



A COMPILATION OF REPORTED FISH KILLS IN THE MALDIVES

S. Naeem & S. A. Sattar

Marine Research Centre
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(July – December, 2007)

Shafiya Naeem and Shahaama A. Sattar

Marine Research Centre, Male' Maldives

Executive Summary

This preliminary report of a compilation of reported fish kills in the Maldives (July to December 2007) was prepared upon the request of the Presidents' Office. The report details a time-line of fish-kill incident reports, current status, actions taken and results obtained to date. Such incidents of fish-kill of the scale currently being observed in the Maldives could occur due to a variety of reasons: changes in physical and chemical conditions, biological phenomena such as algal blooms, infections of bacterial or viral origin and/or a combination of these and other factors.

Marine Research Centre continues to receive reports of fish-kill incidents, which began in July this year. Although the first report received from Meemu Atoll maybe suggestive of an algal bloom, subsequent events have not been linked to any such cause. Furthermore, all but the first two incidents of fish-kill (which might be unrelated to the subsequent events), report mass death of triggerfish, predominantly the red-tooth trigger fish, *Odonus niger* (Vaalaa Rondu). As MRC did not have the capacity to carry out relevant tests and analyses to identify the cause of these mass mortalities, suggestions to seek foreign expertise were put forward to relevant officials at the ministry. These suggestions were turned down on the basis that the costs associated with such a mission was, at the time, not a financially viable option.

Oxygen level measurements of seawater in areas of fish-kill have shown normal readings, thus eliminating oxygen depletion as a cause for this incident. Tests by the National Health Laboratory on fish samples of *O. niger* showed the presence of *Staphylococcus* sp. in the spleen of these samples. This has not been made public to date, as these results are inconclusive of anything for the time-being. As MRC and Maldives lack the capacity

to deal with such large scale incidents of fish-kill, we have to resort to collaboration with institutes and individual parties from overseas. As such we are awaiting results from fish samples which have been sent to laboratories in India and Denmark.

Assoc. Prof. Jacob Larsen of the IOC Science and Communication Centre in Denmark will be arriving in the Maldives on the 9th of December. He is a specialist in harmful microalgae and was invited to the Maldives by Banyan Tree Maldives Vabbinfaru, in collaboration with MRC. Furthermore, MRC is also in collaboration via email with Professor Wolfgang K. Vogelbein of the Virginia Institute of Marine Science who is a specialist in microbiology. We have collected and preserved organs of fish samples as per his instructions. It is hoped that we will be able to secure funds for his visit to the Maldives.

MRC is unable to make any definite conclusions from the results at present. A more comprehensive report of this fish-kill incident will be published once we have explored all possible theories and completed all investigations.

Introduction

Fish mortalities of variable scales have been reported to have occurred in different regions of the Maldives from July 2007. Previous records of similar events in the Maldives are available (Hussein Zahir, pers. comm), and the reasons for the current event are still unknown.

The localized die-off of marine fish also known as “fish kill” is a common occurrence in coastal waters and estuaries worldwide. They may result from a wide variety of causes including changes in physical conditions (storms, temperature fluctuation, changes in salinity), chemical conditions (toxic contaminants, oxygen depletion etc), as well as biological phenomena (algal blooms, biotoxins, spread of disease causing agents, etc), or a combination of such factors (Austin, 1999; Bondad-Reantaso, et al., 2001; Claereboudt, et al., 2001; Hallegraeff, et al., 2003; Reid, et al., 2002; vanDuijn Jnr., 1973).

Phytoplankton (or algal) blooms are reported to be a very common cause of fish kills around the globe. Controlled populations of several groups of potentially harmful algae (usually belonging to the dinoflagellates) exist in the marine environment. When conditions become favourable (nutrient enrichment of the waters, changes in physical conditions of the surrounding waters, etc) the microalgae (usually also associated with the secretion of toxins) populations burst causing mass mortalities of fish (Glasgow, et al., 2000; Hallegraeff, et al., 1995). These toxins are not necessarily always associated with fish kills, but rather the planktivores that feed on these dinoflagellates accumulate the toxins, which in turn affects higher predators (including human beings) that feeds on the toxin-accumulated fish.

Associated with phytoplankton blooms is an increase in primary productivity, and in turn, an increase in the oxygen content of the surrounding waters in the presence of sunlight (Falkowski, et al., 1997; Hallegraeff, et al., 1995). In the absence of sufficient light however, the phytoplankton begin to respire, consuming a considerable amount of oxygen dissolved in the water. This could result in the depletion of dissolved oxygen content to the extent of causing mortalities of living organisms in the water due to oxygen starvation. Oxygen depletion in bottom waters may also result from nutrient enrichment of the waters due to increased sedimentation of organic matter, leading to higher

decomposition rates. This increased biological activity, in combination with a limited replenishment of oxygen by mixing, can lead to the exhaustion of oxygen content near the bottom. During upwelling events the oxygen-depleted bottom waters are brought to the surface making the surface waters limited in dissolved oxygen content (Claereboudt, et al., 2001; Kruse, et al., 1995)

In addition to the potentially harmful phytoplankton, fish are constantly exposed to several other disease-causing agents (bacteria, viruses, parasites, fungi, etc). Most of these organisms normally occur in low numbers unless the conditions become favourable (for example an increase in the temperature of the surrounding waters) for their growth. Although their occurrence is rare, outbursts of disease-causing microorganisms do occur, causing fish kills at epidemic levels (Austin, 1999; Bondad-Reantaso, et al., 2001; vanDuijn Jr., 1973). Colonisation of internal and external surfaces of fish by various microorganisms occurs at different stages of their lives. Fish diseases, or in the worst case, mortalities can occur if pathogenic microorganisms colonise the surfaces. The extent of mortality is related to the extent of pathogenicity of the colonizing microorganism.

Another important contributor to the mass mortality of fish could be the limited availability of food, either due to a population explosion of the fish stock (which usually indicates a considerable amount of competition for the limited supply of food, and thus results in the stronger fish thriving and the weaker ones dying from starvation), or as a result of a reduction in the population of the food organisms. Changes in the condition of the existing environment can contribute to both (or either of) the changes.

Widespread fish mortalities of massive scale have been occurring in different regions of the Maldives. The species compositions of dead fish and the characteristics of the surrounding water suggests that most of the reported events were closely related to each other, however there were occasional reports of fish kills that were probably unrelated. All the suggestively related fish kills consisted predominantly of the red-toothed triggerfish, *Odonus niger* (Vaalán Rondu), which comprised approximately 95% of the fish that were found dead. There are unconfirmed reports of similar fish kills, again *O. niger* being the major group affected in the Arabian Sea in November 2007. Another similar event was reported in Reunion Island in 2000 and 2001, where groups of triggerfish were found dead. Test results from the latter event reported the presence of *Streptococcus* sp.,

a species of bacteria known to be associated with fish kills, in the spleens of the dead fish.

This paper describes the series of fish kill events that were reported since the initial event in July 2007, investigative studies carried out to date and their results, as well as the limitations faced during the investigation.

Time line of fish kills

Possible algal bloom related fish death – M. Raiymandhoo

Reports of fish deaths were received from M. Raiymandhoo on the 8th of July 2007. Associated with these fish deaths, there was also a presence of a red substance floating in the lagoon M. Raiymandhoo. Water samples taken by a team from the Marine Research Centre (MRC) did not show the presence of any microorganisms which could have been due to the time lag between when the sample was taken and observed after having been brought back to the island. The dead fish composition predominantly consisted of different species surgeonfish (*kaalhu*) and parrotfish (*Landaa*). These were found floating amongst the red substance and were in the initial stages of the decaying process (Photo 1). Seawater samples taken by a team from the Environment Research Centre (ERC) on the 9th of July showed the presence of a microorganism suggestive of *Alexandrium* species (ERC, 2007).



Photo 1. Dead fish floating amongst the red substance found in M. Raiymandhoo in July 2007
(Photo by Adam Rasheed, MRC).

deaths of varying fish species, the other atolls report the death of a high number of *O. niger*. Additional species observed mainly belong to the same family (i.e. balistidae), although some islands report the occasional deaths of unicorn fish, snappers and groupers.

MRC has been following the trend of these fish deaths. Since we did not have the capability to deal with such a large scale event, MRC had consulted the Intergovernmental Ocean Commission (IOC), from the first instance when we received reports of mass deaths of *O. niger* in Ari Atoll. We were advised by IOC that they could send a consultant on a fact-finding mission for a certain fee, and this was communicated to relevant officials at the Ministry of Fisheries, Agriculture and Marine Resources (MoFAMR) who at that time decided that the cost of bringing a consultant could not be afforded. As we did not have the expertise to carry out the tests required at the time no field studies were carried out and no samples were collected. However, a phone survey was conducted by MRC in September, to all islands and resorts in North and South Ari Atoll. A similar survey has been conducted to all other atolls that have since reported fish deaths.

Methodology

In November 2007, teams from MRC visited Vaavu, Baa and Dhaalu atolls as well as Sun Island Resort and Spa and White Sands Resort and Spa.

Dissolved Oxygen levels in the seawater

Water samples at different depths (0, 10, 20, 30 and 40 m) were collected by diving on these trips. Samples were collected from:

- Rakeedhoo channel
- Bottom reef of B. Dharavandhoo
- Nearby V. Felidhoo B. Undoodhoo and B. Milaidhoo, Sun Island Resort and Spa and White Sands Resort and Spa

Samples were collected in small bottles, ensuring that no air bubbles were trapped inside the bottle. These samples were tested for oxygen levels using a HI 9142 portable waterproof Dissolved Oxygen meter as soon as they were brought on board. It should be noted here that the samples collected in Baa atoll from Dharavandhoo and Undoodhoo were measured after having being brought back to the island (i.e a time lag of 1 to 2 hours between collection and testing).

Samples have also been collected and fixed with Lugol's solution. These will be tested for the presence of harmful algae by a visiting consultant who will arrive in the Maldives on the 9th of December.

Semi-quantitative surveys of fish counts

Semi-quantitative surveys were also carried out in the following islands to identify the species composition and abundance of dead fish on the beaches: V. Bodumohoraa, V. Rakeedhoo, V. Anbara, V. Felidhoo, V. Keyodhoo, B. Dharavandhoo, B. Eydhafushi, B. Undoodhoo, B. DHonfanu, B. Maalhos, B. Kihaadhoo, B. Kamadhoo, B. Milaidhoo, Dh. Kudahuvadhoo, Dh. Vaani, Sun Island Resort and Spa and White Sands Resort and Spa.

Samples of freshly dead/dying fish were collected from Vaavu Atoll and Dhaalu Atoll. These samples were frozen upon collection. 3 samples of *O. niger* and 2 samples of

unidentified fish species were collected from Vaavu atoll and sent to the National Health Laboratory (NHL) to test for the present of ciguatoxins and *Staphylococcus* sp. Samples of *O. niger* collected from Dhaalu Atoll have been sent by ERC to a laboratory in India (unsure of what these are being tested for) while *O. niger* samples from Vaavu atoll have been sent to the IOC Science and Communication Centre on Harmful Algae of UNESCO in Copenhagen, Denmark to test for the presence of harmful algal blooms.

Results and Discussion

Dissolved Oxygen levels in the seawater

Oxygen levels reported from the dives are shown in tables 1 to 3 below:

Table 1-Dissolved Oxygen readings in Vaavu Atoll

Island	Rakeedhoo	Rakeedhoo	Rakeedhoo	Felidhoo
Date	21-Nov-07	22-Nov-07	22-Nov-07	23-Nov-07
Time of dive	1500 hrs	0930 hrs	1515 hrs	1500 hrs
Depth	Oxygen readings (mg/l)			
0				
10	7.3	7.5	7.1	7.0
20	8.1	7.7	7.2	7.4
30	7.2	7.1	7.4	7.6
40	7.1	7.3	8.7	8.4

Table 2-Dissolved Oxygen readings in South Ari Atoll

Island	Sun Island Resort and Spa	Sun Island Resort and Spa	White Sands Resort and Spa
Date	30-Nov-07	30-Nov-07	1-Dec-07
Time of dive	0630 hrs	1315 hrs	0600 hrs
Depth	Oxygen readings (mg/l)		
0	5.2		5.5
10	4.7	7.5	5.5
20	4.7	7.6	5.7
30	4.2	7.5	5.5
40	4.6		

Table 3 Dissolved Oxygen readings in Baa Atoll

Island	Dharavandhoo	Undoodhoo	Milaidhoo
Date	22-Nov-07	23-Nov-07	24-Nov-07
Time of dive	1610 hrs	1530 hrs	1530 hrs
Depth	Oxygen readings (mg/l)		
0	6.5	6.7	7.8
10	6.5	6.3	7.0
20	6.4	6.4	7.1
30	6.35	6.2	7.0
40		5.8	7.0

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In normal circumstances, an average of 6 mg of dissolved oxygen can be found per litre of seawater (Garrison, 2005). Therefore the above results which on average range between 6 to 7 mg/l of dissolved oxygen can be considered as normal levels, thus nullifying our hypothesis of low oxygen levels being a cause of this fish kill incident. Although the dawn dives showed lower oxygen levels, this is expected for that time of the day and might not necessarily be linked to these fish deaths.

Semi-quantitative surveys of fish counts

Results from our semi-quantitative survey of the species composition and abundance of the dead species are shown in table 4, 5 and 6 below:

Table 4 Fish counts from Vaavu Atoll

Island Date	Bodumohora 21-Nov-07	Rakeedhoo 22-Nov-07	Anbara 22-Nov-07	Keyodhoo 23-Nov-07	Felidhoo 23-Nov-07	Bodumohora 25-Nov-07	Total	%
TRIGGERFISHES								
<i>Odonus niger</i>	481	393	240	2	33	542	1691	95.2
<i>Balistapus undulatus</i>	23	4	3	0	1	4	35	2.0
<i>Canthidermis maculatus</i>	1	0	0	0	0	0	1	0.1
<i>Melichthys indicus</i>	1	0	0	0	1	0	2	0.1
<i>Pseudobalistes flavimarginatus</i>	1	0	0	0	0	0	1	0.1
<i>Pseudobalistes fuscus</i>	0	1	0	0	0	0	1	0.1
<i>Rhineacanthus aculeatus</i>	1	1	0	0	0	1	3	0.2
<i>Sufflamen bursa</i>	3	1	0	0	0	0	4	0.2
<i>Sufflamen chrysopterus</i>	3	3	0	0	0	3	9	0.5
<i>Sufflamen fraenatus</i>	2	1	1	0	0	0	4	0.2
<i>Subtotal</i>	<i>516</i>	<i>404</i>	<i>244</i>	<i>2</i>	<i>35</i>	<i>550</i>	<i>1751</i>	<i>98.5</i>
OTHER FISHES								
<i>Anyperodon leucogrammicus</i>	0	1	0	0	0	0	1	0.1
<i>Cephalopholis miniata</i>	0	1	0	0	0	0	1	0.1
<i>Epinephelus ongus</i>	1	0	0	0	0	0	1	0.1
<i>Variola louti</i>	0	1	0	0	1	0	2	0.1
Unidentified grouper	0	0	2	0	0	0	2	0.1
<i>Lutjanus gibbus</i>	0	1	0	0	0	0	1	0.1
<i>Lutjanus kasmira</i>	1	0	1	0	0	0	2	0.1
<i>Caesionid</i>	0	1	0	0	0	0	1	0.1
<i>Pterocausio pisang</i>	0	1	0	0	0	0	1	0.1
<i>Gerres</i>	0	0	1	0	0	0	1	0.1
<i>Mulloidichthys vanicolensis</i>	0	1	0	0	0	0	1	0.1
<i>Kyphosus cinerascens</i>	0	0	0	0	0	1	1	0.1
<i>Sphyræna sp.</i>	0	0	0	0	0	1	1	0.1
<i>Chromis ternatensis</i>	0	1	0	0	0	0	1	0.1
<i>Cheilinus fasciatus</i>	1	0	0	0	0	0	1	0.1
<i>Ctenochaetus striatus</i>	1	0	0	0	0	0	1	0.1
<i>Naso brevirostris</i>	1	0	0	0	0	0	1	0.1
<i>Naso vlamingii</i>	0	1	0	0	0	1	2	0.1
<i>Arothron hispidus</i>	0	0	0	0	0	1	1	0.1
<i>Diodon hystrix</i>	0	1	0	0	0	0	1	0.1
Unidentified	0	0	0	0	2	0	2	0.1
<i>Subtotal</i>	<i>5</i>	<i>10</i>	<i>4</i>	<i>0</i>	<i>3</i>	<i>4</i>	<i>26</i>	<i>1.5</i>
TOTAL	521	414	248	2	38	554	1777	100

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Table 5 Fish counts from Baa Atoll

Island	Dharavandhoo	Undoodhoo	Dhonfanu	Maalhos	Kamadhoo	Milaidhoo	Total	%
Date	22-Nov-07	23-Nov-07	24-Nov-07	24-Nov-07	24-Nov-07	24-Nov-07		
TRIGGERFISHES								
<i>Odonus niger</i>	12	0	3	3	193	18	229	90.9
<i>Balistoides conspicillum</i>	1	0	0	0	2	0	3	1.2
<i>Balistoides viridescens</i>	2	0	0	0	0	0	2	0.8
Unid triggerfish	1	0	2	0	1	3	7	2.8
<i>Subtotal</i>	<i>16</i>	<i>0</i>	<i>5</i>	<i>3</i>	<i>196</i>	<i>21</i>	<i>241</i>	<i>95.6</i>
OTHER FISHES								
<i>Naso vlamingii</i>	2	1	0	0	1	1	5	2.0
Unidentified	0	0	1	0	1	0	2	0.8
Unidentified pufferfish	1	0	0	0	0	1	2	0.8
<i>Synanceia verrucosa?</i>	1	0	0	0	0	0	1	0.4
<i>Synanceia unid?</i>	0	0	1	0	0	0	1	0.4
<i>Subtotal</i>	<i>4</i>	<i>1</i>	<i>2</i>	<i>0</i>	<i>2</i>	<i>2</i>	<i>11</i>	<i>4.4</i>
TOTAL	20	1	7	3	198	23	252	100

Table 6 Fish counts from South Ari Atoll

Island	Sun Island resort and Spa	White Sands Resort and Spa (tourist beach area)	White Sands Resort and Spa (construction area)	Subtotal (without construction site info)	% (without construction site info)	Total	Total %
Date	30-Nov-07	1-Dec-07	1-Dec-07				
TRIGGERFISHES							
<i>Odonus niger</i>	176	52	833	228	92.7	1061	97.2
<i>Balistapus undulatus</i>	3	1	3	4	1.6	7	0.6
<i>Balistoides conspicillum</i>	2	0	0	2	0.8	2	0.2
<i>Melichthys indicus</i>	0	0	1	0	0.0	1	0.1
<i>Pseudobalistes fuscus</i>	0	1	0	1	0.4	1	0.1
<i>Rhinecanthus aculeatus</i>	1	0	0	1	0.4	1	0.1
<i>Sufflamen chrysopteris</i>	1	0	0	1	0.4	1	0.1
Unid triggerfish	0	0	8	0	0.0	8	0.7
<i>Subtotal</i>	<i>183</i>	<i>54</i>	<i>845</i>	<i>237</i>	<i>96.3</i>	<i>1082</i>	<i>99.1</i>
OTHER FISHES							
<i>Epinephelus fuscoguttatus</i>	0	1	0	1	0.4	1	0.1
<i>Naso vlamingii</i>	2	0	1	2	0.8	3	0.3
Unidentified	2	0	0	2	0.8	2	0.2
<i>Ostracion meleagris</i>	2	0	1	2	0.8	3	0.3
Unid Cephalopholis	1	0	0	1	0.4	1	0.1
<i>Chromis ternatensis</i>	1	0	0	1	0.4	1	0.1
<i>Subtotal</i>	<i>8</i>	<i>1</i>	<i>2</i>	<i>9</i>	<i>3.7</i>	<i>10</i>	<i>0.9</i>
TOTAL	191	55	847	246	100	1092	100

Similar to the reports from the various islands, the above tables show the following general trends:

- approximately 94% of all dead fish observed are individuals of *O. niger* species
- approximately 97.7% belong to the triggerfish family (Balistidae)
- other species include those of the unicornfish, grouper and parrotfish families.

Majority of the fish washed up on the beaches had been dead for sometime and were decayed. This could be indicative of the fact that they had died in areas far from these islands and had been washed up onto these beaches due to the winds and currents at the

time. However a minor portion of the dead individuals were freshly dead indicating that they had died nearby or within surrounding reef of the island.

Photos 2 to 10 show various species which were observed by MRC teams on their visits to the various islands.



Photo 2. A small sample of the individuals of *O. niger* washed up on the beach of B. Kamadhoo (Photo by Ahmed Najeeb, MRC)



Photo 3. *Balistoides conspicillum* (Photo by Ahmed Najeeb, MRC)

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Photo 4. *Rhinecanthus aculeatus* (Photo by Shahaama Sattar, MRC)



Photo 5. *Balistapus undulatus* (Photo by Shahaama Sattar, MRC)



Photo 6. Unidentified triggerfish (Photo by Ahmed Shan, ERC)

Photos 3 to 6 show different species of triggerfish washed up dead

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Photo 7. Puffer fish which also belongs to the same order as triggerfish (i.e. Tetraodontiformes)
(Photo by Shahaama Sattar, MRC)



Photo 8. *Naso vlamingii* (Photo by Shahaama Sattar, MRC)



Photo 9. Parrotfish (Photo by Ahmed Shan, ERC)

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Photo 10. Grouper (Genus: *Cephalopholis*) (Photo by Shahaama Sattar, MRC)



Photo 11. *Epinephelus fuscoguttatus* (Photo by Shahaama Sattar, MRC)

From the fish samples which were tested by NHL, the 3 samples of *O. niger* tested positive for bacteria belonging to *Staphylococcus* species. This was however absent on the other 2 fish samples of different species. This has not been made public on the basis that currently, it would not be wise to draw any conclusions from these results, as these were all samples from the same area. To draw any conclusions, we need to test healthy *O. niger* samples as well as those found freshly dead from other affected regions. At the time of writing, we are still awaiting results from the fish samples which have been sent to India and Denmark.

Limitations and Preliminary Conclusions

With respect to the tests and analyses conducted so far, no definite conclusions can be drawn on the cause of these incidents without further investigation. It is very likely that the samples sent overseas for pathological testing, as well as those sent abroad to confirm the presence/absence of harmful varieties of algae will not show any meaningful results. What is usually required in these situations is the ability to carry out primary testing procedures at the site where the samples are taken, and since MRC or other institutions that actively took part in the investigation lacked this capacity, it was impossible to carry out *in situ* analyses.

The dissolved oxygen measurements obtained in the current study were all found to be in the “normal” range, which was to be expected considering sampling was carried out in daylight where any phytoplankton in the waters would normally be consuming carbon dioxide dissolved in the water in their photosynthetic processes, and releasing oxygen into the water, enriching the dissolved oxygen content of the water. In the absence of sunlight however, these phytoplankton will begin to respire, drawing the dissolved oxygen from the surrounding waters (Falkowski, et al., 1997; Hallegraeff, et al., 1995; 2003). Minimal dissolved oxygen is observed at dawn where the oxygen maybe depleted enough to affect the other living organisms. We should have obtained oxygen readings at dawn, which could have provided us with different results from what we currently have. Further, the initial samples from Baa Atoll (Dharavandhoo and Undoodhoo) were analysed on arrival at the respective islands, which would have severely affected the results obtained from these sites. Dissolved oxygen analyses should be made *in situ*, unless the oxygen is chemically fixed in the water (using various chemical reagents) so that the air-water interactions have minimal impact on the results obtained.

Not having basic sampling gear prevented us from obtaining plankton samples from water to prove/disprove the presence of harmful algal species in the waters. The mentioned samples are obtained by towing plankton nets in the water for a given distance. Concentrated samples of planktons are obtained this way as the planktons get trapped within the net. The samples obtained this way are then analysed with basic microscopy, using special chambers for counting the number of individuals present. Further analyses are then conducted to identify the species present in the samples. If we had this capacity

prior to this incident, we would have been able to either confirm the presence of harmful algae and relate it to the fish mortalities, or confirm the absence of such organisms, and eliminate this factor from a wide range of possibilities that could cause fish deaths at such a massive scale.

Time between fish death and sampling is an important factor when conducting microbiological investigations, as bacteria involved with decomposition quickly colonises the dead tissues and could very well cause damage to cell structures to the extent that it would be difficult, if not impossible, to make any inference on the cause of death (Bondad-Reantaso, et al., 2001). All samples obtained in the current investigation were frozen prior to being processed for microbiology and chemical analyses. Furthermore, most samples were also obtained as freshly dead fish, whereas microbiological analyses require that the samples be either live or moribund (weak and/or nearly dead). Although the spleens of healthy fish are generally considered sterile (i.e. free of any microorganisms), finding *Staphylococcus* sp. in spleens of the samples from the current study may or may not reveal anything. One reason for not being able to make any conclusions from this finding is the fact that the samples were frozen for as long as 2-3 days prior to being sampled. Further, samples of apparently healthy animals will need to be analysed for the same microorganisms, to obtain a reference on the extent of colonization of the spleen in healthy individuals of triggerfish, and compared to samples from individuals that are suspected to be infected. Not much is known about the microbiology of triggerfish, and it could very well be that normally healthy individuals contain *Staphylococcus* sp. in their spleens. Obtaining healthy samples of triggerfish for the purposes of this study was made difficult as this “epidemic” was very widespread. Thus there was no real control to compare the results from this study.

The fact that majority of fish affected throughout the country were individuals belonging to the triggerfish family possibly indicates an infection (or a disease) of triggerfish. Unfortunately there is not enough evidence to prove or disprove this hypothesis without further testing and analyses. Epidemics causing fish mortalities on a large scale are usually associated with infections of bacterial or viral origin (Austin, 1999; vanDuijn Jnr., 1973). Unpublished research following a similar event that occurred in Reunion Island (France, in SW Indian Ocean) between 2000 and 2001 confirmed the presence of *Streptococcus iniae* in the spleen of all affected individuals. Species of *Streptococcus* are

known to be associated with fish mortalities (Akhlaghi, et al., 1996; Bowser, et al., 1998; Bromage, et al., 1999), and diseases, and could very well have been the cause of the mortality observed in Reunion Island. We were unable to test for the presence of the mentioned bacteria, either through the National Health Laboratory (Maldives Food and Drug Authority) or the laboratory at the Indira Gandhi Memorial Hospital. The case may be entirely different for the Maldives, but this event strongly stresses the importance of being able to test for a wide range of microorganisms with potential to cause harm to aquatic organisms.

As we lack the capacity to properly diagnose the causes of fish mortality or disease, we have to resort to seeking assistance from overseas experts. It was probably too late into the incident that an active effort was made to diagnose the cause of these events. The events (although they may not be related) have been ongoing since July 2007, and it was not until November 2007 that any reasonable investigative tasks were carried out. The fish kill events are reasonably unpredictable, and timing is of critical importance even if foreign experts are willing to provide their assistance.

Current Status

Current fish kill reports are being received from North and South Ari Atoll, Kaafu Atoll and Baa Atoll.

As prior mentioned fish samples from Dhaalu Atoll have been sent to a laboratory in India and samples of *O. niger* have been sent to the IOC Science and Communication Centre on Harmful Algae of UNESCO in Copenhagen, Denmark, we are currently awaiting the results from these tests. It will be interesting to see whether there are any similarities between these results and those which have been obtained from tests done by the National Health Laboratory.

Banyan Tree Resort, in collaboration with MRC will be bringing a visiting consultant, Assoc. Prof. Jacob Larsen of the IOC Science and Communication Centre in Denmark, who will be arriving on the 9th of December. He will visit relevant sites where fish deaths are being reported from at the time and conduct tests on water samples collected to confirm the presence or absence of harmful algal species. Water samples have also been collected from the current survey and are being preserved to be analyzed upon his arrival.

Assoc. Prof. Jacob will also be meeting with relevant officials from the Ministry of Fisheries, Agriculture and Marine Resources, Marine Research Centre, Ministry of Environment, Energy and Water, Environment Research Centre, Ministry of Health, Indira Gandhi Memorial Hospital and the Maldives Food and Drug Authority (National Health Laboratory), to carryout discussions regarding the preparation of a Standard Operations Procedure for future instances of such occurrences.

Additionally, MRC is also in communications via e-mail with various specialists. Amongst them is Professor Wolfgang K. Vogelbein of Virginia Institute of Marine Science, who was kind enough to share with us his expertise. We have collected specimens of dying fish and preserved their liver, spleen, intestine, gonads, heart and kidney as per his instructions. Professor Vogelbein has also offered to visit the Maldives and we are currently looking into securing funds for his visit. However, if this is not possible, samples collected will be sent to him for analysis.

Recommendations to GOM

1. Actions on similar events in the future should be made faster than what was done in the current event
2. As the institution with a responsibility to carry out marine research, MRC should have the professional and technical capacity to deal with similar events, and be equipped with proper equipment and gear to facilitate the relevant actions that need to be taken
3. Better institutional coordination needs to be established and maintained to ensure that relevant measures are taken in a timely manner
4. Given the potential for broader implications of these incidents it is recommended to formulate guidelines on how these incidents are reported and managed in the future. To this end it may be important for the Maldives to develop a national monitoring and management strategy for fish death and HAB incidents.

Capacity building for future incidents

MRC lacks expertise on fish diseases and harmful algal blooms, which made the investigations for the current study very difficult. It is important that MRC develops the capacity to handle similar events in the future, and to this end it is strongly recommended that MRC staff be trained in the fields outlined below (Table 7). Furthermore MRC needs to be better equipped with basic sampling equipment to facilitate, at minimum, primary sample collection. It is therefore also recommended that the laboratory at MRC be equipped with the equipment outlined in table 8, so as to provide MRC with the capacity to carry out proper sampling in similar events in the future. .

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Table 7. Areas of training required and estimated costs

Area of training	Level of training	Possible Institute/Estimated cost	Urgency
Disease Diagnostics/ Aquatic Animal Health	PhD/MSc	Virginia Institute of Marine Sciences/ USA; /US\$ 40,000 (1.5-2 yrs)	***
Environmental and Aquatic Animal Health	BSc	Virginia Institute of Marine Sciences/ USA; /US\$55,000 (3 yrs), or alternative from the Asia Pacific region	***
Histopathology	MSc	Virginia Institute of Marine Sciences/ US\$ 35,000 (1.5-2 yrs), or alternative from the Asia Pacific region	**
Coastal Oceanography	BSc / MSc	University of Bangor / University of Southampton / GBP 33,000	**
Laboratory Technician (microbiology/ histopathology)	Diploma?	South Asia / Australasian region; US\$ 10,000	**
General Environmental Science	Diploma	University of South Pacific, US\$ 25,000	*

Table 8. Equipments required and estimated costs (Prices of those items market with '*' are not readily available. They will be published in the more comprehensive report to be prepared at the end of this investigation)

Equipment Required	Estimated Cost
Plankton nets	US\$ 1,500.00
Microscope with camera	US\$ 8,000.00
Haemocytometers and Sedgewick Rafter Counters	US\$ 500.00
Submersible thermometers for <i>in situ</i> measurements	US\$ 1,200.00
DO/Salinity/pH probes; fixatives + accessories	US\$ 2,000.00
Culture media for basic microbiological analyses	US\$ 1,200.00
Dissecting kits	US\$ 5,00.00
Nansen Bottles for water sampling	US\$ 8,00.00
Chemical reagents for water quality analysis	US\$ 1,300.00
Glassware & Plasticware for microbiology	*
Laminating flow cabinets for microbiological work	*

Fish death in B. Kamadhoo on 24th and 25th November 2007

Apart from the reported fish deaths of the above mentioned species, an isolated unprecedented incident of fish death of mainly blue striped herrings (*Herklotsichthys quadrimaculatus*, gumbalha) and big eye scads (*Selar crumenophthalmus*, mushimas) occurred during this time period in B. Kamadhoo. This large-scale event of the death of big eye scads and herrings occurred in the island harbour starting from the 24th of November 2007 (refer photos 12 - 14 below).



Photo 12. and 13. The eastern side of the harbour where all the dead had accumulated
(Photos by Ahmed Najeeb, MRC)



Photo 14. Blue striped herring (Gumbalha) (Photo by Shahaama Sattar, MRC)

The harbour has a single entrance on its northern side and the absence of a second opening decreases the amount of circulation in the area. The water in the harbour was murky and greenish/grey in colour.

Methodology

Coincidentally as this occurred in Kamadhoo while a team from MRC were present in the atoll, we were able to visit the site and obtain fish and water samples.

Water samples were collected using a modified Nansen bottle (Photo 15) which was sunk to the bottom of the basin and unplugged. Once the collector was full, this was then brought up to the surface and oxygen levels then measured using a HI 9142 portable waterproof Dissolved Oxygen meter. Samples were also collected and frozen to be sent to NHL. These samples were tested for the presence of nitrates, sulphates, phosphates, trihalomethanes and turbidity.

Fish samples of dead individuals collected were frozen on spot and sent to NHL to test for ciguatoxins and *Staphylococcus aureas* infections of the spleen.



Photo 15. Modified Nansen bottle used for water collection in B. Kamadhoo

Results and Discussion

Oxygen readings in the harbour

The dissolved oxygen readings in the harbour varied depending on the area of the harbour sampled. Readings ranged from 1 to 7.6 mg/l where the lowest reading of 1mg/l was shown from the water sample taken from the area where all the dead fish had accumulated. The highest reading of 7.6 mg/l was observed at the outer entrance to the harbour. Although these levels of oxygen are reasonably low for sea water, it is not possible to conclude that this is the cause of this mass death. Death brings about decomposition which uses up large amounts of oxygen. Death of this scale in a small enclosed area would naturally use up almost all the available oxygen in the water. Thus in this event the low oxygen levels could be an effect rather than its cause.

Presence of chemicals in the water samples

Results for the presence of nitrates, sulphates, phosphate, trihalomethanes and turbidity are shown in table 9.

Table 9. Results of the analysis of water samples obtained from B. Kamadhoo

	Sample 1	Sample 2	Reference levels
Nitrates	0.0mg/l	0.0mg/l	0.5mg/l*
Sulphates	75mg/l	75mg/l	885mg/l*
Phosphates	0.20mg/l	0.24mg/l	0.07mg/l*
Trihalomethanes (as CHCL ₃)	0ppb	0ppb	
Turbidity	0NTU	0NTU	

*Reference levels taken from Gross (1972)

According to the above, water samples did not show any turbidity and did not have any nitrates or trihalomethanes present. However, test results for nitrates could also be limited by the capacity of the laboratory to test for very low levels of nitrates.

Toxic and Microbiological tests on fish samples

Fish samples were negative for ciguatoxins and positive for the presence of *Staphylococcus aureas*. However, as with those of *O. niger*, the presence of *S. aureas* does not conclude anything and we need to carry out further tests prior to making any conclusions from these results.

Sightings of dead baby dolphins (October – November 2007)

Apart from the reports of mass deaths of *O. niger* in various regions of Maldives, we have also had coinciding sightings of dead baby dolphins in different regions of Maldives during this period (Table 10). Although we have always received records of dolphin 'standings' and sightings over the years, these have normally been of adults and larger individuals.

Table 10. Sightings of dead baby dolphins

Date	No. found dead	Approx. Length (ft)	Location
Early Oct 2007	1	2-5	Near V. Bodumohora
Early Nov 2007	6	2-2.5	Near V. Felidhoo & Rakeedhoo
24-Nov-07	1	2-2	Near K. Gulhi
30-Nov-07	1	1.5-2	Near Rakeedhoo Channel

According to Dr. Charles Anderson who has been keeping records of cetacean sightings and strandings in the Maldives for nearly 20 years, there have never been recordings or sightings of young deaths over such a short period of time. He has also states the following:

“Possible causes include changes in food availability, leading to failure of milk production by mothers. Or disease (marine mammals are know to be susceptible to Morbilivirus).”

In consultation with Dr. Charles Anderson it would be worthwhile to obtain specimens of these dead dolphins in order to identify the species and also carry out pathological tests. These are critical steps which should be taken before we can draw conclusions as to what is causing the deaths of these baby dolphins. Furthermore additional tests need to be carried out before making any connections between the deaths of these baby dolphins and the mass fish kill being experienced throughout Maldives.

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