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Mosquitoes in Belnem on the island of Bonaire: Sources and management implications.

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Mosquitoes in Belnem on the island of Bonaire: Sources and management implications.

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Foreword

Dear reader,

This report is the product of my final thesis project, which I have carried out between October 2015 and April 2016. In this period I have learned a lot about mosquitoes, their habitat and doing research in the field. It was really cool to see what can be achieved with the data on a policy level, and how the right management approach can possibly save lives.

During this project I had help from several people whom I would like to thank now.

My sincere gratitude for Sabine Engel from STINAPA, who was so kind to show me the island and help me wherever she could.

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Executive summary

The people of Bonaire are affected by diseases that are transferred by mosquitoes. The problems are related to specifically the *Aedes aegypti* mosquito. It was unknown if these mosquitoes originate from the residential areas themselves or from a natural mangrove area close by. The general perception of the residents is that the mangrove area is a possible source for the mosquitoes in residential areas. In order to decide on the best management approach to deal with human health risks connected to mosquitoes, the origin of the mosquitoes in the residential areas was examined as well as the current mosquito control methods.

In order to find out what an effective way is to deal with the mosquitoes that are posing a risk to the residents of Belnem a field study was performed. In the field, mosquitoes and abiotic factors were measured across a transect of study sites from the residential area to the natural area. Five main study sites were selected, and for each study site 3 areas were selected (approximately 50 meters apart) to conduct measurements (15 locations in total). For 13 weeks, data was collected on all study sites. During each measurement, the human landing catch (HCL) method was applied for 10 minutes and all mosquitoes during that time were caught with an aspirator. Furthermore, data about various abiotic variables was collected in order to understand the fluctuations of mosquito abundance over time and location. (6 locations were added later because more insight about the range of the mosquitoes was desired)

Commented [BNMP1]: Acronym HLC? Human Landing Catch

Four species of mosquitoes have been found during 219 HCL sessions of 10 minutes on 21 different locations. A total of 208 mosquitoes were caught. The species were the *Culex quinquefasciatus*, *Aedes taeniorhynchus* and the RTU 3 mosquito (until species is identified the term Recognizable Taxonomic Unit is applied). On the additional study sites near the residential areas *A. aegypti* was found. The pattern shows that the population of *C. quinquefasciatus* is only present in the residential area of Belnem. It also shows that the *A. taeniorhynchus* and the RTU 3 mosquito population are located only in the salt marsh mangrove area of Lac Bay. No mosquitoes were caught between Belnem and Lac Bay. This indicates that there are two separate sources of mosquitoes, Lac Bay and Belnem, each with their own species.

In the first two weeks no mosquitoes were caught. In week 7 a peak in abundance took place and then declined again after one week. This could be explained by a positive and significant correlation between the number of mosquitoes and the quantity of rain. Relative humidity also had a positive significant relation with mosquito abundance. Furthermore, the species found in the residential area, *A. aegypti* and *C. quinquefasciatus,* cannot survive the hypersaline environment found in Lac Bay (>40‰). This may explain why those species were not found in the mangroves in dry conditions.

The *A. aegypti* acts as a vector for dengue, chikungunya and zika, this mosquito cannot survive the saline environment and is not found in the mangroves it can be concluded that the mosquitoes in the mangroves of Lac Bay do not pose a risk to the residents of Belnem in the current dry conditions. An effective way to deal with the mosquito issue in Belnem is to keep using the current strategy of eliminating habitat and in addition start monitoring the quantity of rain. After heavy rainfall breeding places can be checked immediately and high

mosquito abundance's can potentially be prevented. Another addition is to instigate more participation of the residents by using a door-to-door strategy. This way, mosquito abundance can potentially also be controlled effectively in a wet year.

However, the results of this project are based on data that were collected during a very dry fieldwork period, so the results and recommendations should be viewed in that light. In order to have a more accurate idea of the mosquito distribution in wet conditions, more research should be performed in a wet year.

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1. Introduction

Mangroves are important wetlands and have significant roles worldwide, they prevent soil erosion and are an important nursery habitat for many aquatic species. Almost 35% of the world's mangroves have disappeared (WWF, 2016). This is due to the anthropogenic pressure which is increasing, mangroves are cleared, overharvested or otherwise misused, so mangroves are in need of protection (WWF, 2015). In the Caribbean Sea on the island Bonaire is a Bay with the largest Dutch Caribbean mangrove wetland area of around 700 hectare which is called Lac Bay. The Bay is protected by an intergovernmental treaty that provides the framework for national action and international cooperation for the conservation and wise use of wetlands and their resources (Ramsar, 2015). This programme is aiming at ecosystem based management for conservation and sustainable use of wetlands and their resources, for the benefit of people and nature (Ramsar, 2014). This treaty, Ramsar, is interested in Lac Bay because it is an important ecological area; it provides a nursery for reef fish and is a habitat for the endangered conch and sea turtles. The Bay also acts as a resting and feeding ground for many species of water birds and is therefore appointed as an Important Bird and Biodiversity Area (IBA) (Debrot et al. 2012) (OLB, 2014).

Mosquitoes are an essential part of this mangrove ecosystem. Lac Bay is a nursery and a habitat for mosquitoes, and they have a central place in the food-web (Xerces, 2013). The larvae are food for other aquatic organisms and the adults are hunted by birds, bats and arthropods such as spiders and dragonflies. Moreover, mosquitoes act as pollinators for plants. These roles of the mosquito are important in the mangrove ecosystem (Connelly and Carlson, 2009). Therefore a sufficiently large mosquito population is desirable. To maintain the mosquito population the mosquitoes need to reproduce. The female mosquito lays eggs for which she needs protein. She gets this protein from blood, either from animals or humans. When the female mosquito sucks up blood from one human or animal and then continues to feed from another, she can transmit several serious deceases like dengue, yellow fever, microfilaria, malaria, chikungunya, West-Nile virus, elephantiasis and zika (Debrot, 2014). Therefore mosquitoes are a possible public health threat to the local population. In the history of Bonaire several epidemics of dengue and yellow fever transferred by mosquitoes have occurred (Kuyp, 1973).

So, on the one hand the mosquitoes are essential to the mangrove ecosystem, but on the other hand they might form a public health threat to the inhabitants of the island. A bridge must be built between the interests of keeping the mosquitoes for the roles they play in the ecosystem and reducing their numbers to protect the local population. In this project under supervision of IMARES (Institute for Marine Resources & Ecosystem Studies) and STINAPA (Stichting Nationale Parken Bonaire) a field study has been conducted to collect data on the mosquitoes and their source habitat. Also, in order to see if mosquito control on the island is effective, an official has been interviewed concerning the current mosquito control management approach and literature study has been conducted about mosquitoes in general and mosquito control management strategies.

1.1 Problem description

The people of Bonaire are affected by diseases that are transferred by mosquitoes, specifically the *Aedes aegypti*, in residential areas of Kralendijk (C – Figure 1) and Belnem (B – Figure 1). In 2014 on Bonaire a total of 39 cases of chikungunya, which is a disease transmitted the *A. aegypti*, have been reported to the European Centre for Disease Prevention and Control (ECDC, 2014). The name "chikungunya" derives from a word in the Kimakonde language from an ethnic group in southeast Tanzania, meaning "to become contorted", and describes the stooped appearance of sufferers with joint pain (WHO1, 2015). Another problem is dengue, from 2000 to 2008 the Caribbean reported 193,491 dengue cases including 3,685 cases of dengue haemorrhagic fever (DHF) causing 353 deaths (PAHO, n.d.). In 2011 a total of 648 people were infected with dengue on Bonaire (Eenvandaag, 2012). In order to effectively control the mosquito population, reducing human health risks, it is important to know the main sources and breeding locations of the mosquitoes. Currently, on Bonaire, that has never been studied. The general perception of the residents is that Lac Bay mangrove area (A – Figure 1) is a possible source for the mosquitoes in residential areas such as Belnem and Kralendijk. This hypothesis is supported by the fact that the trade winds are coming from the east (Windfinder, 2015) and might transport the mosquitoes towards the area of Belnem.

Figure 1, Map of Bonaire, The mangroves are visible in Lac Bay (A) The residential areas of Belnem (B) and Kralendijk (C) are visible on the west side of the island (Google Maps, n.d.).

Should Lac Bay indeed be the breeding place for the mosquitoes in the residential areas, then a management approach to reduce the public health risk that mosquitoes cause should be focussed on the area of Lac Bay. But should the origin of the mosquitoes lie in the residential areas themselves, then the mosquito management approach should focus on the residential areas.

Nowadays Lac Bay has more and more shallow semi-dry areas caused by the construction of roads that cut off water circulation, and by excessive grazing of the mangrove by donkeys and goats. Therefore a restoration plan has been formed to re-establish water circulation to stagnant areas of the Lac Bay. This is achieved by dredging and clearing channels, which improves the water quality and reinstates the natural functions of the area (Debrot, 2014) (OLB, 2014). The plan aims to improve Lac's water circulation. This would mean a decrease in breeding locations to the original level when the water circulation was not obstructed.

The restoration plan is a form of Open Marsh Water Management (OMWM) which is an open water system using physical changes in the habitat for controlling mosquitoes (DNREC, 1985). Further adjustments of the restoration plan in terms of effective mosquito control might be necessary should Lac Bay prove to be the source of the mosquitoes that are causing a public health threat.

To protect Lac Bay and help the people of Bonaire reduce their mosquito problem, fieldwork has been conducted, qualitative data was collected from residents and an official, and a literature study on mosquito control has been conducted so that advice can be given for a fitting management approach.

1.2 Research specification

Problem statement

The people on Bonaire are at risk from viruses transmitted by mosquitoes. So far it is not known if these mosquitoes originate from the mangroves or in the residential areas themselves. This knowledge is crucial to decide on the best management approach to reduce the public health threat caused by the mosquitoes and protect the mangrove ecosystem with its natural mosquito population at the same time.

Research goal

In order to decide on the best management approach to deal with human health risks connected to mosquitoes, the origin of the mosquitoes in the residential areas must be examined as well as the current mosquito control mechanisms.

Main research question

What is an effective way to deal with the mosquito issue in the area of Belnem, and do the mosquitoes in the mangroves of Lac Bay pose any risk to the residents of Belnem, Bonaire?

Sub-questions

1. What mosquito species are occurring in the area from Lac Bay to Belnem and how are they distributed over the area?

2. Which abiotic parameters, such as humidity, pH, water temperature, wind speed, salinity, water body size and vegetation can be linked to high mosquito abundance of these locations?

3. What is the current mosquito control management approach and how can this be improved?

2. Materials & methods

In the first phase of the project a research proposal was written, for this proposal a desk-study was performed about open water management, mosquito life cycles, species and habitat. Another part of the desk-study was to prepare the method for fieldwork. Information about these subjects was taken from scientific papers and research reports. The guidelines for entomological surveillance of malaria vectors in Sri Lanka gave a standardized and comprehensible guide on 'direct landing catches' method of mosquitoes from human bait (Abeyasingha *et al*, 2009). This information and was used in designing the methods described below.

2.1 Fieldwork methods

On the 15th of November 2015 the fieldwork period started. The first week was spent exploring the area and setting up transect from Lac Bay to Belnem in order find sources of mosquitoes causing a potential health threat in the residential areas. Then, for 13 weeks data was collected on all study sites once a week.

2.1.1 Selecting the study sites

To select the study sites, a straight line was drawn from Lac Bay to the residential area of Belnem. It appeared to be impossible to select study sites on a straight line through the area directly from Lac Bay to Belnem (Red line from A to E in figure 2). There were no roads or passages through the uneven rocky grounds, cactuses and thorny bushes. Therefore, the following 5 main study sites were selected (see Figure 2), and for each study site three areas were selected approximately 50 meters apart to conduct measurements (so in total, there were 5*3 = 15 locations where measurements took place). The fieldwork period took 13 weeks, every study site was visited once a week so that means 13 measurements for each of the 15 locations. Figure 2 shows a part of Bonaire with the location of the study sites.

Figure 2, Map of the location of the study sites (Google Maps, n.d.) From east to west, study site A is a natural mangrove area, study site B, C and D are intermediate study sites and study site E is an urban study site.

Table 1, Overview of the characteristics of the study sites. Not all study sites had water bodies which makes it not available (N/A).

Study site	Water body	Vegetation
A - Mangrove	Large saline water body	Shrub layer cover
B - Intermediate	Sweet water wells	Patchy shrub layer
C - Intermediate	N/A	Herbaceous layer
D - Intermediate / Urban	N/A	Shrub layer cover
E - Urban	N/A	Patchy herbaceous layer

2.1.2 Additional locations

The last three weeks of fieldwork, six study sites were added in order to give more insight how far the mosquitoes from A and E were venturing land inwards. This was because locations B, C and D were only giving measurements of 0 mosquitoes. The area of these study sites are shown in the green circles in figure 2. Three of them near the mangroves and three near Belnem. The locations had similar conditions as locations A and E.

Also, it was suspected that the *Aedes aegypti* (vector for dengue, chikungunya and zika) would be caught on the transect. When this was not the case near the end of the fieldwork period an ad hoc attempt was made to find a population in the residential area on the north side of the airport as shown in figure 3. Here proof of existence was found for *A. aegypti.*

Figure 3, study site F and G are located in urban areas just north of the airport (Maps, n.d.). The *Aedes aegypti* was not found on the transect but on the other side of the airport in Kralendijk.

Interviews local residents

While exploring to find the best study sites contact was made with two households. The first was located close to study site B, the other was near study site D. The leading question was if they had noticed any mosquitoes lately and if they had experienced any high mosquito abundances in the past. This data was used to see if the negative outcomes of the measurements at the study sites were bad luck or that there were just no mosquitoes. The same people were interviewed in week 2, 3 and 4 of the fieldwork period.

2.1.3 Study site A

Study site A was located at the west side of the mangrove area. This study site represented the natural salt marsh mangrove part of the transect. The study site was on the edge of the mangrove near a large water-body. Figure 4 shows a zoomed in map of the location showing the exact places of study sites A-1 A-2 and A-3. The study sites are 50 meters apart parallel to the road as close to the edge of the mangroves as possible. Figure 5, 6, and 7 are pictures from the study sites and indicate a dense layer of shrub vegetation. The study sites were on the border of impenetrable mangrove vegetation and water bodies.

Figure 4, locations A-1, A-2 and A-3 were located on the western edge of the mangrove area. The locations were approximately 50 meters apart (Maps, n.d.).

Figure 5, Study site A-1 was a mangrove area and has a large saline water body which is covered with a shrub layer of vegetation. The ground looks muddy but it was in fact hard substrate with a thin layer of sediment.

Figure 6, Study site A-2 was on the edge of the mangrove area, it had a large saline water body and a shrub layer cover.

Figure 7, Study site A-3 was located between the mangrove vegetation and was the furthest in to the mangroves. It had a large saline water body and a dense shrub layer cover.

2.1.4 Study site B

Figure 8 shows a map of the exact location of the three study sites at B. Water bodies in the form of wells were found at this study site, with low salinity. The terrain is open and usually windy with cacti and thorny trees. Figure 9, 10 and 11 are pictures of location B-1, B2 and B-3. This location was selected because of its water bodies and relative close distance to Lac Bay. The vegetation was shrub layer cover but only in patches.

Figure 8, Locations B-1, B-2 and B-3 are located to the north west of study site A. The study sites were approximately 50 meter apart (Maps, n.d.).

Figure 9, Study site B-1 was near an open well which was used by wildlife (donkey's and goats) and local farmers. The area was open except for patches shrub layer vegetation of cactuses and trees.

Figure 10, Study site B-2 had a deeper open well that could be breeding habitat for mosquitoes, surrounding the well was an open area with patches of shrub layer vegetation.

Figure 11, Study site B-3 was the furthest inland of study site B. It had another open well and was surrounded by patchy shrub layer cover of trees and cacti..

2.1.4 Study site C

Study site C was located in the middle of the island along the road. There were no water bodies in the direct vicinity. Vegetation was limited to herbaceous layer cover and some shrub layer cover on the side of the road. This location was chosen because of its central position between A and E. This location was essential in giving data about the possibility of mosquitoes crossing the island using stepping stones from Lac to Belnem or vice versa. Figure 12 shows a map with the exact locations of the study sites at location C. Figure 13, 14 and 15 are pictures of study site C-1, C-2 and C3 which show study sites next to the road in a wide open area.

Figure 12, Location C-1, C-2 and C-3 were located in the middle of the island surrounded by uninhabited wilderness. The locations were approximately 50 meters apart (Maps, n.d.).

Figure 13, Study site C-1 was located next to the road that goes straight across the island. The area was dry and had some shrub layer cover next to the road and elsewhere herbaceous layer cover.

Figure 14, Study site C-2 was very similar to C-1. It is dry, close to the road and mostly had a herbaceous layer cover.

Figure 15, Study site C-3 was also close to the road. There was no water body and had a herbaceous layer cover next to the road.

2.1.5 Study site D

Study site D was located just outside of the residential area of Belnem. This site was protected from the wind by a dense shrub layer cover and had no water bodies in the vicinity. This location was picked because of its close distance to E. Also, according to local residents, the area contained high mosquito abundances in the past. Figure 16 shows the exact location of the three study sites at D. Figure 17, 18 and 19 are pictures of location D-1, D-2 and D-3. The picture of D-1 shows the view of looking south from the study site in to the residential area. The picture from study site D-2 looks north in to the dense shrub layer and the picture form study site D-3 is further away from the urban area.

Figure 16, Location D-1, D-2 and D-3 were located next to a residential area on the west side of the island and just south of the airport. The study sites were approximately 50 meter apart (Maps, n.d.).

Figure 17, Study site D-1 was located on the border of the residential area. There were no water bodies and this site was protected from the wind by a shrub layer cover.

Figure 18, Study site D-2 was located between dense shrub layer cover and had no water bodies.

Figure 19, Study site D-3 was located even further between the dense shrub layer cover and was the furthest away from the residential area of the D study sites.

2.1.6 Study site E

Study site E was the most eastern study site situated in the residential area of Belnem. The area had no natural water bodies and mainly a very patchy herbaceous layer cover for vegetation. There were open areas with roads and buildings but the study sites were well covered against the wind. Figure 20 shows a map of the exact locations of the study sites. Figure 21, 22 and 23 show a picture of D-1, D-2 and D-3. This study site was picked because the houses and buildings on Bonaire had open spaces of uncontrolled growth between them with patchy vegetation. This was also the case in Belnem which makes it representative for many urban areas.

Figure 20, Location E-1, E-2 and E-3 were located in Belem at 'Punt Vierkant'. This residential area was located on the west side of the island close to the ocean. The study sites were approximately 50 meters apart (Maps, n.d.).

Figure 21, Study site E-1 was located between the houses of the residential area. It had a patchy herbaceous layer cover but the study site was well covered from the wind by buildings. There were no water bodies.

Figure 22, Study site E-2 was located next to the road between buildings and herbaceous layer cover, there was no water body in the vicinity.

Figure 23, Study site E-3 was located between the houses of the residential area. It had a patchy herbaceous layer and no water body.

2.2 Data collection

2.2.1 Abiotic variables

Table 2 shows the abiotic variables which have been collected (when possible) during every mosquito measurement, and the devices that were used. The terrain data (vegetation and water body size) was ascribed to several categories. The level of vegetation that provides cover for mosquitoes was judged by the height of the overall vegetation in which the measurement was performed. The width of the water body was measured to show any increase or decrease in the amount of water surface area that could act as mosquito breeding habitat. Measuring depth was not possible in the mangroves.

Table 2, The first column states the measured variable, the second column indicates which device was used to do the measurement and the last column shows which scale, unit or level that was used.

Variables	Device used	Data unit
Wind speed	Wind/thermo meter	m/s
pH	pH/thermo meter	pH scale
Water temperature	pH/thermo meter	Celsius (°C)
Salinity	Refractometer	‰
Humidity	Humidity loggers	RH (%)
Air temperature	Wind/thermo meter	Celsius (°C)
Vegetation cover	Visual assessment	1. Grass/moss layer cover 10cm high 2. Herbaceous layer cover 135cm high 3. Shrub layer cover 800cm high 4.Tree layer cover, Anything over 800cm high
Water body width	Visual assessment	1. Small water body 10cm wide 2. Medium water body 100cm wide 3. Large water body 1000cm wide 4. Major water body Anything above 1000cm wide

2.2.2 Mosquito collection

The collection of adult mosquitoes was achieved by attracting the biting female using human bait, by sitting quietly on a location with exposed arms and legs. The method is called Human Landing Catches (HLC) (Abeyasingha *et al*, 2009). The researcher sat down very still with exposed legs and arms. When the mosquito landed on one of the exposed parts of the researcher it was directly sucked up the mosquito using a mechanical aspirator. Sometimes the mosquito would fly up in a failed attempt but was almost always caught when the mosquito landed again. All mosquitoes that landed during a time frame of 10 minutes per location were caught. The mosquitoes of one 10 minute session were placed in a plastic cup. The cups were labelled with time, date and location. Then they were transported carefully to the lab for determination data processing.

2.2.3 Management data collection

The current management approach on mosquito control was explored by conducting an interview with Mr. van Slobbe. He is a government official of Bonaire responsible for pest (mosquito) control (see interview questions in Appendix 1 and results in Appendix 2).

Another source of information were interviews conducted in the light of vector control in 2012 attended by Dr. Debrot. In this research two government officials (Mr. van Slobbe and Mr. van Arneman) were questioned about vector control on Bonaire. The data was taken from the meeting itinerary.

Interview van Slobbe

Mr. van Slobbe, policy advisor infectious diseases, was interviewed face to face. He is the head of the team that works on mosquito control on Bonaire. Mr. van Slobbe preferred no audio recordings of the interview. The interview questions in Appendix 1 were sent ahead of the interview, but could not be addressed because the information was considered too sensitive, especially in the light of recent rise of the mosquito-related Zika virus (WHO2, 2016). Therefore, more neutral and factual questions about the current mosquito control strategy were asked concerning the following topics: strategy, monitoring and prevention and source reduction methods.

2.3 Data processing

The data downloaded from the humidity loggers and the other abiotic parameter data was taken from the field data sheet and was entered into Excel. The mosquitoes were sprayed with and preserved in alcohol the day after the catch before being identified and counted. This data was also fed into the same data matrix in Excel. Figure 24 and 25 show the equipment used to collect and identify mosquitoes.

Figure 24, Samples and data from the field. Every study site has its own code. On the right of the picture the mechanical aspirator (the black device) used for catching the mosquitoes is visible.

Figure 25, Microscope setup that was used to identify the several mosquito species. This microscope was later switched for a binocular in order to be able to see the entire mosquito and not just parts of it.

2.3.1 Data analysis in SPSS

The data matrix in Excel was put in to the statistical analysis program SPSS. The distribution of the mosquitoes was visualized by making *Boxplots.* This gave an overview of the 15 locations from Lac to Belnem each with their own species distribution and quantity of mosquitoes.

The Spearman rank correlation tests were used to calculate the correlation between the rain in mm and the abundance of mosquitoes. The data was adopted by putting the rain data one week ahead to take in account the week that it approximately takes for mosquito eggs to go through the process of becoming an adult. This way the moment of rainfall and the emerging of adults were approximately at the same time and were tested for correlation.

To find out which abiotic factor, or combination of factors, had an influence on mosquito abundance the *General Linear Mixed Model* (GLMM) was used. This test was chosen because there were several variables measured over time that might have had influence on mosquitoes. Furthermore, a nested design was used to ensure valid results. Locations B, C and D were not taken in to the model because no mosquitoes were caught at these study sites. Also, because only 5 study sites had water bodies, variables concerning water were not taken into account. Only humidity, wind speed and air temperature could be taken in to the GLMM. The model was executed several times in all possible combinations with the different variables in order to find the best covariance structure using the Akaike information criterion. This resulted in a scaled identity covariance structure. This covariance structure was then used in the GLMM model to find out which model is the top model by using the Akaike information criterion. Bad models were deleted until one model explained over 95% of the variances. The results out of this top model were then used in the report. The quality of the GLMM model was determined by calculating the R^2 by generating a predicted value variable while executing the model. This predicted value variable was then correlated with the sum of mosquitoes using the Spearman correlation test.

2.3.2 Qualitative data analysis

The data from the interview with Mr. van Slobbe was combined with the interview data received from the meeting itinerary written by Dr. Debrot (2012), to establish an overview of the current management approach concerning mosquito control on Bonaire.

The current management approach was then analysed by comparing the approach with the available options in mosquito control according to a desk study on the available options of mosquito control in residential and salt marsh areas.

3. Results

3.1 Abiotic parameters and study site characteristics

Table 3 shows an overview of the study site characteristics. Of each study site the average of the three locations was calculated. Location C, D and E did not have a water body so some measurements were not available. The measurements were performed starting at dusk. The average time that the measurements took place was 19:29 with a standard deviation of 42 minutes.

Table 3, Overview of the average value of abiotic variables per study site. Wind speed is measured in meter per second (m/s), air temperature in degrees Celsius (°C), humidity in relative humidity percentage (RH %), acidity with the pH scale, salinity in per mil (‰), water temperature in degrees Celsius (°C) and for vegetation and water body size a scale from 1 to 4 is used, also the range is displayed. For vegetation: 1. Grass/moss layer cover 10cm high 2. Herbaceous layer cover 135cm high 3. Shrub layer cover 800cm high 4.Tree layer cover is anything over 800cm high. For the width of the water body the following levels were used: 1. Small water body 10cm wide 2. Medium water body 100cm wide 3. Large water body 1000cm wide 4. Major water body was anything above 1000cm wide. Some study sites had no water body so data was not available (N/A). Values are displayed with a standard error (SE).

	Study site A	Study site B	Study site C	Study site D	Study site E
Wind speed in m/s	1.79 SE 0.11	2.61 SE 0.18	3.13 SE 0.17	1.68 SE 0.13	1.64 SE 0.14
Air temp in °C	27.88 SE 0.19	27.26 SE 0.17	27.32 SE 0.17	27.84 SE 1.15	27.69 SE 0.15
Vegetation in 1-4	3.00 3	2.56 2-3	2.10 2-3	3.00 3	3.00 3
Humidity in RH %	72.82 SE 0.54	76.31 SE 0.45	74.81 SE 0.54	76.36 SE 0.49	76.47 SE 0.62
Water body in 1-4	3.54 3-4	2.00 2	N/A	N/A	N/A
Acidity in pH	8.80 SE 0.06	9.29 SE 0.07	N/A	N/A	N/A
Salinity in ‰	65.46 SE 2.01	5.45 SE 1.15	N/A	N/A	N/A
Water temp in °C	27.62 SE 0.18	27.42 SE 0.25	N/A	N/A	N/A

Air temperature, humidity, acidity and water temperature appear to be constant over all study sites. On the other hand wind speed, water body and salinity do have differences. Wind speed peaked in the middle of the island, especially at study site B and C. The difference in salinity indicates that study site A has salt water and site B did not and the difference in water body size shows that the sweet water sources at study site B are much smaller that the water body at study site A. The vegetation did not change in fieldwork period. The possible influence of these differences will be discussed in chapter 3.5.

3.2 Species

During thirteen weeks of fieldwork, four different recognizable taxonomic units (RTU) of mosquitoes were found. In total **208** mosquitoes were caught at all study sites combined. The RTU 3's identity is being examined in a research lab on Curacao, until the results are known this species name is not available and will be called RTU 3. In two ad hoc standard measurements outside the transect more than 6 *Aedes aegypti* per measurement were caught. An overview is given in table 4.

Table 4, Overview of the 4 different recognizable taxonomic units, their scientific name, regular name and quantity of mosquitoes caught during the fieldwork period. RTU 3's identity is being examined therefore its name is still not available (N/A).

Recognizable taxonomic units (RTU)	Scientific name	Regular name	Quantity of mosquitoes caught
RTU 1	*Culex quinquefasciatus*	The Spanish house mosquito	**119**
RTU 2	*Aedes taeniorhynchus*	The black salt marsh mosquito	**71**
RTU 3	N/A	N/A	**18**
RTU 4	*Aedes aegypti*	The yellow fever mosquito	**>12**

In the following 4 text boxes more information about the habitat, characteristics and vector role of the four RTU's will be given.

Box 1: *Culex quinquefasciatus*, RTU 1

Figure 26, A picture of several *Culex quinquefasciatus*.

The *C. quinquefasciatus* lives in close relation to humans and prefer an urban landscape. The larvae of these mosquitoes are found in any nutrient rich standing sweet water. After a blood meal the female mosquito oviposits (lays her eggs) directly in the water. Five to eight days later depending on the temperature of the water adult mosquitoes emerge out of the water. The *C. quinquefasciatus* is the primary vector of St. Louis encephalitis virus (SLEv) and next to that it could transmit the West Nile virus (WNv) but is not the primary vector (IFAS3, 2013).

Box 2: *Aedes taeniorhynchus*, RTU 2

Figure 27, Several *Aedes taeniorhynchus* in the lid of a cup.

Larval habitats are restricted to the coastal salt marshes. Eggs are laid in moist soil, not in water. Only with sufficient flooding eggs will hatch. This species can lay its initial clutch of eggs without a blood meal. Egg development can occur in any salinity from fresh- to ocean water. Eggs hatch in 5 to 15 days depending on the water temperature (IFAS1, 2014). This species is not a primary vector concern but the *A. taeniorhynchus* can act as a vector for Eastern Equine Encephalitis (EEE) and Venezuelan Equine Encephalitis (VEE). Normally the viruses affect horses but during an outbreak humans can be affected to (IFAS1, 2014).

Box 3: RTU 3

Figure 28, Five RTU 3 mosquitoes.

This mosquito was caught 18 times at the study site in the mangroves and nowhere else. This indicates a species with a saline habitat. The mosquito was approximately half the size of RTU 2 and had a green colour when still alive. The identity of this species is being examined.

Box 4: *Aedes aegypti* RTU 4

Figure 29, Six *Aedes aegypti* in an alcohol solution.

This mosquito is also known as the yellow fever mosquito. The *A. aegypti* prefers an urban landscape and is present in the tropical and sub-tropical regions (IFAS2). The *A. aegypti* are container-inhabiting mosquitoes. That means that they use standing water in any container to oviposit. The female's main blood sources for protein to produce eggs are humans (IFAS2). This species is the main vector for yellow fever but also for dengue, zika and chikungunya. Their preference for urban areas, humans as their main source of protein and their role as a vector make the species a public health threat (IFAS2).

3.3 Frequency of appearance

During fieldwork, 3 main species of mosquitoes were caught, RTU 1, 2 and 3. The fieldwork period was 13 weeks, over these 13 weeks the amount of mosquitoes that was caught fluctuated. Figure 30 shows the mean of the amount of *Culex quinquefasciatus* (RTU 1) that was caught in the fieldwork period at study site E. In 35 (of 219) measurements the *C. quinquefasciatus* occurred and only at study site E.

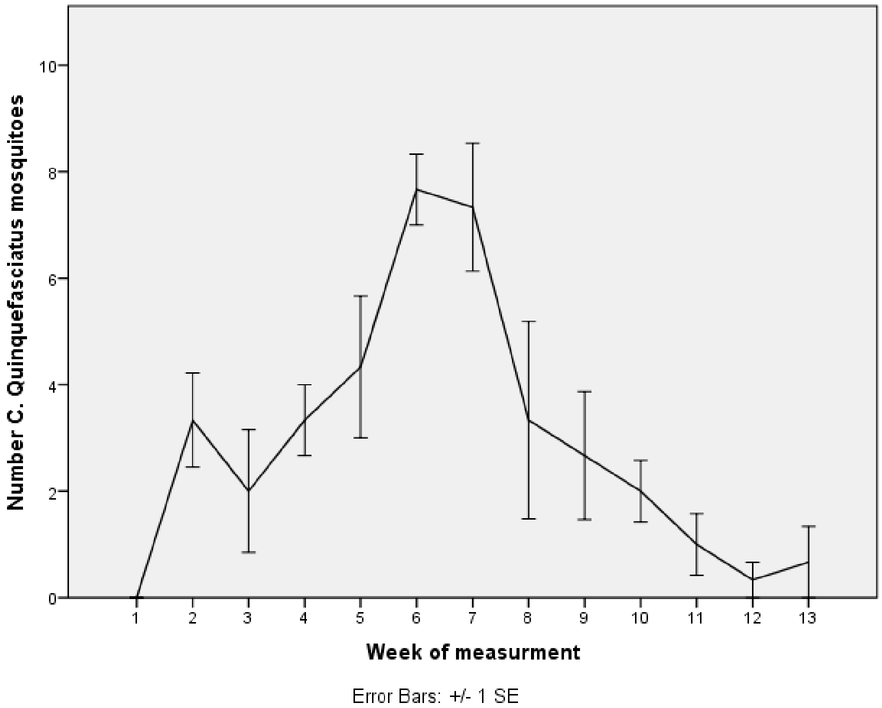

Figure 30, Graph of the mean (+SE) of the amount of *Culex quinquefasciatus* caught at study site E over the fieldwork period of 13 weeks.

Figure 30 indicates a mean amount of approximately 3 mosquitoes per measurement of 10 minutes in the first four weeks. After this initial period the mosquito abundance peaks in week six and seven (approximately 8 mosquitoes in 10 minutes). After the peak the numbers steadily declined to a low point of approximately 1 mosquito in 10 minutes. Over the entire fieldwork period a mean amount of 2.92 *C. quinquefasciatus* with a standard error (SE) of 0.44 was caught at study site E.

The *Aedes taeniorhynchus* (RTU 2) was only caught at the study site A in the mangroves. In 23 (of 219) measurements the *A. taeniorhynchus* occurred. The mosquito abundance fluctuated and is visualized in figure 31.

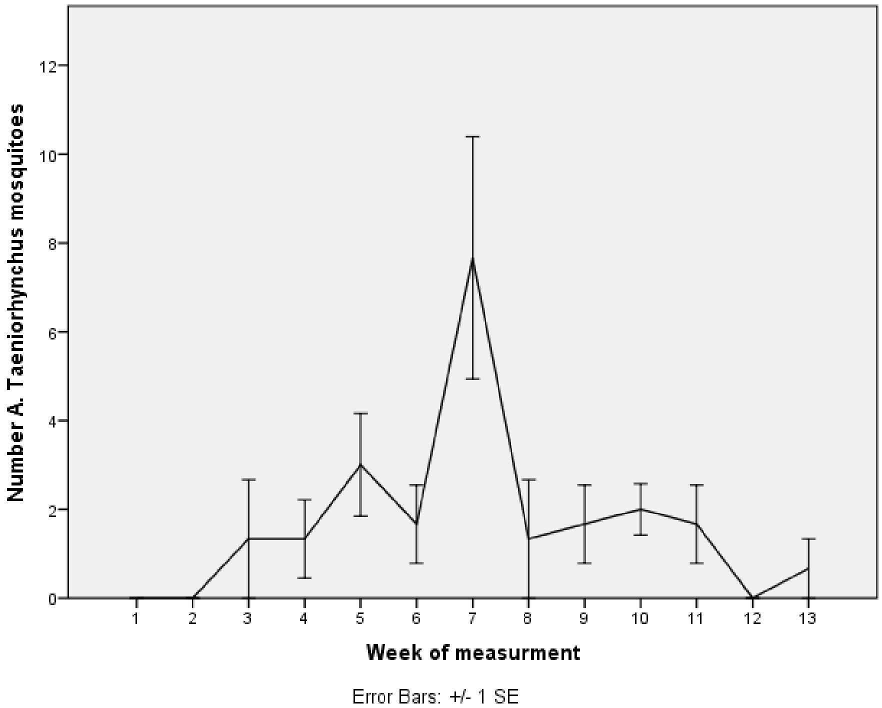

Figure 31, This graph shows the mean (+SE) amount of Aedes taeniorhynchus that was caught at study site A during the fieldwork period of 13 weeks.

In the first two weeks no mosquitoes were caught, then until week six a very low abundance of mosquitoes occurred (approximately 2 mosquitoes during 10 minutes). In week seven a peak in the abundance of the *A. taeniorhynchus* took place (approximately 8 mosquitoes in 10 minutes). The peak in abundance declined again after one week (to approximately 2 mosquitoes in 10 minutes). Over the entire fieldwork period a mean amount of 1.72 *A. taeniorhynchus* with a standard error (SE) of 0.40 were caught at study site A. The peak in abundance of this species in the mangroves happened at approximately the same time as the peak of *C. quinquefasciatus* in the urban area of study site E.

In 11 (of 219) measurements RTU 3 occurred only at study site A in the mangroves. This species had the lowest abundance but is also showing similarities to the graphs of RTU 1 and 2. Just like the other species this species peaked in abundance in week six and seven after which the numbers declined again. Figure 32 visualizes the mean amount of RTU 3 mosquitoes caught over the fieldwork period at study site A.

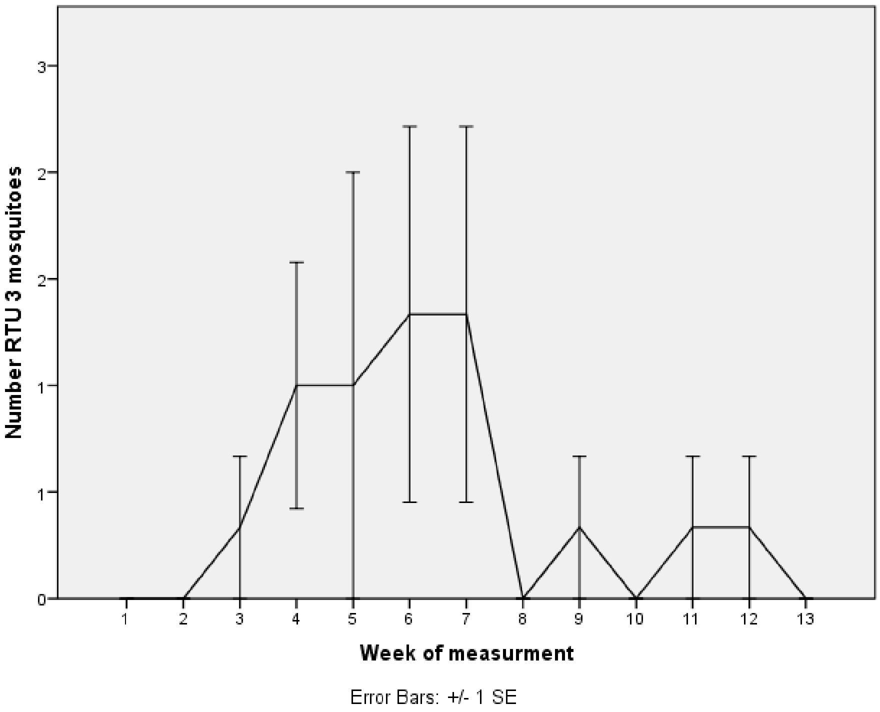

Figure 32, Graph of the mean (+SE) amount of caught RTU 3 mosquitoes over the fieldwork period of 13 weeks.

Over the entire fieldwork period a mean amount of 0.46 RTU 3 mosquitoes with a standard error (SE) of 0.14 was caught at study site A.

3.4 Abiotic variable: Rain

The sudden peak in mosquito abundance in approximately week 7 as described in 3.3 could have something to do with the rainfall that occurred from December 22 till 28 (week 5 and 6 of the fieldwork). Especially on December 26 a total of 31.2 millimeters of rain fell. In total 42.4 millimeters of rain fell in this period. Figure 37 shows this peak of rainfall at week 6 of fieldwork in the graph.

Before this period, on 4 days in the fieldwork period, a little bit of rain fell (starting the 14th of November) with a maximum of 1.9 millimeters. The peak in mosquito abundance was measured at the 6th and 7th of January. This was approximately a week after the heavy rainfall and therefore the heavy rainfall could explain the sudden increase in mosquito abundance.

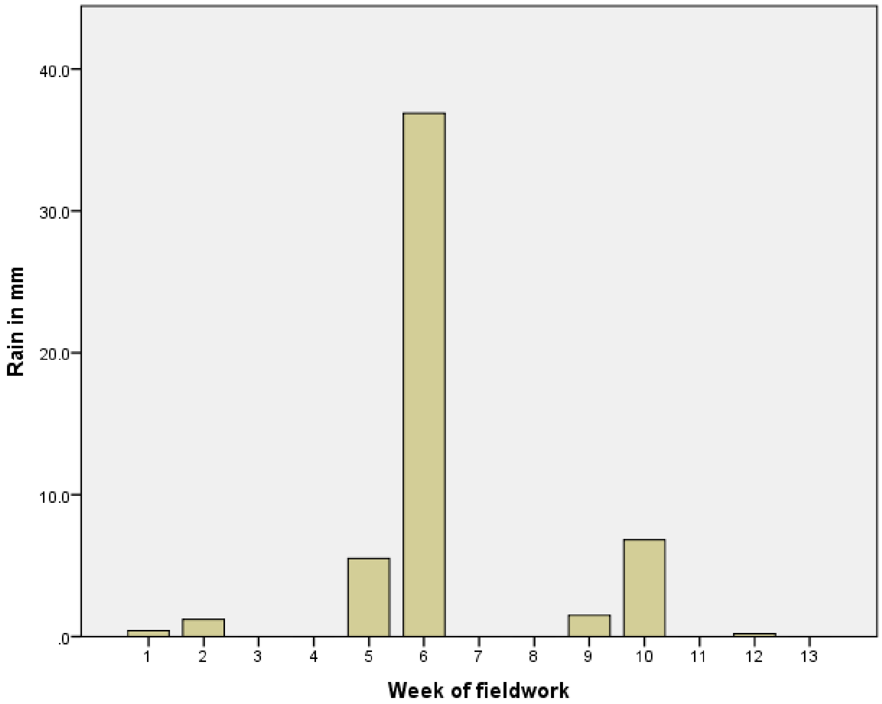

Figure 33, This graph shows the quantity of rain in mm for each week of fieldwork, the red circle indicates that week six had the most rain.

The frequency of appearance of the several mosquito species seems to be correlated to the rain that occurred a week before the peak in abundance.

The Spearman correlation test indicates a significant (N=78 R=0.293 P=0.009) but weak positive correlation (R^2=0,086). This indicates that more rain is associated with more mosquitoes. The scatterplot of the data is visible in figure 34.

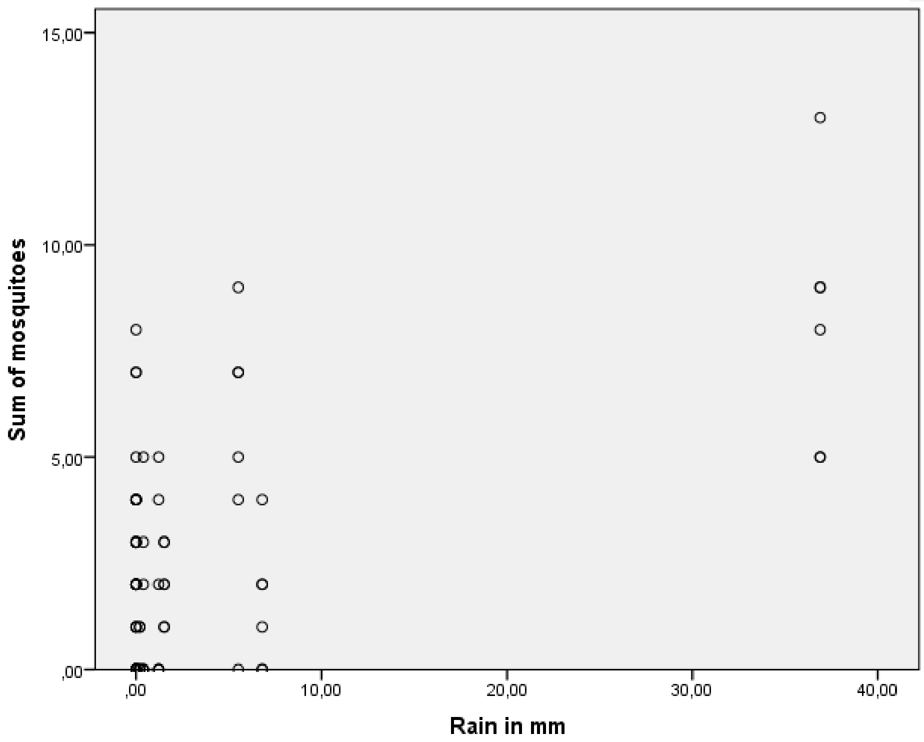

Figure 34, Scatterplot of the total number of mosquitoes and the quantity of rain in a week.

After the peak of rainfall in week 6 of the fieldwork period there was only 8.1 millimetres of rain in the month of January spread out over 4 rainy days. This could explain the decline in mosquito abundance's at the end of the fieldwork period as described in chapter 3.3.

3.5 Distribution of mosquitoes

The previous chapter showed that the quantity of the three different mosquito species that were caught on the transect fluctuated over time. In this chapter the distribution and densities of those species along the transect are given.

Culex quinquefasciatus (RTU 1) was found only at study site E (Figure 34). The mean was 2.92 mosquitoes per measurement at location E. The mean had a standard error (SE) of 0.44. The maximum number of mosquitoes that was caught in one session was 9 and the minimum 0. This species was caught the most compared to the other species.

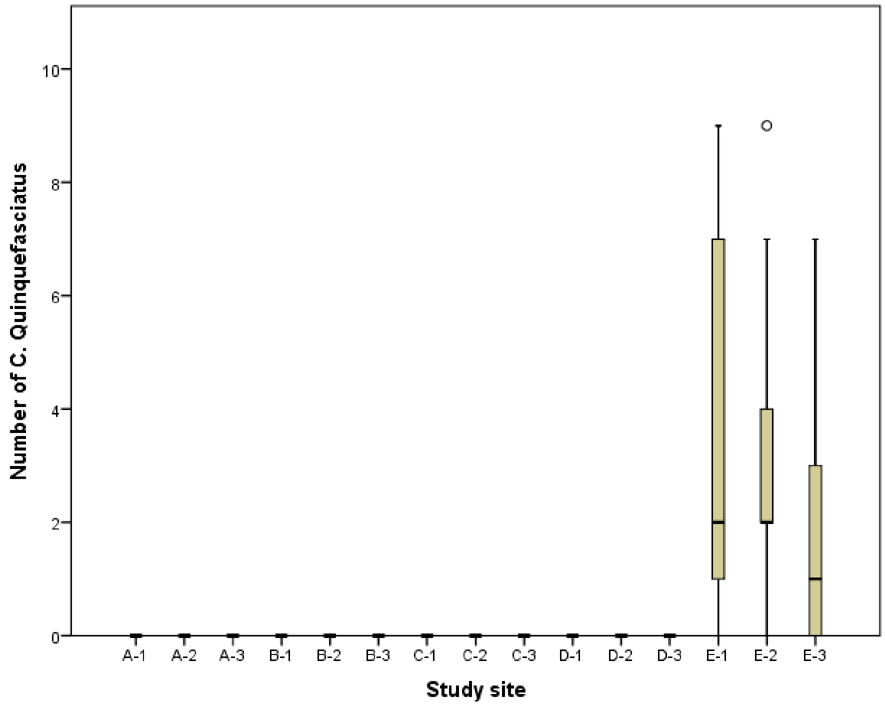

Figure 34, Distribution of the *Culex Quinquefasciatus*. On the horizontal axis the different study sites are given. From left to right is from the mangroves (study site A) to Belnem (study site E). On the vertical axis the amount of *C. Quinquefasciatus* that was caught is shown.

Figure 35 indicates that the *Aedes taeniorhynchus* only occurred at study site A in the mangroves. Location A-1 has an outlier value that was approximately 13 mosquitoes, this instance occurred a week after the peak of rainfall in week 6 as described in figure 31. The mean number of *A. taeniorhynchus* that was caught at location A is 1.72 with a standard error (SE) of 0.40. The minimum number of mosquitoes that was caught is 0 and the maximum 13.

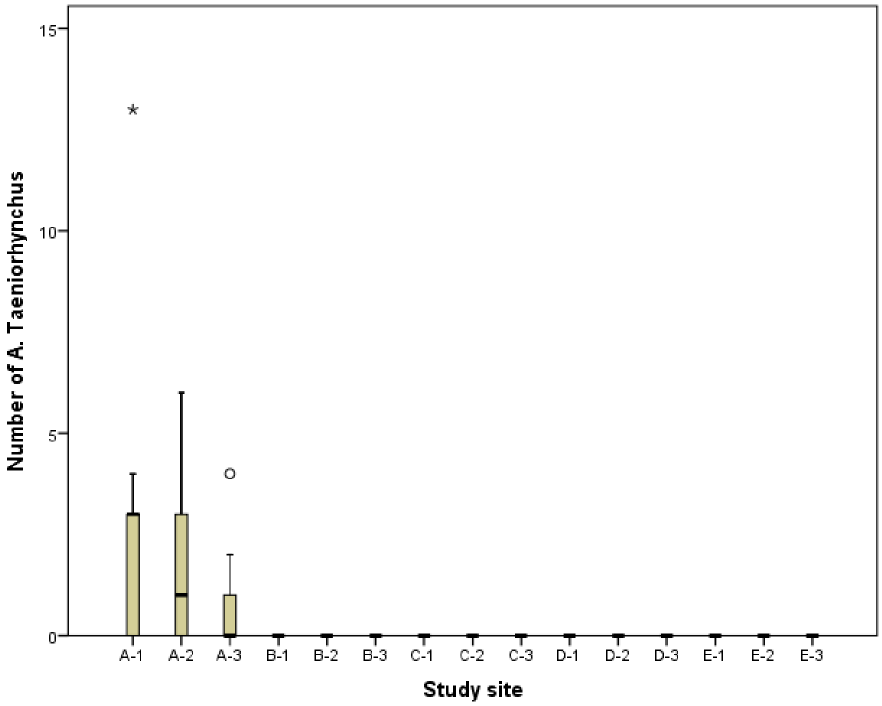

Figure 35, Distribution of the *Aedes taeniorhynchus. On* the horizontal axis the different study sites are given. From left to right is from the mangroves (study site A) to Belnem (study site E). On the vertical axis the amount of *A. taeniorhynchus* that was caught is shown.

Figure 36 indicates that appearance of the RTU 3 mosquito was also limited to study site A. On all other locations this species was not caught. This mosquito species was caught the least compared to the other species with a mean number of 0.46 and a standard error (SE) of 0.142 per measurement.

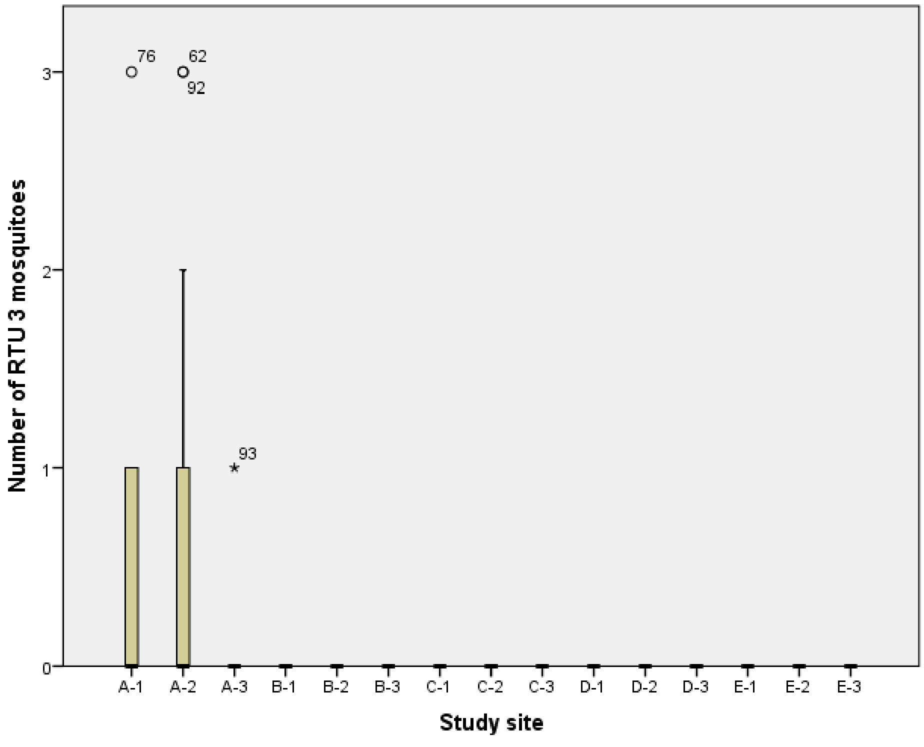

Figure 36, Distribution of the RTU 3 mosquito species. ***On t*he horizontal axis the different study sites are given. From left to right is from the mangroves (study site A) to Belnem (study site E). On the vertical axis the amount of *RTU 3* that was caught is shown.**

The population of *Culex quinquefasciatus* is limited to study site E in de residential area of Belnem and *Aedes taeniorhynchus* and the RTU 3 mosquito population are located only in study site A. No mosquitoes were caught at study sites B, C and D. This indicates that there are two sources of mosquitoes, Lac Bay and Belnem, each with their own species.

3.6 The influence of abiotic variables on abundance

Chapter 3.4 indicated a significant correlation between the quantity of rain and the fluctuation of mosquitoes over time. The location's abiotic variables might also have had an influence on the abundance.

Water (temperature)

The influence of rain was also noticeable in the variable water temperature. The low water temperature in relation to both mosquito species in the mangroves was significantly correlated with the Spearman correlation test (*Aedes taeniorhynchus* R= 0.366 P= 0.022) (RTU 3 R= 0.401 P= 0.011). This means that when the water temperature is low, the abundance of mosquitoes goes up. The decline in water temperature likely caused by rainfall because the air temperature is very constant (mean 27,6 °C SE 0,07 °C).

Salinity

The salinity of the habitat has an influence on species of mosquitoes that breed there because some species have tolerance to a saline habitat and are able to breed there but other species cannot. The *A. aegypti,* the main vector for serious viruses, and the *C. quinquefasciatus* are not tolerant to saline habitat over 30‰ salinity (Ramasamy *et al,* 2011). The measured salinity of water bodies at study site A (mangroves) and B1 and B2 (wells) are shown in figure 38.

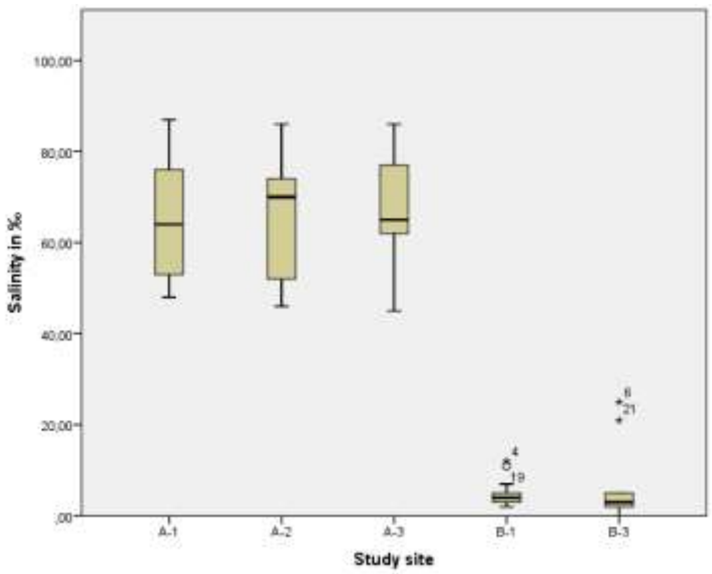

Figure 38, Boxplot of the salinity in pro mille (‰) at the study sites with a water body. A = Lac Bay and B = wells.

Study site A had a mean salinity of 64,23 ‰ with a SE of 3.61 ‰. This was considered a hypersaline environment. The study sites at B have a mean salinity of 5,46 ‰ with a SE of 1.15 and were considered a sweet to brackish environment. In text box 5 the relation between water temperature, salinity and mosquitoes is discussed.

Box 5: Water temperature and salinity effect on mosquito abundance.

The mosquitoes are most likely not affected by the water temperature drop but when rain falls, and because of that the water temperature declines, mosquito abundance goes up. This theory is supported by the fact that the *A. taeniorhynchus* lays eggs in the moist soil on the border of a water body so when it rains and the water temperature declines the eggs are submerged and after a minimum of five days the adult mosquito emerges (IFAS1, 2014). This could explain the correlation between the increase in mosquito abundance in the mangroves and the quantity of rain. In the urban area this correlation could also be explained because the *Aedes aegypti* and *Culex quinquefasciatus* live in close vicinity with humans in and around houses. These urban areas have containers like old car tires, barrels and a variety of other stuff that could hold water. When it rains these containers fill up and become habitat for mosquitoes. Approximately a week later adult mosquitoes emerge (IFAS3, 2013).

The *A. aegypti* and other not saline tolerant species like the *C. quinquefasciatus* are not able to survive in the mangroves which explains there abundance in residential areas and absence in the mangroves.

Humidity

To test if the other abiotic variables (wind speed, air temperature and humidity) had an effect on the abundance of mosquitos a general linear mixed model (GLMM) was used. In this model only the variables wind speed, air temperature and humidity were tested because these variables were measured every time at every location. The only variable that could explain the fluctuation of mosquito abundance was humidity. The relation between humidity and mosquito abundance was positive with a factor of 1.112 (95% Confidence interval 1,042; 1,187). The model had an R^2 of 0,138 which indicates a significant but weak correlation. This indicates that on average with an increase of 1 % in humidity there was an 11.12% increase in the abundance of mosquitoes.

3.7 Mosquito management approach

From the previous chapters it became clear that there are two locations, with different abiotic characteristics, containing different population of mosquitoes; one urban area and a natural mangrove area. There are also two mosquito management approaches in place, one for the urban area and one for the natural area. In both cases the solution is sought at the source of the problem. In mosquito management this is called ‘source reduction’ and it is usually the most effective and economic option of reducing mosquito populations. The ‘source reduction technique’ is accomplished by eliminating mosquito habitat (by draining or filling water bodies), by altering them (by water management) or treating them (with a control agent to kill the mosquito larvae). This way the water bodies are no longer functional for breeding. This technique is also called ‘physical or permanent control’ and can be applied to residential domestic sources and natural areas like salt marshes (Connelly & Carlson, 2009).

On Bonaire a mix of the above mentioned source reduction strategies are applied in the mosquito management approach. In this chapter the current mosquito management approach will be explained first. Then the generally available mosquito control strategies are explained, as well as their applicability on Bonaire.

3.7.1 Current mosquito management approach on Bonaire

The mosquito control strategy of Bonaire is based on the guidelines of the WHO (World Health Organisation). These guidelines are described in the IMS (Integrated management strategy) of the PAHO (Pan American Health Organisation). This is a working strategy designed for countries with the support of international help. The plan focuses on a national, regional and local level on six different key aspects for vector control (PAHO, n.d.) (see figure 37). The strategy focuses on optimal use of resources and funds to reduce the social, economic and health impacts in the Caribbean Sub-region caused by vector borne viruses. When people are ill they cannot function and thus vector borne viruses has impact on all levels of society. From these guidelines, plans have been composed suitable for Bonaire. These plans focus on monitoring and prevention, eliminations of possible habitat, alteration of habitat and treating of habitat with biological and or chemical pesticides (Slobbe, 2016).

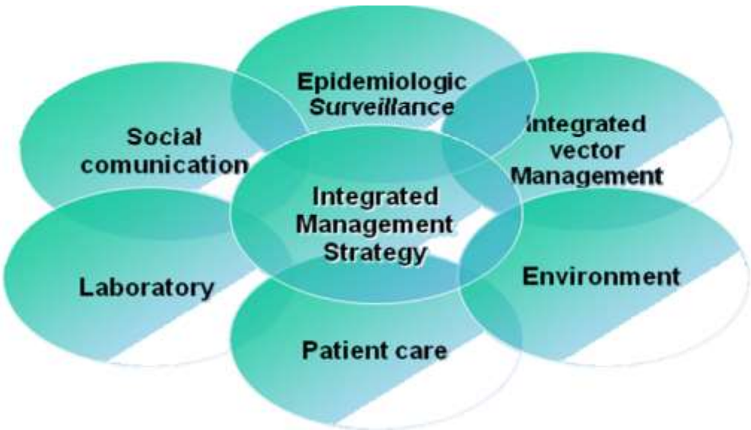

Figure 37, Key components of the IMS vector control (PAHO, n.d.)

Monitoring and Prevention

The vector control team consists of 6 inspectors and one coordinator. Every three months the team of inspectors checks every known breeding place for mosquito larvae and mosquito populations. New breeding places can be reported by telephone. Cases of dengue, chikungunya or zika must be notified to the authorities. When somebody who is infected is taken to the hospital, the municipal health service (GGD) is warned and the inspectors visit the person's home to find out where the infected mosquito possibly came from and then proceed with eradication or treatment of the source.

To prevent infection and increase public awareness and participation, a constant campaign is implemented using most media (Social-media, radio, television, leaflets and brochures) with the aim of education and enabling the local population to eradicate mosquito habitat in and around their homes (Slobbe, 2016) (Arneman, 2012).

Elimination

The elimination of mosquito habitat is achieved by the local municipality by cleaning up illegal waste dumping sites, to limit places with stagnant water (Slobbe, 2016). Arneman and Slobbe indicate that elimination of habitat in peoples own back yards is a crucial point in the mosquito control strategy and should be performed by the residents themselves after rainfall.

Alteration

Alteration in residential areas is achieved by closing off cesspools, which are underground containers holding liquid waste (such as sewage) from a building, by closing off oil drums, and by making buildings mosquito proof by placing fly screens in residential areas. In the mangrove area of Lac Bay there are plans to make channels and build culverts to improve water circulation, thus altering the area and reducing mosquito breeding grounds (Slobbe, 2016).

Treating

On Bonaire, treating habitat with chemicals is a last resort. This means adding chemicals to a water body where mosquitoes breed so that the larvae die (called 'larviciding'). Where possible, more environmentally friendly treatment is achieved by placing Guppy fish in the water that eat the mosquito larvae and pupae. When this is not possible, for instance in case of bigger water containers and reservoirs, habitats is treated with the chemical larvicide *Bacillus thuringiensis israelensis* (Bti). In serious cases, when (infected) mosquito populations reach very high densities, adulticides are sprayed in houses and gardens (Slobbe, 2016). Adulticides are chemicals sprayed in to the air with the intention of killing adult mosquitoes.

3.7.2 General available mosquito control strategies

This study showed that mosquitoes occurred in both residential areas and the mangroves of Lac Bay. The general mosquito control strategies for salt marsh and residential areas are given in table 5.

Table 5, Overview of the general available mosquito control strategies in residential and natural salt marsh areas (Connelly& Carlson, 2009) (IFAS, n.d.) (DNREC, 1985) (EPA, 2000).

	Residential area	Natural salt marsh area
Elimination of habitat	- Draining	- Draining by ditching - Flooding by impounding
Alteration of habitat	- Water management	- Rotational Impoundment Management (RIM) - Open marsh water management (OMWM)
Treating of habitat	- Bacterial - Chemical	- Bacterial - Chemical

The options in elimination, alteration and treating of habitat will now be explained in more detail.

Elimination

The elimination of habitat is usually performed by filling or draining the area from or with water. This technique is not only eliminating breeding habitat for mosquito species, but also for other flora and fauna in an area. Residential areas can benefit from elimination of habitat.

Residential: Elimination (draining)

The *Aedes* and *Culex* species only need very little water to breed in. A simple pet bowl or flowerpot with sufficient water in it can be an excellent breeding location. This is way in Florida there are extensive programs to educate the population in how to eliminate mosquito habitat by disposing or covering standing water containers. There is an extensive communication strategy and sometimes house to house surveillance. The main goal of this approach is educating the local population in a risky area to be able to recognise and eliminate mosquito habitat (Connelly & Carlson, 2009).

Natural: Elimination by ditching (draining)

In salt marshes, ditching is used by making channels (ditches) to increase drainage and thus eliminating mosquito producing sites. These ditches also act as channels for predatory fish that feed on mosquito larva and pupa. When possible the ditches should be connected to a larger pond or channel all year round that acts as a fish reservoir so a population of predatory fish can be sustained. The way a ditch is constructed is important to the effects it has to the area. A very deep ditch accumulates all the soil and thus destroys parts of the area. To overcome this problem, a 'rotary ditching' has been implemented on both coasts of the U.S. Rotary ditching uses high speed rotary equipment to dig shallow ditches and spread the collected soil evenly over the marshes. Environmental damage of rotary ditching is marginal and only concerning the area where the ditch is being dug. This approach is strictly used in vegetated areas, very sandy and loose soils are not suitable because the ditches will not sustain. Planning and maintenance are essential to the success of this approach (Connelly& Carlson, 2009) (IFAS, n.d.).

Natural: Elimination by impounding (flooding)
Habitat elimination by impounding usually means building dikes around an area that is considered a breeding location for mosquitoes. When the dikes are finished, the area is flooded so there is no possibility for laying eggs. Impounding eliminates salt marsh mosquitoes and sand-fly production, but almost all natural marsh functions are eliminated too (IFAS, n.d.).

Alteration
Source reduction is by alteration of habitat is usually achieved by water management in an open water management system. This means that water, nutrients and organisms can flow through the area. Although the system is open, impoundment and ditching are used to steer the water. This could be achieved in urban and natural areas.

Residential: Alteration
In residential areas, roads, buildings and other man-made structures can obstruct the water flow or contain stagnant water. Altering habitat could mean closing off water containment structures or creating an infrastructure to let the water flow. Mosquito habitat is altered by water management.

Natural: Alteration ('RIM')
Rotational Impoundment Management (RIM) is an intermediate between a totally closed and an open impoundment by controlling the inlets and exits. This management approach is aimed at *Aedes* mosquitoes and biting midges that are laying their eggs in soil. After impoundment and flooding the area with a layer of water the habitat for breeding is eliminated. Only impounding and flooding the marshes would mean an environmental disaster for native flora and any natural flow of nutrients and organisms. So the impounding of the salt marshes needs to be managed with pumps and culverts. The aim is to significantly reduce mosquito population while improving the fish habitat and water quality. Many natural marsh functions are maintained or re-established. Seasonal source reduction of mosquitoes and minimal flooding allows indigenous marsh vegetation to survive. However, additional source reduction options like larvicides are necessary to further decrease the size of the mosquito population. Furthermore the flooding of marsh wetlands in summer seasons is not natural and might negatively influence indigenous flora and fauna (Connelly & Carlson, 2009) (IFAS, n.d.).

Natural: Alteration ('OMWM')
Open marsh water management (OMWM) is an open system using physical changes in the habitat for controlling mosquitoes. The main idea is to excavate ponds and ditches throughout the marsh to establish connected waterway system. The ponds act as a reservoir for predatory fish and are attractive as habitat for water birds like the waterfowl. Flowing water and ponds with predatory fish are unsuitable for egg deposition and larval maturation therefor the source of mosquitoes is reduced. Significant advantages of the OMWM are that the use of chemical insecticides is greatly reduced, the adverse effects on marsh flora and fauna are minimal, the created ponds are a habitat for water birds and the approach is cost effective (DNREC, 1985).

Treating

Treating means that the water in which the mosquito breeds is treated with chemicals in order to kill the eggs of larvae. Larvicides kill insect larvae, there are larvicides that use bacteria (microbial larvicides) and larvicides that use chemicals. Microbial larvicides are bacteria that are registered as pesticides, such as the microbial larvicides *Bacillus sphaericus* and *Bacillus thuringiensis israelensis* (EPA, 2000).

Chemical larvicides include substances such as temephos, methoprene, oils, and monomolecular films. The oils and monomolecular films create a layer over the water and suffocate or drown the larvae and emerging adults (EPA, 2000).

Treating a mosquito habitat is usually a part of the mosquito control strategy mostly in urban areas. This is because the use of larvicides imposes risks to the environment in which it is used.

3.7.3 Applicability of management approaches on case Bonaire

This study indicates that there are two separate areas where different species (and populations) of mosquitoes occur. The urban areas of Kralendijk and Belnem on the west side of the island (with the species *Aedes aegypti* and *Culex quinquefasciatus*) and the natural salt marsh mangrove area of Lac Bay on the east side of the island (with the species *Aedes taeniorhynchus* and the RTU 3 mosquito). The mosquito that is causing a public health threat, the *Aedes aegypti,* was not found in the mangrove area of Lac Bay but only in residential areas. These findings have consequences for the choice of a mosquito control management approach.

The natural area of Lac Bay contains the *A. taeniorhynchus* and the RTU 3 which both cause no public health threat, but could be a nuisance in high abundances. The restoration plan will increase water circulation and thereby cause a decline in of stagnant water bodies. This reinstate normal marsh land habitat that will support a normal population of mosquitoes for this kind of ecosystem. A balanced mosquito population is important for the mangrove's ecosystem, as stated in the introduction, therefore no other form of source reduction is applicable. The restoration plan is in fact a form of the previously mentioned open marsh water management (OMWM) and is applicable.

In the residential areas the available options in eliminating, altering and treating mosquito sources are already used and are all applicable. The current mosquito management approach covers the main aspects of an effective mosquito control strategy. The possible improvements can be found in fine tuning the current activities (see recommendations).

4. Discussion & Conclusion

The weather conditions during this fieldwork period were very dry and rainfall did not occur except for a few minor occasions. This had influence for the mosquito abundance. Personal communication with local residents indicated that in other years high abundance's of mosquitoes were present during the same months. The rain season of 2015-2016 is below average. The results and recommendations of this report are based on this period. The low abundance and even absence of mosquitoes measured in the beginning of the fieldwork period could be explained by a below average rainfall for a long period of time starting in April 2015. Table 6 shows the difference between the historical average rain fall since 1968 and the average rainfall this year.

Table 6, Rainfall in 2015 compared to the average since 1968.

	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sept.	Oct.	Nov.	Dec.
2015	49.88	4.93	24.76	0.05	7.62	1.52	2.42	0.21	15.65	15.9	15.89	36.98
AVG.	46.3	21.6	12.3	18.9	19.2	14.9	29.1	28.9	42	68.5	95.4	74.10

The salt marsh mangrove area of Lac Bay is populated by the *Aedes taeniorhynchus* and the RTU 3 mosquito. Their habitat is saline and separated from the human population. On the other side of the island, separated by approximately 5 kilometres of open wilderness, the *Culex quinquefasciatus* and the *Aedes aegypti* have been found. Their habitat is residential areas of Belnem and Kralendijk. These species live together with humans in and around houses. Furthermore, the *A. aegypti* was not found on the initial transect, but was present residential areas. No mosquitoes were found at the three study sites in between Lac Bay and Belnem. The data indicates that in the current dry conditions there are two different separated populations of mosquitoes, a population with the *A. taeniorhynchus* and RTU 3 mosquitoes in Lac Bay and a population consisting of the *C. quinquefasciatus* and the *A. aegypti* in Belnem.

The abundance of mosquitoes was affected by abiotic variables. Significant relations were found with the abiotic parameters humidity, salinity, water temperature and the quantity of rain. Variables like pH, water temperature, wind speed, water body size and vegetation did not have much to do with the mosquito abundance of locations according to the data set.

There was a positive significant correlation between the quantity of rain and the quantity of mosquito's. The influence of rain was also probably found in the relation between low water temperature and both mosquito species in the mangroves (study site A). This indicates that when rain falls, and because of that the water temperature declines, mosquito abundance goes up. The water characteristic salinity of the water body in the mosquito habitat had influence on which species of mosquitoes occurred. Some species (like the *A. aegypti*) cannot survive in saline environments while others like the *A. taeniorhynchus* can. Humidity was significantly associated to the abundance of mosquitoes. The relation between humidity and mosquito abundance was positive and significant but weak.

The first three phases of the mosquito's life cycle are in water. Abiotic parameters such as the quantity/availability or the characteristics of water bodies in an area have the most influence on mosquito abundance on locations. It could be discussed that the low abundance of mosquitoes measured in the beginning and end of the fieldwork period could be explained by the well below average rainfall for a long period of time, starting in April 2015. The low humidity over a longer period of time can decrease the lifespan or be fatal to mosquitoes (Yamana & Eltahir, 2013). Mosquitoes prefer high humidity and this could explain the correlation between an increase in humidity and an increase in the abundance of mosquitoes. The conditions in the mangroves were hypersaline. When the salinity drops because of better water circulation and/or more rain it could be that other species with a lower level of tolerance to salinity will occur in the mangroves.

The data indicates that the abundance or absence of mosquito species on a study site is related to the quantity of rainfall, more rain means more mosquitoes. Furthermore, the salinity of the water determines which species occur at a location and humidity plays a role in the number of mosquitoes that will occur.

Mosquito management

The mosquito management strategy for the natural salt marsh area of Lac Bay is already in place. The restoration plan is focussed on restoring the water circulation to stagnant parts of the area to sustain and improve the natural functions of the mangroves. This plan is in fact a form of OMWM and will reduce the quantity of breeding locations for mosquitoes in the Bay to a natural level. The *A. taeniorhynchus* and the RTU 3 mosquito appear to be no threat to public health but could be a nuisance. With the purpose of managing the mosquitoes in the Lac Bay the restoration plan will most likely suffice. The current mosquito management strategy for the residential areas uses all the available options in eliminating, altering and treating mosquito sources. The details of the current mosquito management strategy are not available because the policy is not fully transparent. There have been no major outbreaks of illnesses transferred by mosquitoes in the last few years so the current strategy appears to be working in the current dry conditions. The question is if the strategy will hold in a wet season.

It could be discussed that for a wet season certain alterations to the current management could be made and more participation from the residents could be necessary for early detection. Recommendations on this subject are given in chapter 5.

Conclusion

In short, it was found that in the current dry condition there are two different separated populations of mosquitoes, a population with the *A. taeniorhynchus* and RTU 3 mosquitoes in Lac Bay and a population consisting of the *C. quinquefasciatus* and the *A. aegypti* in Belnem. The salinity of the water probably determines which species occur at a location and the abundance or absence of mosquito species on a study site is mostly related to the quantity of rainfall.

This means that the mosquitoes in the mangroves of Lac Bay do not reach the residents of Belnem and therefore they do not pose a risk to the residential areas. Furthermore, the main vector for illnesses, the *A. aegypti* cannot survive in the hypersaline environment of Lac Bay and therefor the management approach to minimize the risk on dengue, chinkunguya and zika should focus on the residential areas.

An effective way to deal with the mosquito issue in Belnem is to keep using the current strategy and in addition start monitoring the quantity of rain so that after heavy rainfall breeding places can be checked immediately and high adult mosquito abundance's can be prevented. Another addition to the current strategy is to instigate more participation of the residents by using a door-to-door strategy. This could be helpful to also be able to keep mosquito abundance down effectively in a wet year.

6. Recommendations

1. The first and most important recommendation is to perform this research in a relatively normal to wet year with an average or high amount of rain. The results of this project are based on data that were collected during a very dry fieldwork period this probably affected the results and recommendations.

2. It is recommended to start implementing the mangrove restoration plan as soon as possible, to limit the quantity of breeding habitats. This in order to prevent nuisance to people enjoying recreational activities in and around Lac Bay. The results of the restoration plan should be monitored and evaluated before changes are made to the mosquito control strategy for Lac Bay.

3. The mosquito management approach for the residential areas should be improved by collecting rain data. When heavy rain occurs known mosquito sources should be checked and if necessary eliminated or treated. This should be done instead of, or on top of, the regular three monthly checks of known breeding locations. Three monthly checks is too low a frequency for effective mosquito control, because adults will emerge in approximately a week after heavy rainfall.

4. Participation of local residents is a matter of major importance to keep the populations of *Aedes Aegypti* low, as Mr. van Slobbe and Mr. van Arneman indicated. This is because this species shares its habitat with humans, so the source of this species can be found on private property and residents could take care of breeding locations themselves. Therefore, it is recommended to implement a door-to-door surveillance to show people where mosquitoes breed and how to eliminate mosquito habitat. This personal approach should instigate people to eliminate breeding habitat on their property after heavy rain. The team of mosquito control Bonaire can't do it alone; it is a matter of public health.

5. In order to examine the range of a mosquito species it is recommended to perform an active monitoring test. This means creating habitat for mosquitoes along a transect instead of looking or already excising habitat. Artificial mosquito habitat could be created at a certain range from the established mosquito population. Then, this habitat could be checked often for mosquito larvae. If the mosquito larvae are present then the artificial mosquito habitat is within the range of the current population. If no larvae are found then it is outside of the range of the population. Using this method, mosquito management approaches can be written with a better understanding of the source of mosquitoes and therefore can be made more efficient.

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Appendix

1 Interview questions

For: Government official of Bonaire responsible for pest (mosquito) control

Goal: Document the current management approach concerning mosquito control and subject it to a SWOT analysis

1. **What is currently being done about mosquito control in residential areas?**

(Find out what the current management approach is)

2. **What is the goal of the current mosquito control management approach?**

(Find out what the goal of the management approach is)

3. **Is the effectiveness being monitored and measured?**

(Find out how effective the management approach is)

4. **How effective is the current mosquito control strategy? Strong points, Weak points?**

(Find out strong and weak points)

5. **What could be done to improve the current mosquito control approach?**

(Find out opportunities)

6. **Are there any threats to this management approach?**

(Find out threats)

7. **Is this mosquito control strategy communicated with the local population?**

(Find out communication strategy)

2. Interview Joey van Slobbe

The interview was conducted on Monday 29th of February on Bonaire at the 'openbaar lichaam Bonaire afdeling publieke gezondheid'.

The following information was given during the conversation:

Strategy
The mosquito control strategy of Bonaire is based on the guidelines of the WHO (World Health Organisation). These guidelines are described in the IMS (Integrated management strategy) of the PAHO (Pan American Health Organisation) (PAHO, n.d.). From these guidelines plans have been made that are fitting for Bonaire.

Monitoring and Prevention
Every three months the team of inspectors checks every known breeding place for mosquito larvae and mosquito populations. New breeding places can be reported by telephone.

Casus of dengue, chikungunya and zika have a notification obligation.

When somebody who is infected is taken to the hospital the GGD is warned and the inspectors go to the person's home and work to find out where the infected mosquito possibly came from and then proceed with eradication.

Prevention campaign on most media: social-media, radio, television and with leaflets and information newspaper with the aim of education and enabling the local population to eradicate mosquito habitat in and around their homes.

Elimination
The elimination of habitat is achieved by cleaning up waste dumping sites to make sure that there are no places that will keep water.

Alteration
Alteration in residential areas is done by closing off cesspools, oil drums and by making places mosquito proof.

Treating
Treating mosquito breeding places done by placing Guppy fish in the water that eat the larvae and pupae.

Bigger water containers and reservoirs are treated with larvicide Bti.

When mosquito populations reach very high densities adulticides are sprayed in houses and gardens.

Extra info
There used to be waves of infection but now there are some cases spread throughout the year and with pauses of months.

The correlation between rain fall and the mosquitoes that come after is obvious.

The world is changing, people travel all the time so viruses have a bigger reservoir of victims and viruses spread more easily.