number of seahorses attached to stable non-motile holdfasts may indicate the beginning of the habitats recovery to its former condition.

In the long term, as the original habitat continues to recover, it is hoped that other seahorse species will return to the area in greater numbers.

Seahorse population

During the first quarter of 2013 the average number of seahorses seen per dive is 0.53 with 48 seahorses recorded over 90 surveys. In January, February, and March the average seahorses observed per dive were 0.68, 0.45, and 0.47 respectively. It would appear that since November 2012, with the exception of December, the number of seahorses is leveling off around 0.5 per dive.

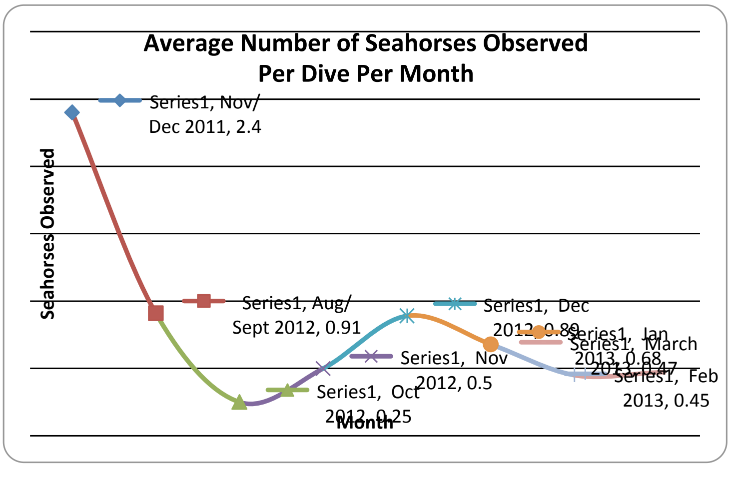
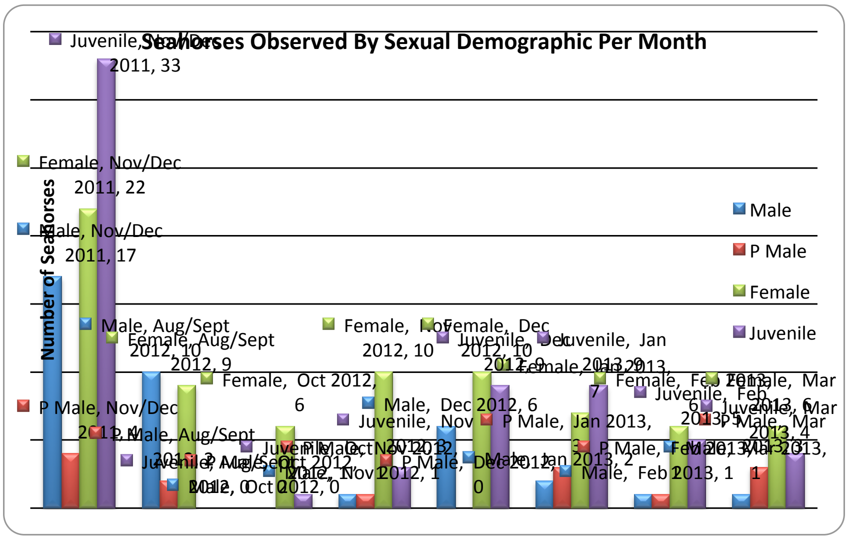


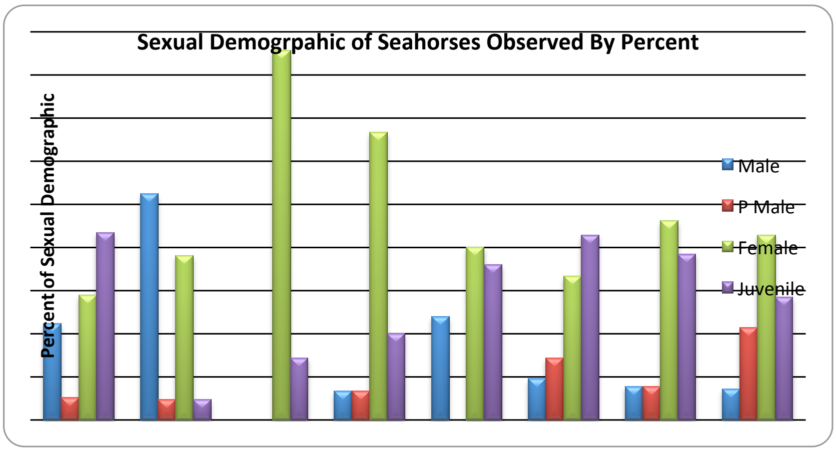
Figure 1: Averaged observed seahorses per survey over time

During the Nov/Dec 2011 surveys there was an average of 2.4 seahorses over 32 surveys observed. When compared to November 2012 there is a decline of 79% in seahorses seen per dive in just one year. It is all the more startling when you look at the total number of seahorses found. In Nov/Dec 2011 there were 76 seahorses recorded and just one year later that number is down to 15 when surveying the same site. The total number of seahorses recorded in December 2012 and January 2013 is 25 and 21 seahorses correspondingly, showing a marked increase compared to Oct. and Nov., and returning to the levels seen in Aug/Sept 2012. In February and March the numbers again drop to 13 and 14 seahorses becoming even with the numbers for November 2012. While still far lower than the numbers seen in Nov/Dec 2011 the figures for February and March are still twice that of the lowest seen in Oct. 2012. The possible cause for the drop in observed seahorses in Oct. 2012 is addressed in more detail subsequently.

As can be seen in Graph 8 and 9 on the following page. Every single sexual demographic is consistently lower than it was in Nov. 2011, with no demographic ever exceeding 10 individuals. Males and females observed have taken a noticeable drop in numbers, but the most troublesome figures however, are the ones associated with reproduction. In one year the number of pregnant males recorded has been reduced to a quarter of its former value, and where 33 juvenile were seem in Nov. 2011, in Nov. 2012 only 3 were observed.



Graph 8: Seahorses observed by sexual demographic

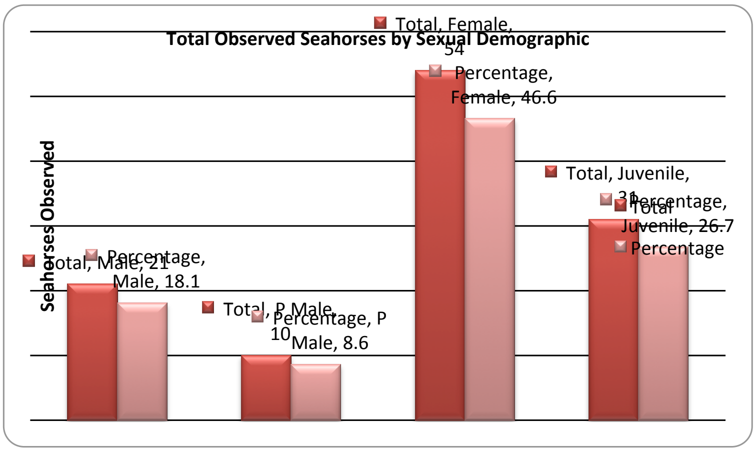


Graph 9: Sexual demographic of seahorses by percent

In Dec. 2012 there is an increase in the juveniles identified at 9 individuals, which is considerably more than the rest of 2012 combined at 5. In the first quarter of 2013 18 juveniles are recorded which is an increase of 29% over all 2012 surveys. The number of reported pregnant males while very low in 2012 is showing signs of increase in 2013 with 7

PM recorded. This includes 3 pregnant males observed in Jan. as well as March, just one less than the maximum of 4 pregnant males recorded in any given month, reported in Dec. 2011. This is also a 133% increase over 2012 in pregnant males observed.

Since Oct. 2012 the seahorse numbers increased during Nov. and Dec. In Jan. the numbers remain comparatively high, and then in Feb. and March the drop back to the Nov. 2012 levels. During these surveys the ratios of sexual demographics remain relatively constant. One thing that remains unclear is the consistent finding of very low numbers of males in comparison to females as seen in Graph 10. The reason for this is unknown. However it may be possible that there is some outside factor that is inhibiting the development of males in the wild, continued research will be able to determine the source of this anomaly.



Graph 10: Total observed seahorses by sexual demographic from Aug. 2012 to March 2013

Fewer juveniles mean a smaller population for the next generation of males, pregnant males, and female seahorses. The low juvenile count in Aug/Sept 2012 may speak for the dip in population in following months. Conversely, Dec. and Jan.'s reemerging numbers in juveniles may be a hint at growth in population in future months. Unfortunately, without accurate information on growth rates in the wild it is impossible to say with any certainty when these numbers will be realized.

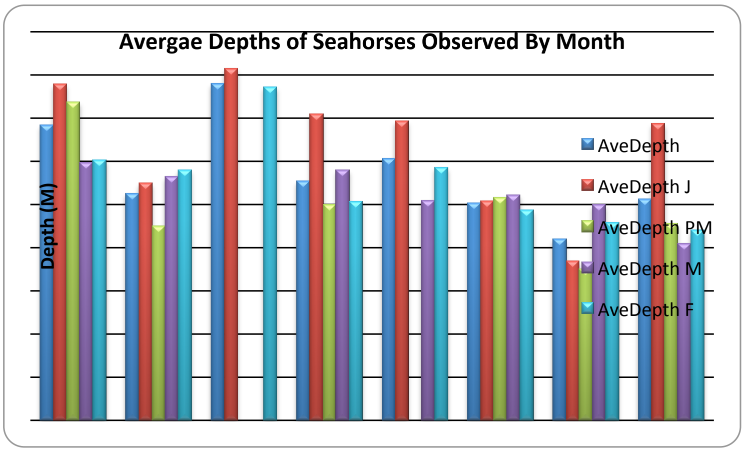
It is possible that the increase in juveniles during Dec. and Jan. is the result of some form of mating season or breeding cycle. However this theory is complicated by the data, which shows that pregnant males are increasing in Jan and Mar. Through captive observation it is known that H. spinosissimus can have a gestation period as short as 14 days. If H. spinosissimus is breeding year round it would explain the fluctuation in numbers from month to month. And any attempt to identify a breeding cycle would be futile. This then begs the question, if the local population is breeding year round; why where there are only 3 pregnant male identified in Aug-Dec 2012? Further research will confirm or deny these possibilities and give insight into the breading habits and behaviors of the local population, particularly when analysis of water temperature and other seasonal variations such as current flow, direction and possible lunar cycles are considered when the data set is larger.

In the rest of this discussion the attempt is made to make some correlation out of the numbers and data that we have collected through March 2013. And indeed there do appear to be patterns emerging that will hopefully aid our future research and conservation efforts. However, interpreting these patterns is hindered by a relatively small data set, and low numbers of seahorses identified. It was stated in the end of year Dec. 2012 report that the low number of seahorse identified associated with reproduction was a concern for the recovery of the population. In the first quarter of 2013 juvenile numbers are up compared to 2012 and most importantly pregnant male numbers are showing an increase.

One possible reason for the drop in seahorses recorded in October 2012, compared with Aug/Sept, is that the random selected points for that month gave a disproportionate number of deep survey sites, as seen in Table 2. The average depth for the survey points for this period was 19.4m with only 5 surveys done below 15m. November's average survey depth was 11.4m, with a more varied depth distribution. Dec., Jan., and Mar.'s average dive depth continued to maintain a varied distribution with their depths remaining close to 12m. This is in large part due to the modified methodology for point selection after Oct. 2012 depths were so outside of the average. Under the new grid selection system the depths and distribution remain more dispersed. Feb. is the exception with the average depth dipping just below 11m. October's average dive depth of 19.4m noticeably deviates from the rest of the other average dive depths and falls out of the range of the seahorse's preferred depth range as understood at this time, while all other months averaged dive depths fall within it. Preferred depth ranges are discussed subsequently.

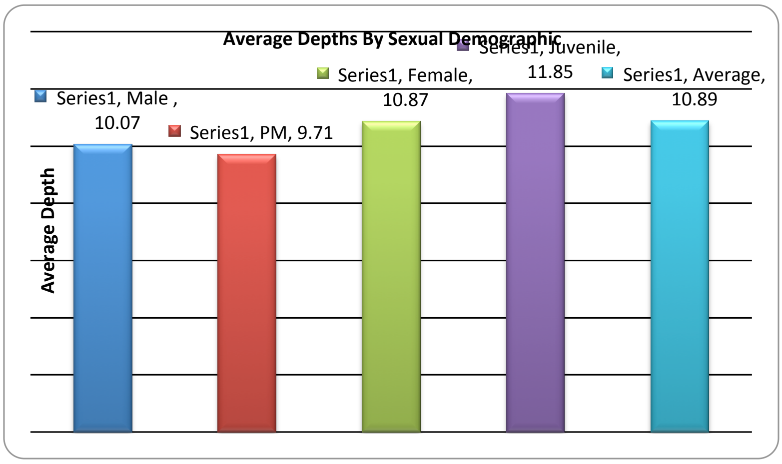
MONTH	AVERAGE DIVE DEPTH
AUG/SEPT 2012	10.396 m
OCT 2012	19.414 m
NOV 2012	11.44 m
DEC 2012	12.121 m
JAN 2013	12.363 m
FEB 2013	10.859 m
MAR 2013	12.197 m

Table 2: Average depth of survey dives by month



Graph 11: Average depth of seahorses observed through March 2013

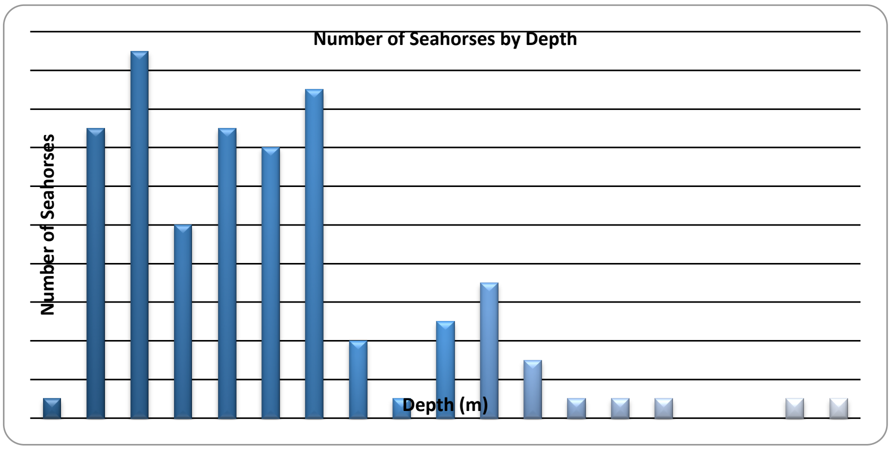
Graph 11 shows the average depth of seahorses by demographic per month. However, it is important to note that a few monthly averages are unfortunately represented by single sightings of seahorses rather than a calculated average of two or more. These single sighting include single males and pregnant males in Nov. 2012 and Feb. 2013, as well as the single juvenile found 16.3m in October, which could be an outlier.



Graph 12: Average depths of sexual demographics from Aug. 2012 – March 2013

Graph 12 shows the average depths of all seahorses by sexual demographic. The average depths for male, female, and pregnant males are all within ~1m of each other. Juveniles are averaging slightly deeper than the other demographics at 11.85m. It is important to note that with the gradually sloping bottom at our study site the difference from 10 to 12m can be several 100 m in places. Of all seahorses recorded, those found between 7-12m account for 77.6% of the total. If 7-12m is indeed a preferred range, it is evident that the average dives in October fall outside of this. August 2012-March 2013 rest comfortably within this range and would thus have higher counts in seahorses.

Note: From observational data large males are often found at depths up to 50m in Cambodia



Graph 13: Number of recorded seahorses per depth from Aug. 2012 through March 2013

As shown in Graph 13, seahorses have been observed at depths of 6 to 24 m. Within this range, there appears to be two groups that the seahorses were found: 7 to 12 m and again at 15 to 17 m. After 18 m, seahorses are solitary if observed at all.

The first group of seahorses from 7 to 13 m holds the majority – 81% – of seahorse data. It is arguable that this may be a favorable depth for the seahorses. The second group of seahorses from 15 to 17 m holds 12.9% of the data. Cumulatively, the two groups account for 94% of all seahorses observed.

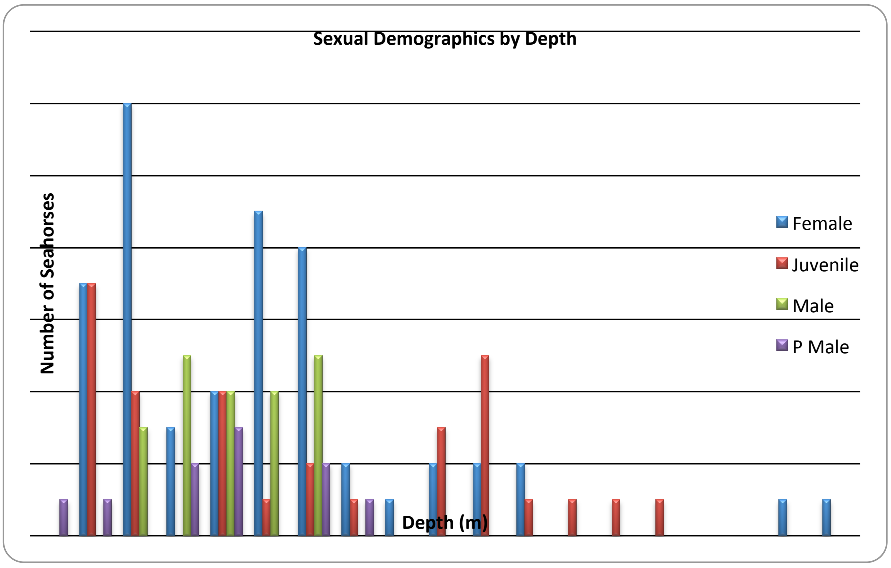
There is a dip at 14 m, which separate the two groups of data. Otherwise, the two groups may have been able to form one larger group. Why are there two groups? Are there two favorable depth ranges for seahorses? If so, what is happening in between the two ranges? Could it possibly be an area passed through during some form of migration due to seasonal variations or breeding habits? It is also possible that currents play a role in the separation observed between these two groups.

In the Aug/Sept report it was hypothesized that pregnant males migrate to deeper water to give birth due to the high proportion of pregnant males and juveniles found at an average depth of 15m, compared with adult males and females found at an average depth of ~11.5m. In October 2012 1 juvenile was found at 16.3 meters and no pregnant males were recorded. In November 2012, 3 juveniles were recorded with an average depth of 14.2 meters, 1 pregnant male was found at 10 meters, with the average for males at 11.6 and females at 10.15. Again in December the average depth of juveniles was 13.87 while the average depth of males and females was 10.18 and 11.72 respectively. This again was consistent with the previous hypothesis as the surveys are finding the bulk of our juveniles in deeper water. In the first quarter of 2013 this hypothesis is weakened as is seen in Graph 4. The averages for all sexual demographics for Jan/Feb/Mar are at 10m or less with the exception of juveniles in March that has an average of 13.75. As seen in Graph 12 this pulls the averages for all demographics much closer together. In Table 3 it is also obvious that this quarters data suggest a far more homogenous population in terms of depth than was previously thought.

Average Depth	**9.67m**
Juvenile	**10.44m**
Pregnant Male	**8.82m**
Male	**9.55m**
Female	**9.24m**

Table 3: Average seahorse depths for Jan, Feb, Mar 2013

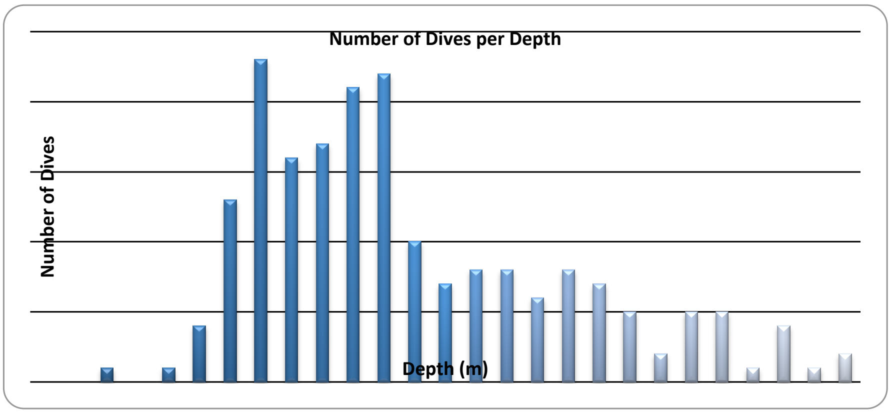
It again must be noted that while the difference in depth between demographics does not appear to be very large, the topography of our study site is a very gradual sloping bottom. The difference in depth a few meters can equate to several hundred meters of distance. All seahorses observed by sexual demographic per depth can be seen in Graph 14.



Graph 14: Sexual demographics of all seahorses by depth Aug 2012-March 2013

It is clear from Graph 14 that females and juveniles are spread out and observed throughout our survey area. Females range from 7m to 24m, and juveniles from 7m to 20m. The interesting thing is that the males, both pregnant and not, are found exclusively at 7-13m within our survey site. Why are the males staying to this particular depth range while the females appear to be far more dispersed? A larger sample size and increased dives at these deeper depths may help answer this question.

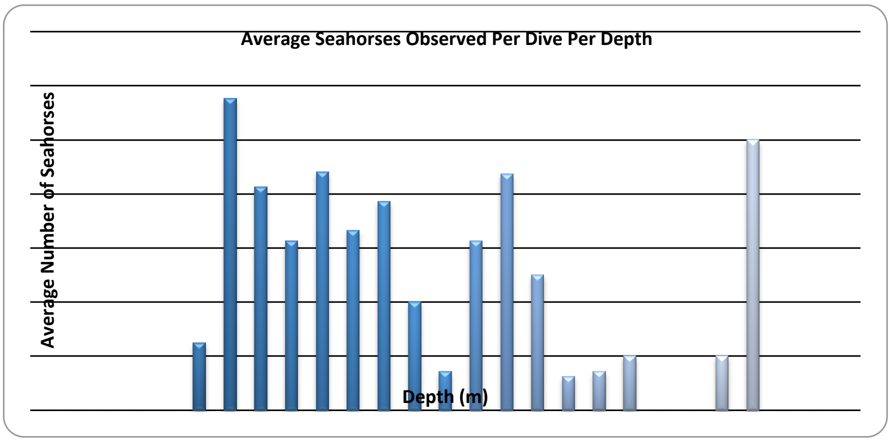
Since Aug. 2012 there have been 197 survey dives made at a range of 3 to 27 m. It is true that the number of dives per depth is favored in the range of 7 to 13m. As can be seen in Graph 15, the number of dives to depths greater than 13m are all lower than 10 and at depths deeper than 19m are all equal or lower than 5m. An increase in the number of dives at these depths may reveal males and PM at deeper depths.



Graph 15: Total number of dives per depth Aug 2012-Mar 2013

While low numbers of dives made at a given depth may explain the absence of certain demographics witnessed regardless of total number of seahorses. Low numbers of dives at a given depth can confound the total number of seahorses recorded. If few dives were made at 13 and 14 m, then it only follows that fewer seahorses would be observed. Similarly, observing more seahorses at a specific depth, such as 8 m, could simply be the result of more frequent dives at that depth. To understand the relationship further, taking the average number of seahorses observed at each depth will help eliminate the variables mentioned above and can be seen in Graph 16.

Note: Adapting the methodology to give a more equal number of dives per depth may help verify these findings. It is also important to note that the topography of the study site is primarily 7-15m deep. So even when the dives are spread out evenly over our entire study site the proportion of dives in this range will be higher than at deeper depths.

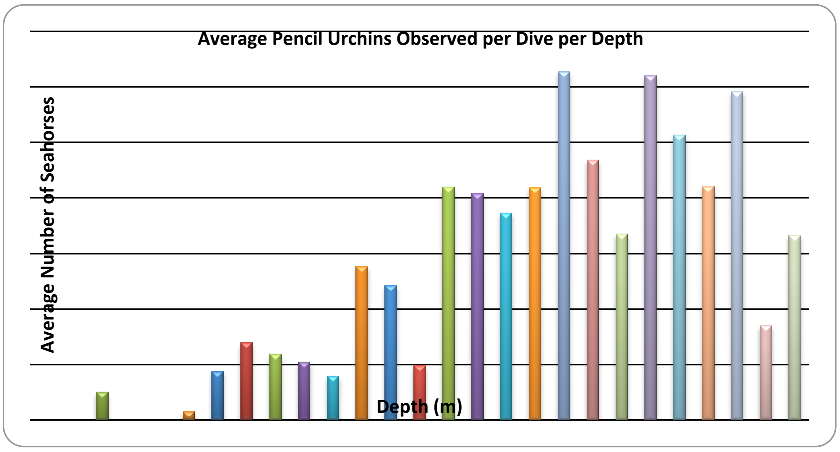


Graph 16: Average seahorse observed per depth per dive Aug. 2012 through Mar 2013

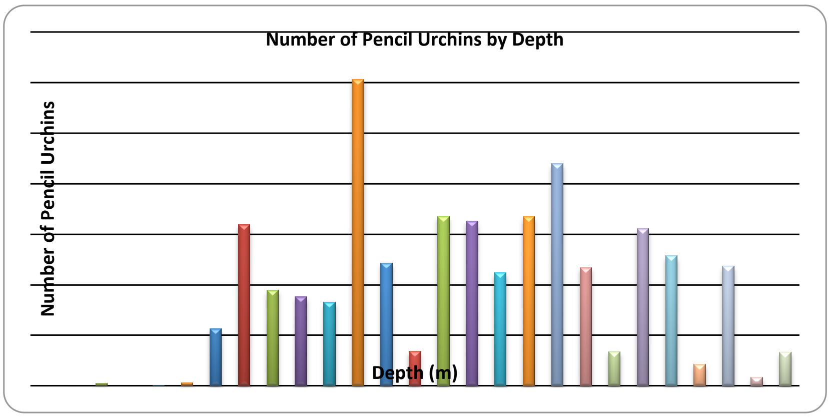
The shape of this graph is smoother and with fewer outlying points. The 1.0 average seahorse per dive at 24 m is because only one dive was made at the depth and a seahorse was observed, which leads to an uninformative average of 1 seahorse. Future research will clarify whether singular anomalies are simply chance or the beginnings of a trend. And while taking the average seahorses per dive per depth helps to eliminate some variables larger data sets and increased numbers are needed to fully understand the relationship.

Nevertheless, both Graph 13 and Graph 16 indicate 7 -13m and 15-17m as a favorable range regardless of the number of dives at any specific depth. This leads to the question of why seahorses prefer that depth range. And why there is this gap at 14m where there have been 7 dives at that depth. Exploring further variable such as temperature, migratory patterns, and holdfasts is the next step.

Previous reports examined the holdfast and habitat preference of seahorses. This quarter the total percent of seahorses found on pencil urchins has dropped to 80% from 85% at the end of Dec. 2012. Investigating a possible relationship between depth of pencil urchins and depth of seahorses could help explain seahorse distribution.



Graph 17: Average pencil urchins observed per depth per dive Aug. 2012 through Mar. 2013



Graph 18: Total number of pencil urchins by depth Aug. 2012 – Mar. 2012

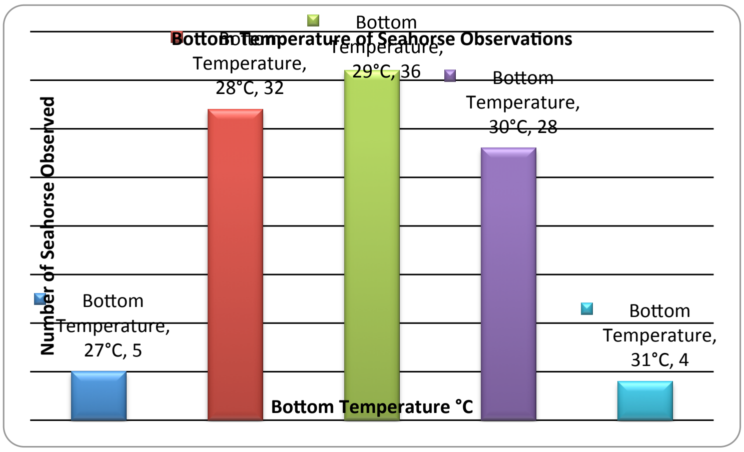
Graphs 17 and 18 give some interesting information. The average urchins per dive is weighted heavily on the depths deeper than 14m as seen in Graph 17. This is to be expected as we see high numbers of urchins and also have low numbers of dives at these deeper depths. When looking at Graph 18 the number of urchins is more evenly distributed. Regardless, it seems that the number of urchins found and the number of seahorses observed is inverted in regards to depth, with the exception of an overlap at 15-17m.

Note: Higher densities of pencil urchins and shell cover make the sighting and identification of seahorses more difficult due to their excellent ability to camouflage and blend into their surroundings than in less dense habitats.

Consequently, at this time it does not seem possible to positively correlate pencil urchins and seahorses through depth. Though seahorses are partial towards pencil urchins, pencil urchins are not an explanation for the favorable depths of seahorses.

Though the seahorse and pencil urchin have a significant relationship, it is not yet possible to determine any correlation or causal relationship between the two. This is because, while seahorses suggest the presence of pencil urchins, the opposite is not true.

In Graph 19 we can see the number of seahorses observed by the bottom temperature they were found in. The most prevalent temperature is 29C with 36 seahorses. 28C and 30C are also well distributed with 32 and 28 respectively.



Graph 19: Average bottom temperature of observed seahorses Aug 2012-Mar 2013

The average seahorses recorded per dive per temperature paints a different picture. There were .73 seahorses at 28C, .52 at 29C, and .64 at 30C. 27C and 31C were both averaged at .5 seahorses per dive. This means that the average seahorse per dive is the same for 27C, 29C, and 31C, while the two highest totals are sandwiched between them. Deeper depths are usually associated with lower temperatures and shallower depths higher temperatures. This makes for a very unusual distribution that is worthy of further study.

One of the most striking observations of the survey data to date is the comparison between November 2011 and 2012 where a dramatic drop off in the number of juveniles has been identified. There has been a significant drop in pregnant males as well, but with so few being observed during surveys this drop could be possibly explained through random survey points just missing them, as sightings are fairly rare in general. The drop in juveniles however, cannot be explained away. Since the survey in 2011 when 33 juveniles were observed, in the 4 months surveys were carried out in 2012 there were just 14 juveniles recorded. This is a very dramatic drop off that has implications beyond the general decline of the local population. It suggests that the local populations ability to recover and continue to breed though their natural behavior, which is already undetermined and possibly quite untraditional due to the very unique habitat they are in, is being impeded. In the first quarter of 2013 the surveys are showing indications that this negative trend in reproduction is shifting, if only slightly. The numbers being recorded are still far lower than in 2011, but are increasing in large proportion to the surveys in 2012. Continued monitoring of the population will make clear the long-term magnitude of this potential shift. If the seahorse's natural ability to reproduce has been so degraded that they can no longer sustain a population, then further efforts to protect the area may be futile in regards to seahorse recovery. It may be necessary to implement a captive breeding and reintroduction program to augment the natural population and give them a chance to recover to their previous densities.

Perhaps the most intriguing question raised during the surveys so far is the irregularity occurring at 14m. There is a noticeable drop in seahorse numbers, average seahorses per dive, pencil urchin numbers, and average pencil urchins per dive. There have been 7 dives at this depth, and while that is not the highest total by any means, it is certainly not the lowest. It is made all the more unusual by the larger numbers in all of these categories that immediately surround this depth. It will be interesting to see what the explanation is for this anomaly.

Conclusion

This is the early stages of a long term survey project and as such the conclusions should be considered to be in there early hypothesis, as the project progresses, results will generate differing answers to those contained within this report.

Each quarterly report will build into yearly reports, which will be collated every 5 years to show a continuous pattern in seahorse species, population assessment, and further data on the condition of the seabed and holdfast preferences.

As can be seen in this report, there is a direct correlation between the condition of the seabed and the number of seahorse species, this has been concluded by looking at the pre trawled data with the post trawling data and looking at the pattern of results post trawling. The majority of pre-trawled data is strictly observational and based around species distribution, diversity, and population densities with little direct data on urchin densities and shell cover before illegal trawling did large-scale damage. However, the impact that trawling has had on density and distribution of seahorses is undeniable, and it reasonable to assume that the removal of holdfasts and the drastic altering of the habit was a driving force behind seahorse declines.

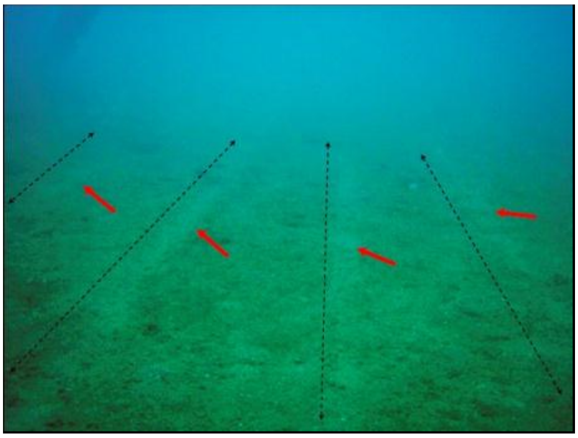


Photo 4: Visible trawling marks on devastated substrate, Koh Rong Somleom 2008

Another possible reason for the drastic reduction in species diversity is the seahorse focused illegal fishing that occurred in 2008-2009 when species numbers and diversity were radically reduced. Further study is needed to determine if the slow population recovery and diversity, after the area was protected, is a result of low density impacting mating behavior, low survivorship, long gestation, slow growth to sexual maturity, etc. All these factors can be possible reasons for the different rates of recovery by different species.

Photo 5: Counting seahorses/other *Syngnathoidae* after confiscation from illegal fishing vessel Oct. 2008

By comparing the type of complete or incomplete holdfasts on the study site it shows clearly that *Hippocampus spinosissimus* is best suited to adapt to this broken and fragmented habitat and even this hardy species is reducing in numbers.

To reestablish traditional seahorse numbers and diversity of species it is necessary to firstly stop the trawling and secondly allow the habitat to reestablish or take mitigating actions to restore it to its former status.

There is a great deal more to learn on this, such as the correlation of depth and time of year to breeding, and as to whether the seahorse species on this site pair bond as is considered normal for seahorses, or if they indeed have adapted as *Hippocampus spinosissimus* appears to have done to the fragmented habitat.

Only by continuing this survey into the future will these questions reach some form of conclusion.