CHAPTER SEVEN

Effects of climatic factors on the occurrence and seasonal variation in populations of *Rastrococcus iceryoides* (Hemiptera: Pseudococcidae) and its associated natural enemies: implications for biological control

ABSTRACT

The fundamental aspect in developing a sound management strategy for a pest is the understanding of its seasonal fluctuation and that of the associated natural enemies. In this study the population dynamics of the alien invasive mango mealybug, *Rastrococcus iceryoides* (Hemiptera: Pseudococcidae) and its associated natural enemies were carried out in two major mango growing areas in coastal Tanzania during the period of December 2008 to June 2010. Destructive sampling, based on random selection of 80 leaves, 20 twigs (~10 cm) and 5 fruits, was carried out on a weekly and monthly basis during the mango and off mango season, respectively. The total number of *R. iceryoides* and the number of mummified mealybug were recorded for each plant part. Upon emergence, parasitoids were counted and identified. The number of predators was collected using beat-sheet techniques. The study revealed that populations of *R. iceryoides* population followed an annual cycle which is synchronized with the mango fruiting season, with a peak incidence occurring during the Northeast monsoon (December-February) at a temperature range of 23 to 33°C, relative humidity of 54 to 86% and total rainfall from 0 to 63 mm. The population trend of *R. iceryoides* is climate dependent and declined sharply following the onset of the heavy rains from March-May, and continues through the coldest and driest period of the year from June-October (Southwest monsoon). The mealybug population and its associated natural enemies were significantly and positively correlated with mean temperature, while it was significantly and negatively correlated with rainfall. The population of the parasitoid and predators were positively correlated with that of the host/prey, *R. iceryoides*. The study also revealed that there was high diversity of hyperparastoids, although their impact on the primary parasitoid was negligible. The findings of this study can be used to model the prediction of *R. iceryoides* population outbreaks and additionally to formulate an effective and sustainable pest management strategy in the agro-ecological system under consideration.

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Key words: Population dynamics, Mango, *Rastrococcus iceryoides*, natural enemies, weather factors, pest management

7.1 Introduction

The mango mealybug, *Rastrococcus iceryoides* Green (Hemiptera: Pseudococcidae), was accidentally introduced from Southern Asia into the African continent in the early 1990s (CABI, 2000). In Africa, *R. iceryoides* together with its close relative *R. invadens* are regarded as the two important exotic mealybug species native to Southern Asia that commonly infest mango. The latter devastated mango production in West and Central Africa but was managed through classical biological control with the introduction of the parasitoids, *Gyranusoidea tebygi* Noyes and *Anagyrus mangicola* Noyes (both Hymenoptera: Encyrtidae) from India (Noyes, 1988; Bokonon-Ganta and Neuenschwander, 1995). *Rastrococcus iceryoides* on the other hand is so far restricted to the East Africa region (mainly Tanzania and Coastal Kenya) and northern Malawi, where it has remained one of the most destructive sucking pests of fruit trees, especially mango as well as various ornamental plants (Tanga, unpublished data; Williams, 1989; Luhanga and Gwinner, 1993; CABI, 2000).

Surveys conducted in Kenya and Tanzania revealed that *R. iceryoides* has an extremely broad host range of about 29 cultivated and wild host plant species from 16 different families. Twenty-one of these host plants are new records for *R. iceryoides*, of which 18 are native to Africa (Chapter 3). The broad host range recorded in Africa strongly suggests that *R. iceryoides* is an emerging invasive polyphagous pest that is capable of rapidly expanding its host range. During the pest outbreak, the high infestation level lead to delays in flowering, fall of floral spikes and leaves, drying up of young fruit-lets, and slowing the growth of new branches due to the severe dieback effects on heavily infested plant parts (Tanga, unpublished data). It is also a nuisance by causing accumulation of large amounts of excreted honeydew that results in the formation of sooty mould, which causes drastic reduction in photosynthesis, leading to reduction in plant growth, flowering and fruiting as well as premature leaf drop. Members of the genus *Rastrococcus* have a potential to become major pests in newly invaded areas. For example, the mango losses caused by *R. invadens* were reported to be as high as 80% in Ghana (Entomological society of Nigeria, 1991), and range between 53% to 100% in Côte d'Ivoire (Hala et al., 2004). Chemical insecticides are among the conventional methods used in controlling *R. iceryoides* in Tanzania and Kenya. However, like other mealybug species *R. iceryoides* proved to be very hard to manage with the use of insecticides as they are covered with powdery hydrophobic wax that repels water-based insecticide solutions (Blumberg and Van Driesche, 2001; Derzelle et al., 2004). The inefficiency of the insecticides in controlling the pest coupled with its unaffordability for most small scale farmers has made mango cultivation uneconomical and has led to the abandonment of mango production in severely affected areas (Tanga et al. unpublished data).

The population fluctuation as well as the distribution of the mealybug pests depend largely upon the prevailing environmental factors (DeBach, 1949), as these pest are known to multiply tremendously during favourable weather conditions leading to population outbreaks (Amarasekare et al., 2008). Climatic conditions also influence natural enemy populations such as parasitoids and predators either directly or indirectly (Arif et al., 2006; Chaudhari et al., 1999). For developing an early warning weather based system for any pest in a specific agro-ecosystem, it is necessary to have basic information regarding population dynamics in relation to prevalent meteorological parameters (temperature, relative humidity and rainfall). This will help in determining appropriate times for intervention, and application of suitable methods of control.

Considering the fact that very little is known regarding the ecology of *R. iceryoides* as a recently introduced pest, and that the role of its indigenous natural enemies is also not documented, a thorough understanding of the interaction between meteorological parameters/pest/natural enemy dynamics is a prerequisite for a weather based pest forecasting model. Therefore, this study was carried out to determine the influence of weather parameters namely temperature, relative humidity and rainfall on population dynamic *R. iceryoides* and its associated natural enemies, which is of great significance in formulating efficient integrated pest management (IPM) strategies for sustainable agriculture.

7.2 Materials and Methods

7.2.1 Study sites

Tanzania is located in East Africa between latitude, 1° to 11°45′ S and longitude 29°21′ to 40°25′ E. The country is divided into four main climatic zones: (1) the coastal area and immediate hinterland, (2) the central plateau, (3) the semi-temperate highland areas, and (4) the high, moist lake regions. This study was carried out in the coastal area, which is characterised by tropical conditions, with temperatures averaging about 27°C, annual rainfall varying from 1000 to 1930 mm and high humidity. In this region the seasons are well defined; these are: northeast monsoon, from December to February (it is hot and comparatively dry), the long rains, from March to May, and the short rains, from November to December and the southwest monsoon from June to October (coldest and driest) (EON, 2011). Within the coastal area the sampling was carried out at two main mango growing localities; Kibaha (06˚ 43' 84" S; 038˚ 46' 07" E, 79 m above sea level) and Dar es Salaam (06˚ 45' 80" S; 039˚ 06' 25" E, 162 m a.s.l). The distance between the two localities is 38.66 km. At both orchard localities where sampling was done the farms were maintained under the same agricultural practices and had not been sprayed with insecticides for the previous two years.

7.2.2 Sampling procedures

The sampling methodology was a slight modification of that described by Pitan et al. (2000) and Bokonon-Ganta and Neuenschwander (1995). During the study period a destructive sampling was conducted weekly during mango fruiting season (October-February), while it was monthly during the mango off season (March - September) for a total period of 19 months (December, 2008-June, 2010). On each sampling date, a group of twenty mango trees were randomly selected at fixed distances and labelled to represent the whole orchard before commencing the sampling. For each sampling date, 80 leaves, 20 twigs (~10 cm long) and 5 fruits were selected at random for mealybug counts. To avoid taking mealybugs only from the upper portions of plants, the order in which plant parts were examined (bottom to top and vice versa) was reversed after each tree. During the survey, care was taken to make sure that no tree was sampled twice within the same month. The samples where then transported to the laboratory at the National Biological Control Program (NBCP), Kibaha. In the laboratory, and on the same day of the sampling, the individual numbers of each developmental stages (first, second, third nymphal instars, ovipositing and non-ovipositing female) were counted and recorded for each plant part. The total number of live individuals was used to represent the infestation level for each plant part and sampling date.

The different plant parts were checked for the presence of mummified *R. iceryoides* nymphs. The mummified mealybug nymphs were carefully removed from the plant parts using a fine hair brush and then placed into transparent plastic rearing containers (22.5 cm height x 20 cm top diameter x 15 cm bottom diameter). An opening (10 cm diameter) was made on the front side of the cage to which a sleeve, made from very fine organza material (about 0.1 mm mesh size) was fixed. The same material was fixed to the opposite opening (10 cm diameter) of the cage to allow for ventilation. A third opening (13 cm diameter) was made on the roof of the cage, which was also screened with the same material. Streaks of undiluted honey were applied to the roof of the cages and maintained in the laboratory at ambient temperatures of 26 – 28 ^{0}C, 70 ± 5 % RH, photoperiod of 12:12 (L : D) h,. The mummies were checked daily until the parasitoid wasp ceased to emerge. Mummified mealybugs from each sampling date and locality were maintained separately. Parasitoids that emerged from the mealybug mummies were counted and recorded. Thereafter, they were kept in vials containing 80% ethyl alcohol and labelled with their respective plant part and sampling date. All parasitoids that emerged were initially identified at Annamalai University, India and later confirmed at the National Collection of Insects, PPRI-Agricultural Research Council (ARC), Pretoria, South Africa. The level of parasitism was calculated as a percentage of the mummified mealybugs in the samples over the total number of mealybug for each plant part and sampling date separately.

Predators of *R. iceryoides* were sampled using the beat sheet technique (Wade et al., 2006), which involved beating 5-10 randomly selected branches of host plants over a 1m^2 cloth screen using a 60 cm long stick. The sampling was done in the morning (8:30-9:30 am). Predators that were dislodged onto the cloth were then counted and recorded before preserving in 80 % ethyl alcohol. Immature stages of predators were reared on mealybugs in perspex cages (15 cm height x 20 cm length x 15 cm width). An opening (10 cm diameter) was made on the front side of the cage to which a sleeve, made of organza material, was fixed. The same material was fixed to the opposite side of the cage to allow for ventilation. The rearing process was carried out in the laboratory set at the National Biological Control Program (NBCP), Kibaha, Tanzania until they developed to the adult stage and were later counted and recorded.

Records of the main climatic factors: daily minimum and maximum temperatures, minimum and maximum relative humidity and total rainfall were obtained from the nearest Meteorological and Agricultural Station in Kibaha. The daily value of these weather parameters were averaged to correspond with sampling dates.

7.2.3 Data analysis

Data are presented as means (± SE) per twig, leaf and fruit. A Poisson generalized linear model was implemented using generalized estimating equations to examine the effect of the weather parameters, namely, mean temperature, mean relative humidity and rainfall on the population dynamics of the mealybugs and the number of mummified mealybugs on the twigs, leaves and fruits. An AR-1 correlation structure was used for the generalized estimating equations (Zuur et al., 2009). Chi-square goodness of fit was used to test if the infestation level by the mealybugs, and the number of mummified mealybug were the same on the different plant parts. During the mango fruiting season, comparisons were made among the three plant parts (fruit, leaves and twigs), while during the mango off-season, comparisons were done only for the leaves and the twigs. The association between the mealybug and parasitoid populations on the different plant parts was assessed using correlation. All the analysis was performed in R 2.13.1 (R Development Core Team, 2011).

7.3 Results

7.3.1 Local variation of ambient temperature and relative humidity

During the entire study period, temperature and relative humidity were generally high, with an average minimum temperature of 23.34 °C and an average maximum of 31.93 °C. The average minimum relative humidity (RH) was 63.20 % and an average maximum of 83.82%. Total rainfall ranged between 0 mm and 259.2 mm throughout the study period. The mean values of temperature and relative humidity recorded during the study period from December 2008 to June 2010 is illustrated in Figure 7.1.

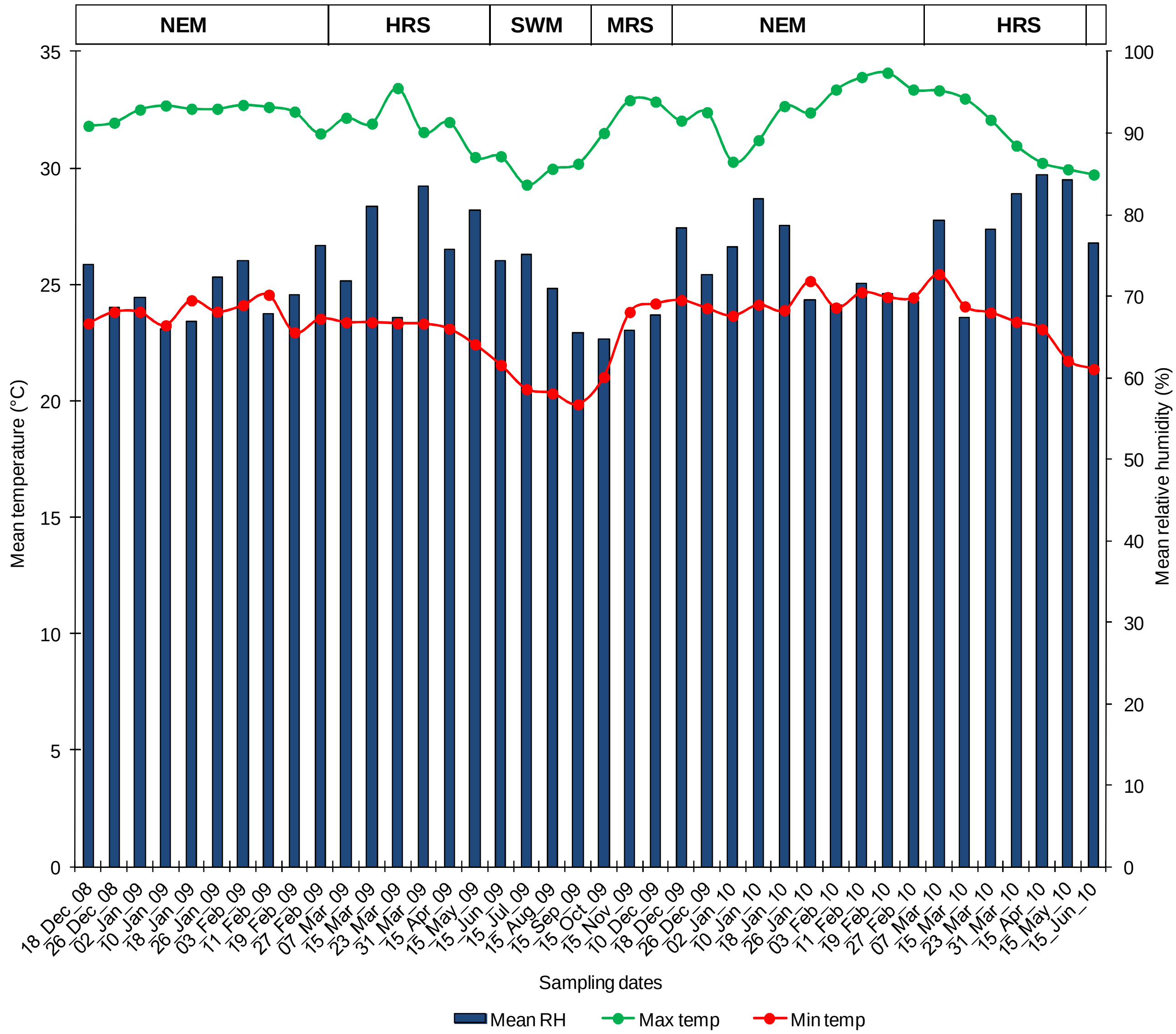

Figure 7. 1: Mean temperature and mean relative humidity in the Coast region of Tanzania from December 2008 to June 2010. NEM (December-March): Northeast monsoon (It is hot and comparatively dry); HRS (March-May): Heavy rainy season; SWM (June-October): Southwest monsoon (It is coldest and driest); MRS (November-December): Mild rainy season. Data obtained from Meteorological and Agricultural Station in Kibaha – Coast region, Tanzania.

7.3.2 Seasonal infestation levels by *R. iceryoides* on mango in Kibaha

The results of the survey showed that there was a great seasonal fluctuation in the population of *R. iceryoides* on the different mango plant parts with respect to rainfall (Figure 7.2). During the 2008/09 mango fruiting season the highest infestation levels on the fruit (171.2 ± 85.82 mealybugs/fruit) was recorded on the 26th of December 2008 (mean temp. of 27. 9°C, mean RH of 68.6%, and total rainfall of 4 mm). While the highest infestation on the twig (324.05 ± 82.26 mealybugs/twig) and the leaves (136.06 ± 17.89 mealybug/leaf) was recorded on 10th of January (mean temp. of 27.9°C, mean RH of 66%, and no rainfall). For the same mango fruiting season the lowest infestation level on the leaves (34.43 ± 8.03 mealybugs/leaf), and twigs (36.5 ± 9.54 mealybugs/twig) was recorded on 18th December 2008 (mean temp. of 27.6°C, mean RH of 73.8%, and rainfall of 44.6 mm), and 27th of February 2009 (mean temp. 27.5°C, mean RH of 76.19%, and rainfall of 6.1 mm), respectively. While the lowest infestation level on the fruit (0 mealybug/fruit) was recorded on 26th of January 2009 (mean temp. of 28.2°C, mean RH of 72.3%, and rainfall of 19.5 mm) and 19th of February 2009 (mean temp. of 27.7°C, mean RH of 70.2%, and total rainfall of 63.2 mm). During this mango fruiting season, the infestations by *R. iceryoides* varied significantly across the different plant parts (twigs, leaves and fruit) ($\chi 2$ = 570.54; *df* = 2; *P* < 0.0001).

During the mango off-season from 07th of March to 15th August 2009, the highest infestation levels was recorded on 15th of March 2009 (mean temp. of 27.5°C, mean RH of 75.7%, and 35.2 mm of rainfall) and 15th of August 2009 (mean temp. of 25.2°C, mean RH of 70.9%, and 0.7 mm of rainfall) on the leaves (47.34 ± 10.36 mealybugs/leaf) and twigs (52.15 ± 19.49 mealybugs/twig), respectively. However, during the mango off-season there was no significant difference in the infestation levels between twigs and leaves ($\chi 2$ = 0.147; *df* = 1; *P* = 0.7019).

During the 2009/10 mango fruiting season, the highest infestation levels on the leaves (88. 51 ± 13.65 mealybugs/leaf) and the fruit (178.4 ± 54.06 mealybug/fruit) was recorded on 10th of January 2010 (mean temp. 27.7°C, mean RH of 82%, and 23.9 mm of rainfall), while that on the twig (207.15 ± 54.55 mealybugs/twig) was on 18th January 2010 (mean temp. of 28.3°C, mean RH of 78.7%, and 16.3 mm of rainfall). On the other hand the lowest infestations was recorded on 15th of September 2009 (mean temp. of 25.0°C, mean RH of 65.6%, and 0.5 mm of rainfall for fruit (4.0 ± 2.67 mealybugs/fruit), and on 10th of December 2009 (mean temp. of 28.5°C, mean RH of 67.6%, and total rainfall of 114.9 mm) for both twigs (8.0 ± 2.96 mealybugs/twig) and leaves (11.49 ±2.92 mealybugs/leaf). Similar to the 2008/9 season, there was a significant difference in the infestation levels amongst the twigs, leaves and fruits ($\chi2$ = 123.4; df = 2; P < 0.0001).

During the mango off-season, the highest infestation levels was recorded on 27th of February 2010 (mean temp. of 28.9°C, mean RH of 69.4%, and 2.6 mm of rainfall) for both twigs (34.8 ± 10.46 mealybugs/twig) and leaves (41.79 ± 9.17 mealybugs/leaf). While the lowest infestation was on 15th of June 2010 (mean temp. of 25.6°C, mean RH of 76.6%, and 63.8 mm of rainfall) for both twigs (2.55 ± 1.42 mealybugs/twig) and leaves (4.69 ± 1.85 mealybugs/leaf). However, there was a significant difference on the infestation levels observed on the twig and leaves during this period ($\chi2 = 12.97$; $df = 1$; $P < 0.0003$).

7.3.3 Seasonal infestation levels by *R. iceryoides* on mango in Dar es Salaam

The population of *R. iceryoides* varied greatly with rainfall on all plant parts (Figure 7.3). During the 2008/09 mango fruiting season, the highest infestation levels by *R. iceryoides* was recorded on 03rd of January 2009 (mean temp. of 28.3°C, mean RH of 68.5%, and total rainfall of 0.2 mm) for the fruits (109.6 ± 45.60 mealybugs/fruit); while the infestation was highest on 11th of January 2009 (mean temp. of 27.9°C, mean RH of 66.6%, and no rainfall) for both twigs (148.55 ± 40.64 mealybugs/ twig) and leaves (82.55 ± 13.35 mealybug/leaf) (Figure 7.3). Thereafter, the infestation level continue to decline reaching the lowest level on 20th of February 2009 (mean temp. of 27.6°C, mean RH of 72.8%, and rainfall of 63.2 mm) on the twigs (10.25 ± 2.99 mealybugs/twig). Whereas, it was lowest on 08th March 2009 (mean temp. of 27.8°C, mean RH of 73.5%, and rainfall of 30.8 mm) on both leaves (10.89 ± 5.35 mealybugs/leaf) and fruits (4.6 ± 1.89 mealybugs/fruit) (Figure 7.3). During the 2008/09 fruiting season, *R. iceroides* infestation level varied significantly amongst the twigs, leaves and fruits ($\chi2 = 36.63$; $df = 2$; $P < 0.0001$).

During the mango off-season, the highest infestation levels was recorded on 16th of March 2009 (mean temp. of 27.7°C, mean RH of 79.9%, and 36.7 mm of rainfall) and on 24th of March 2009 (mean temp. of 28.4°C, mean RH of 68.4%, and no rainfall), on the leaves (13.48 ±

5.41 mealybugs/leaf) and the twigs (16.05 ± 6.55 mealybugs/twig), respectively. On the other hand the lowest infestation was on 16th July 2009 (mean temp. of 25.0°C, RH of 74.4%, and rainfall of 16.5 mm) and 16th October, 2009 (mean temp. of 26.3°C, RH of 64.7%, and rainfall of 0.5 mm) for the twigs (3.65 ± 1.27 mealybugs/twig) and the leaves (5.24 ± 1.38 mealybugs/leaf), respectively. However, during this period there was no significant difference in the infestation levels amongst the different plant parts ($\chi 2$ = 0.023; *df* = 1; *P* = 0.8802).

During the 2009/10 mango fruiting season, the highest infestation levels was recorded on 11th January 2010 (mean temp. of 27.4°C, mean RH of 81.5%, and 30.3 mm of rainfall) on the twigs (75.65 ± 21.22 mealybugs/twig). While it was highest on 27th of January 2010 (mean temp. 28.4°C, mean RH of 69.7%, and no rainfall) on the leaves (86.2 ± 14.05 mealybugs/leaf) and the fruits (82.6 ± 30.77 mealybugs/fruit). The infestation reached its minimum on 11th of December 2009 (mean temp. of 28.3°C, mean RH of 69.1, and total rainfall of 98.2 mm) on twigs (7.75 ± 3.61 mealybugs/twig) and the leaves (4.66 ± 2.04 mealybugs/leaf), and on 28th of February 2010 (mean temp. of 29.2°C, mean RH of 69.9%, and 6.1 mm of rainfall) on the fruits (5.2 ± 2.63 mealybugs/fruit) (Figure 3). There was a significant difference in the infestation levels amongst the twigs, leaves and fruits ($\chi 2$ = 11.89; *df* = 2; *P* = 0.0026).

During the mango off-season of 2010, the highest infestation levels was recorded on 08th of March 2010 (mean temp. of 29.1%, mean RH of 78.4%, and 61.7 mm of rainfall) on twigs (18.1 ± 5.41 mealybugs/twig) and the leaves (21.24 ± 7.79 mealybugs/leaf). Thereafter, there was a gradual decline of the infestation level with the minimum being recorded on 16th of June 2010 (mean temp. of 25.8°C, mean RH of 76.5%, and 63.8 mm of rainfall) for both twigs (1.55 ± 0.95 mealybugs/twig) and leaves (3.48 ± 1.73 mealybugs/leaf). However, there was a significant difference on the infestation levels observed on the twigs and leaves during this period ($\chi 2$ = 7.57; *df* = 1; *P* = 0.0059).

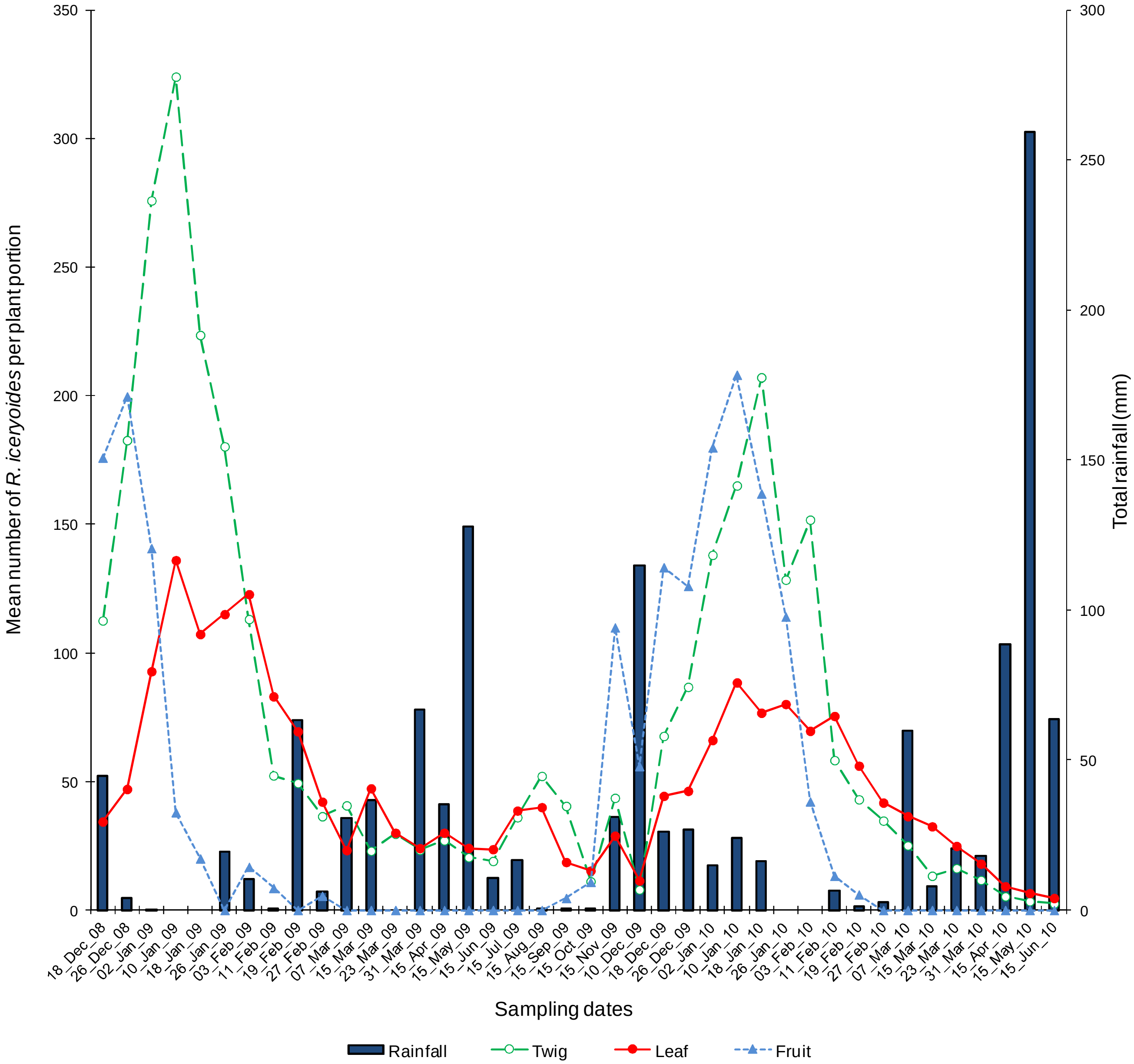

Figure 7. 2: Seasonal fluctuation of *R. iceryoides* on the twigs, leaves and fruit with corresponding rainfall (mm) from December 2008 to June 2010 in Kibaha

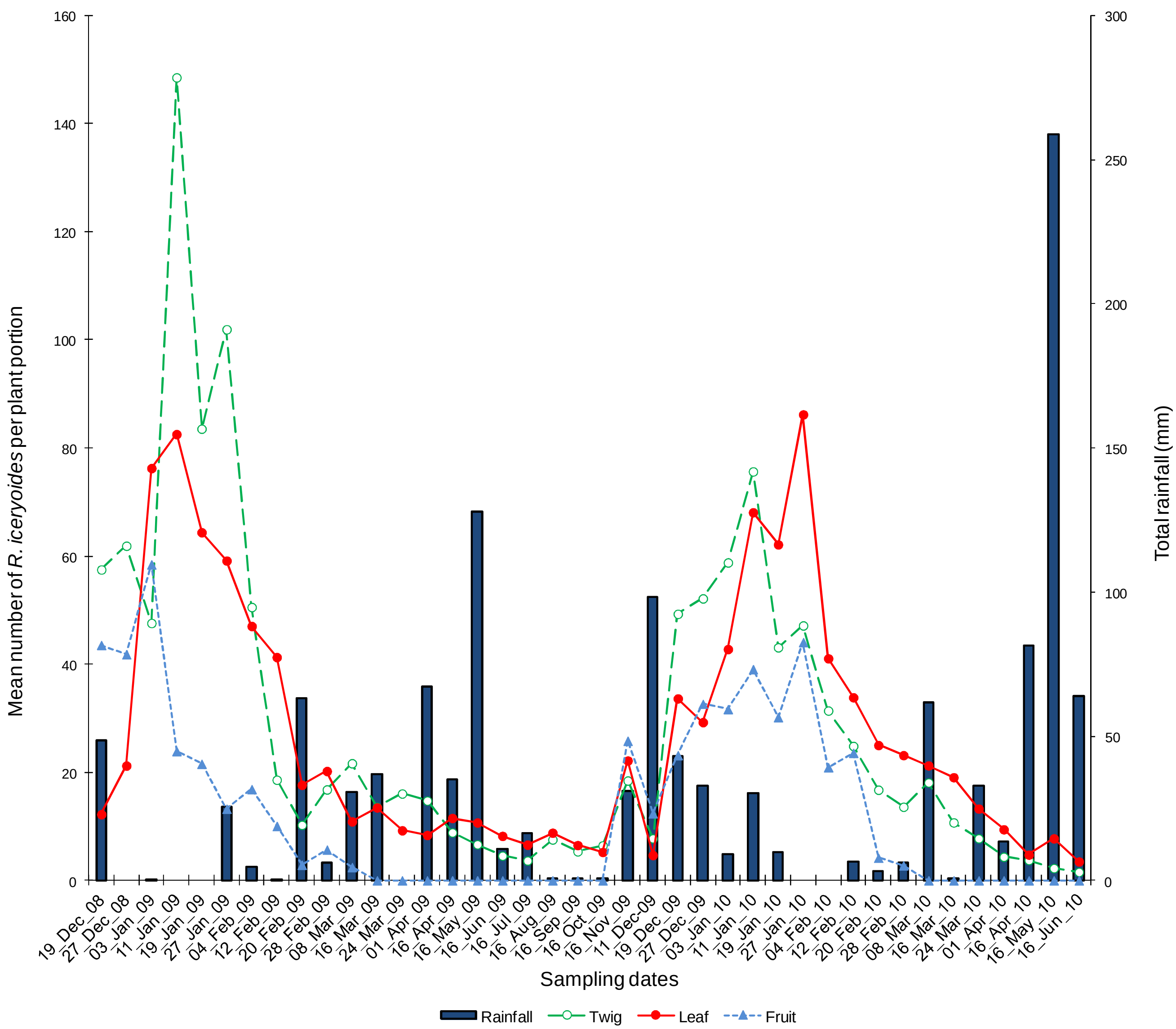

Figure 7. 3: Seasonal fluctuation of *R. iceryoides* on the twigs, leaves and fruit with corresponding rainfall (mm) from December 2008 to June 2010 in Dar es Salaam

7.3.4 Seasonal fluctuation of percentage parasitism of *R. iceryoides* in Kibaha

A total of 3421, 10357 and 261 mummified mealybugs, were recorded from the twigs, leaves and fruits, respectively, with respective mean percentage parasitism of 5.54 ± 0.18, 6.52 ± 0.12 and 3.44 ± 0.41%. The seasonal fluctuation trend in percentage parasitism did not follow that of the host (Figure 7.4). Percent parasitism was quite low at the beginning of the season with least percent parasitism being recorded on18th of December 2008 (mean temp. of 27.6°C, mean RH of 73.8%, and rainfall of 44.6 mm), for twigs (0.75±0.28%) and leaves (0.61± 0.18). While the least percent parasitism on the fruit (0.0 %) was recorded on 26th of January 2009 (mean temp. of 28.2°C, mean RH of 72.3%, and rain of 19.5 mm), and 19th of February, 2009 (mean temp. of 27.7 °C, mean RH of 70.2%, and rain of 63.2 mm). As the season advanced the percent parasitism increased slightly to reached its maximum on 18th of January 2009 (mean temp. of 28.4°C, mean RH of 66.9%, and zero rainfall), 3rd of February 2009 (mean temp. of 28.4°C, mean RH of 77.9%, and rainfall of 0.4 mm), and 11th of February 2009 (mean temp. of 26.6°C, mean RH of 70.19%, and rainfall of 63.2 mm), for the fruit (6.2±3.3%), twigs (4.4±1.3%) and leaves (3.1±1.0%) respectively. The percentage parasitism varied greatly ($\chi 2$ = 33.58; df = 2; $P <$ 0.0001) amongst the different plant parts.

During the mango off-season period of 2009, the lowest percentage parasitism was recorded on 7th of March 2009 (mean temp of 27.8°C, RH of 71.8%, and rainfall of 30.8 mm), and 15th July 2009 (mean temp of 24.9°C, RH of 75.1%, and rainfall of 16.5 mm) for the leaves (0.31 ± 0.14%) and the twigs (0.67 ± 0.4), respectively. While the highest percent parasitism was recorded on 15th March, 2009 (mean temp. 27.7°C of, RH of 80.9%, and rainfall of 36.7 mm) and 15th July 2009, for twigs (2.49 ± 1.09%) and the leaves (1.95 ± 0.48%), respectively. However, during the mango off season there was no significant difference in the percentage parasitism between the twigs and leaves ($\chi 2$ = 0.36; df = 1; P = 0.5485).

During the 2009/2010 mango fruiting season, the lowest percent parasitism was recorded on 15th September, 2009 (mean temp. of 25.0°C, RH of 65.56%, and rainfall of 0.5 mm) and 26th December 2009 (mean temp. of 28.2°C, RH of 72.7%, and rainfall of 26.9 mm) for leaves (0.75± 0.26%) and twigs (0.89 ± 0.4%), respectively. However on the fruit no mummified *R. iceryoides* was recorded on 15th September, 2009 as well as on 19th February, 2010 (mean temp. of 29.3°C, RH of 70.3%, and rainfall of 1.2 mm). With the advancement of the mango season the percent parasitism increased to reach its maximum on 26th January, 2010 (mean temp. of 28.8°C, RH of 69.5%, and zero rainfall) for the fruits (4.39 ± 1.56%) and on 03rd February, 2010 (mean temp. of 28.7°C, RH of 68.9%, and zero rainfall), for both the twigs (4.52 ± 1.59%) and the leaves (4.57 ± 0.98%). . There was a significant difference in the percentage parasitism amongst the twigs, leaves and fruits ($\chi 2$ = 17.81; df = 2; P = 0.0001).

During the mango off-season 2010, the highest percent parasitism was recorded on 07th of March, 2010 (mean temp. of 29.4°C, RH of 79.4%, and rainfall of 59.9 mm) and 15th of March, 2010 (mean temp. of 28.5°C, RH of 67.4%, and rainfall of 8 mm) for twigs (5.64 ± 2.4), and leaves (1.99 ± 0.51), respectively. Thereafter, the percent parasitism started to decline to reach its minimum on 15th of June 2010 (mean temp. of 25.5°C, RH of 76.6%, and rainfall of 63.8 mm) for both twigs (0.2 ± 0.2%) and the leaves (0.45± 0.19%). Unlike that of the mango fruiting season, percent parasitism during this season was not influenced by the plant part from which the mummies were collected ($\chi2$ =0.39; *df* = 1; *P* = 0.5316).

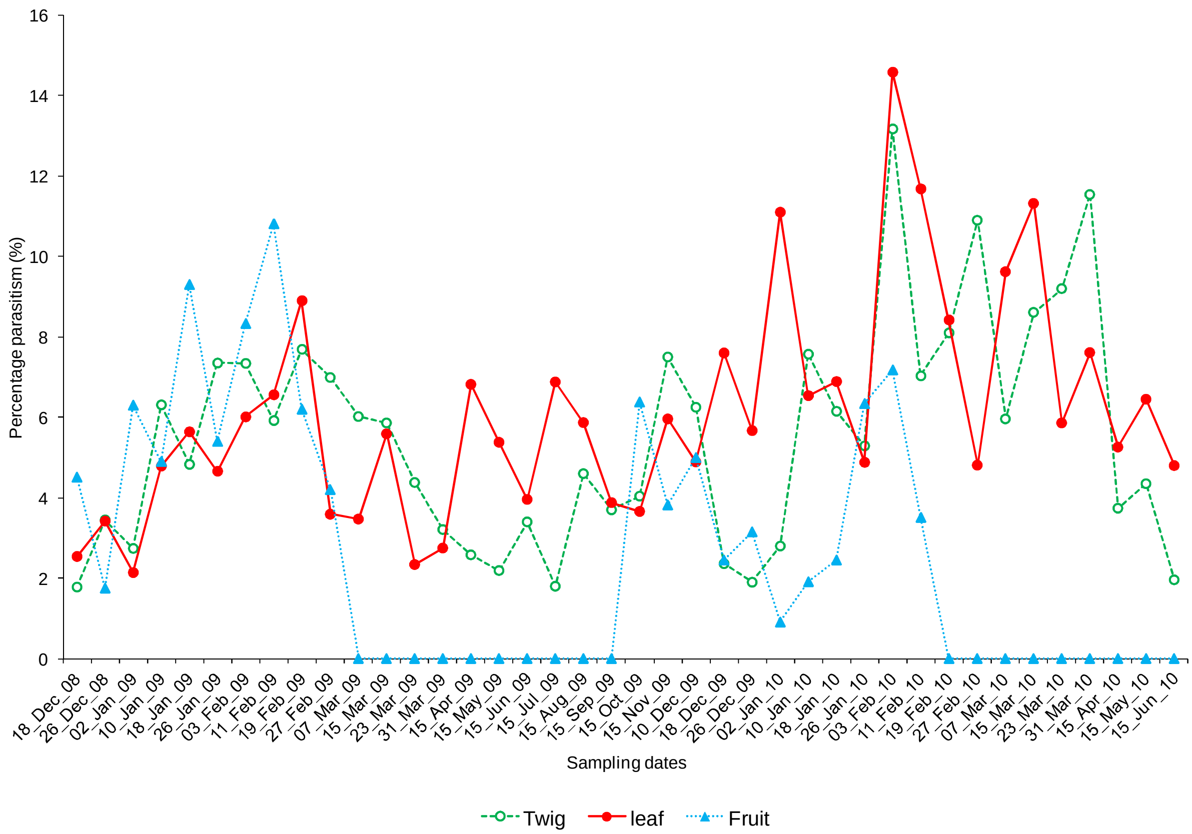

Figure 7. 4: Seasonal variation of percentage parasitism of *R. iceryoides* on twigs, leaves and fruits in Kibaha from December 2008 to June 2010.

7.3.5 Seasonal fluctuation of percentage parasitism of *R. iceryoides* in Dar es Salaam

A total of 1537, 6239 and 244 mummified mealybugs, were recorded on the twigs, leaves and fruits, respectively, with respective mean percentage parasitism of 6.44 ± 0.31, 7.14 ± 0.17 and 4.90 ± 0.6%. Like that in Kibaha, the seasonal fluctuation trend in percentage parasitism did not follow that of the host (Figure 7.5). Percent parasitism was very low at the beginning of the mango fruiting season with least percent parasitism being recorded on19th of December 2008 (mean temp of 27.6°C, mean RH of 73.9%, and rainfall of 48.6 mm), for leaves (1.13 ± 0.4%). While on the 27th December 2008 (mean temp of 27.9°C, mean RH of 67.8%, and zero rainfall), it was lowest on both twigs (1.16 ± 0.47%) and fruits (0.85 ± 0.69%). The percent parasitism increased, thereafter to record its maximum on 12th February 2009 (mean temp of 28.5 °C, mean RH of 67.9%, and rainfall of 0.4 mm), on 28th February 2009 (mean temp of 27.6 °C, mean RH of 74.1%, and rainfall of 6.1mm), and 08th March, 2009 (mean temp of 27.8 °C, mean RH of 73.5%, and rainfall of 30.8 mm) on fruits (5.78 ± 3.44%), leaves (3.91 ± 0.99%) and twigs (6.71 ± 2.24%), respectively. There was no significant difference in the percentage parasitism amongst the twigs, leaves and fruits ($\chi2$ = 4.90; *df* = 2; *P* = 0.0862).

During the mango off-season of 2009, the highest percentage parasitism was recorded on 16th of March, 2009 (mean temp of 27.7°C, mean RH of 79.9%, and rainfall of 36.7mm), and 24th March 2009 (mean temp of 28.4°C, mean RH of 68.4%, and zero rainfall) for the twigs (4.99 ± 2.04%) and the leaves (2.24 ± 0.59%), respectively. While the lowest percent parasitism was recorded on 16th June 2009 (mean temp of 25.9°C, mean RH of 74.9% , and rainfall of 10.8 mm) and 16th July 2009 (mean temp of 25.0°C, mean RH of 74.4% and rainfall of 16.5 mm), for the twigs (0.58 ± 0.41%) and the leaves (0.60 ± 0.24%), respectively. There was no significant difference in the percentage parasitism recorded on the twigs and leaves ($\chi2$ = 0.09; *df* = 1; *P* = 0.7630).

During the 2009/2010 mango fruiting season, the lowest percentage parasitism of *R. iceryoides* was recorded on 11th December 2009 (mean temp of 28.3°C, mean RH of 69.1% and rainfall of 98.2 mm), 19th December 2009 (mean temp of 28.5°C, mean RH of 75.1% and rainfall of 43.0 mm), and 27th December 2009 (mean temp of 28.0°C, mean RH of 73.8%, and rainfall of 32.8 mm) for leaves (0.37 ± 0.18%), fruits (0.84 ± 0.53%), and twigs (1.30 ± 0.59%), respectively. Thereafter, the percent parasitism increased with the maximum recorded on 27th January 2010 (mean temp of 24.2°C, mean RH of 69.7%, and rainfall of 28.4mm), 04th February 2010 (mean temp of 25.1°C, mean RH of 68.8%, and rainfall of 28.9 mm) and 20th of February 2010 (mean temp of 24.8°C, mean RH of 71.2%, and rainfall of 29.2 mm) on leaves (3.81 ± 0.63%), fruits (5.05 ± 2.28%), and twigs (5.08 ± 2.17%), respectively. During this period the parasitism was comparable amongst the different plant parts ($\chi2$ = 0.53; *df* = 2; *P* = 0.7665).

During the mango off-season of 2009/10, the highest percentage parasitism of *R. iceryoides* was recorded on 16th March 2010 (mean temp of 29.5°C, mean RH of 66.8%, and rainfall of 0.7 mm), and 24th March 2010 (mean temp of 28.1°C, mean RH of 80.2%, and rainfall of 32.9 mm) for twigs (3.64 ± 1.72%) and leaves (1.30 ± 0.43%), respectively. Thereafter the percent parasitism declined with a minimum recorded on the twigs (0.60 ± 0.45%) and leaves (0.19 ± 0.1%) on 16th May 2010 (mean temp of 26.6°C, mean RH of 83.9%, and rainfall of 258.7 mm) and 16th June 2010 (mean temp of 25.8°C, mean RH of 76.5% and rainfall of 63.8 mm), respectively.

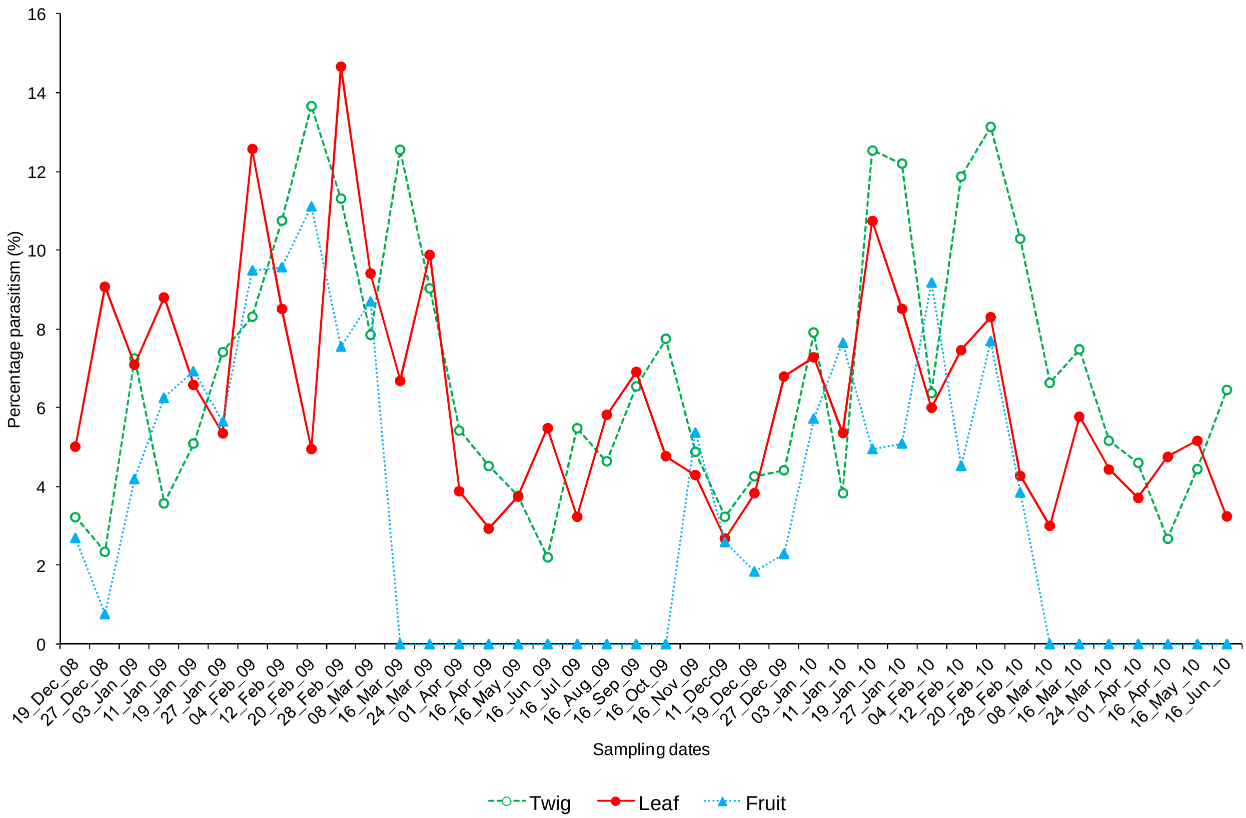

Figure 7. 5: Seasonal variation of percentage parasitism of *R. iceryoides* on twigs, leaves and fruits in Dar es Salaam from December 2008 to June 2010.

7.3.6 Seasonal population fluctuation of primary parasitoid species

In Kibaha, out of 14039 mummified mealybugs collected from the twigs, leaves and fruits during the study period, 7960 wasps emerged. Six primary parasitoid species were identified; accounting for 76.31% of total emergence. All the primary hymenopteran parasitoids were members of the family Encyrtidae. These were namely, *Anagyrus pseudococci* (Girault) accounting for 90.75%, followed by *Leptomastrix dactylopii* Howard (4.61%), *Anagyrus aegyptiacus* Moursi (1.84%), *Leptomastidea tecta* Prinsloo (1.14%), *Agarwalencyrtus citri* Agarwal (1.05%) and *Aenasius longiscapus* Compere (0.61%) (Figure 7. 6). The combined percentage parasitism by these parasitoids ranged from 0.13% on the 15th of June 2009 to 5.2% on the 07th of March 2009. There was a strong and positive association between the parasitoid population and that of their host, *R. iceryoides* on all plant parts (r = 0.91, P < 0.0001; r = 0.87, P < 0.0001; r = 0.94, P < 0.0001 for twigs, leaves, and fruits, respectively).

The highest number of the dominant parasitoid, *A. pseudococci* recovered from the collected mummies were 463 and 406 wasps for 2008/09 (on 10th January 2009, mean temp. of 27.9°C, mean RH of 66% and rainfall of 0 mm) and 2009/10 (on 10th January 2010, mean temp. of 27.7°C, mean RH of 82% and total rainfall of 23.9 mm) mango fruiting seasons, respectively (Figure 7. 6).

In Dar es Salaam, out of 8020 mummified mealybugs collected from the twigs, leaves and fruits during the study period, 3569 wasps emerged. The primary parasitoids accounted for 80.3% of the total emergence, while the remaining 19.7% were hyperparasitoids. The primary parasitoid species were *A. pseudococci* (94.04%), *L. dactylopii* (3.91%), *A. aegyptiacus* (1.19%), *L. tecta* (0.66%) and *A. longiscapus* (0.2%). The percentage parasitism throughout the entire study ranged from 0.47% on the 11th of December 2009 to 6.7% on the 19th of December 2008 (Figure 7.7). The population of the parasitoid was significantly and positively correlated with that of its host, *R. iceryoides* on all plant parts (r = 0.89, P < 0.0001; r = 0.95, P < 0.0001; r = 0.94, P < 0.0001 for twigs, leaves, and fruits, respectively).

The highest number of the dominant parasitoid, *A. pseudococci* was collected on the 19th of January 2009 (n = 274) at mean temperature of 28.5°C, mean RH of 67.01% and no rainfall; and on the 11th of January 2010 (n = 269) at mean temperature of 27.4°C, mean RH of 81.5% and total rainfall of 30.3 mm (Figure 7. 7).

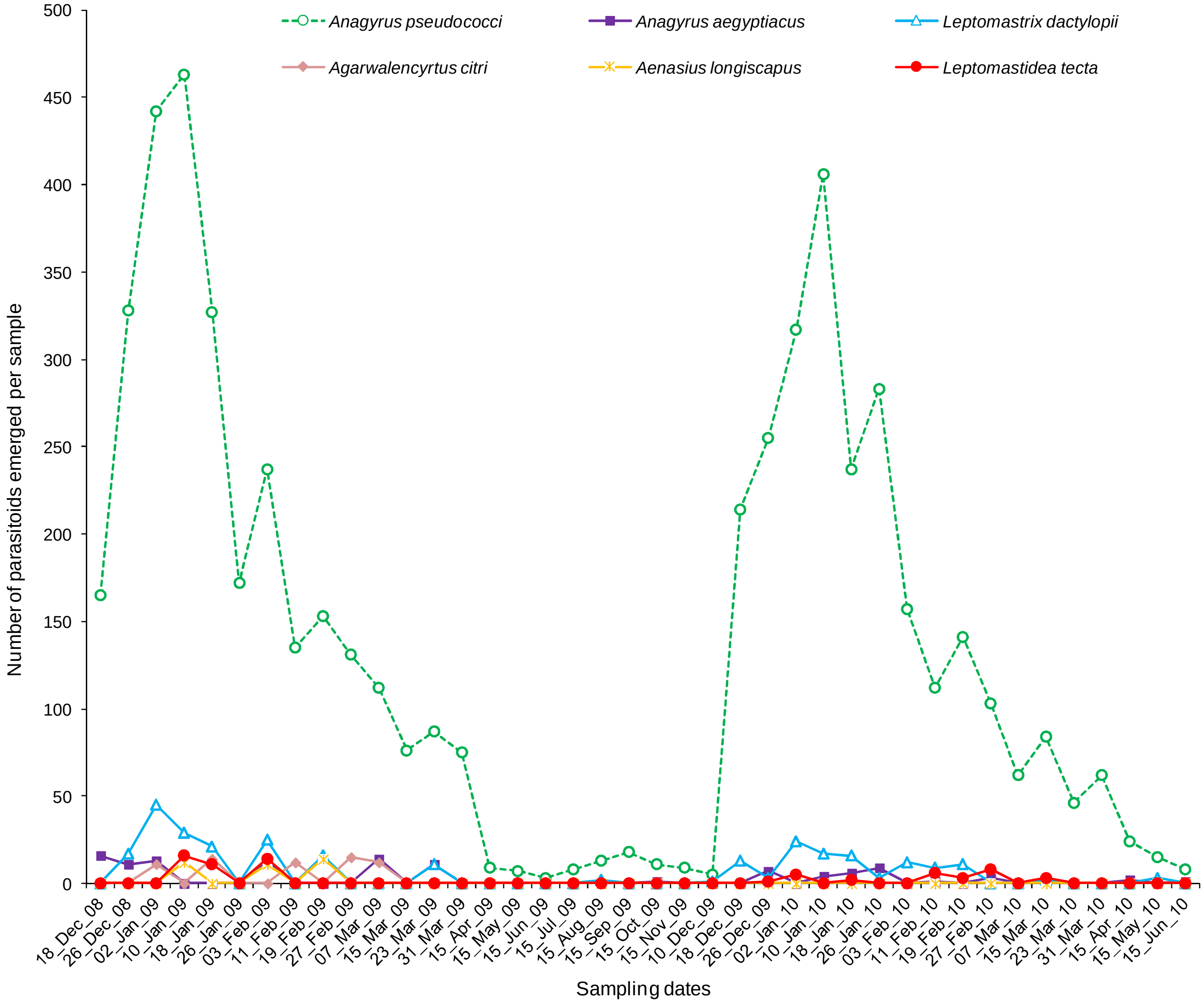

Figure 7. 6: Seasonal population variation of the primary parasitoid species from December 2008 to June 2010 in Kibaha.

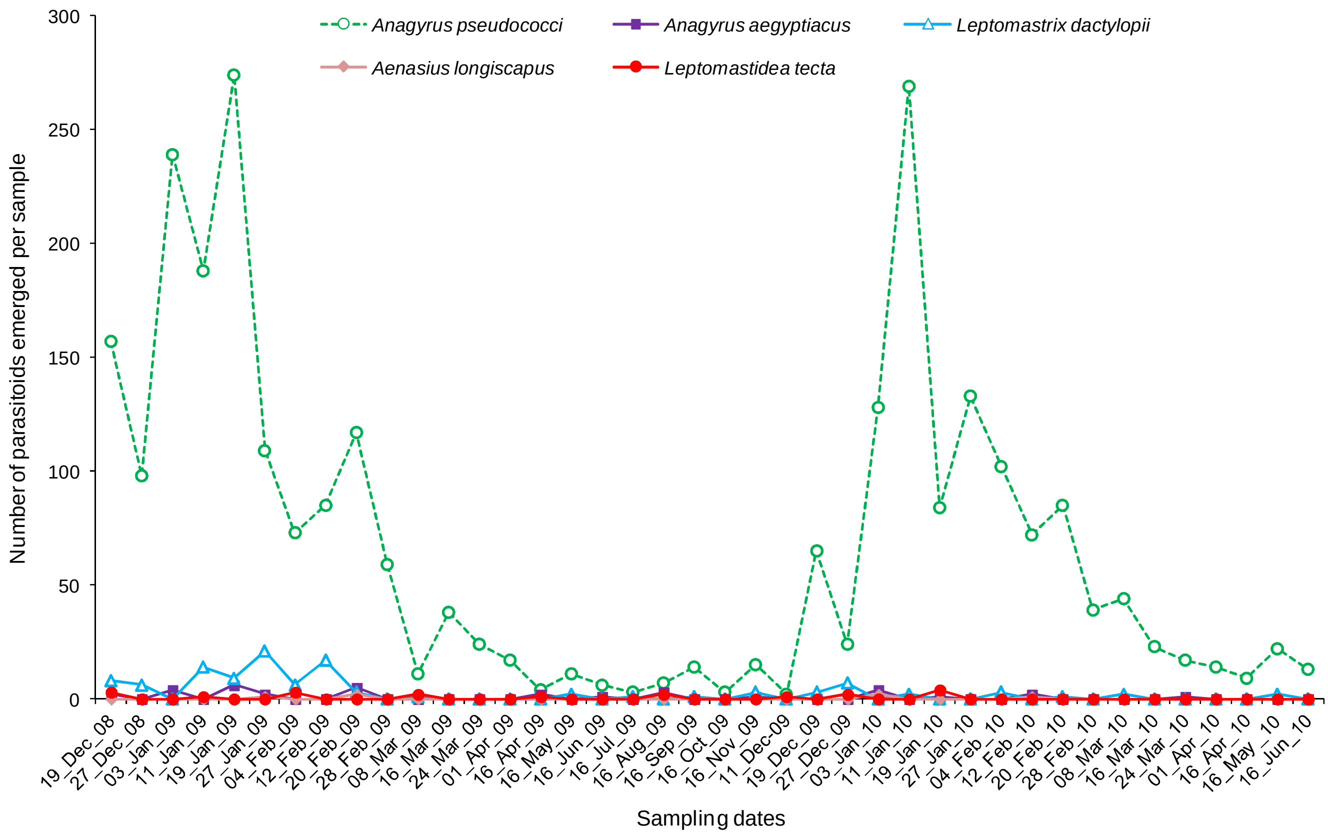

Figure 7. 7: Seasonal population variation of the primary parasitoid species from December 2008 to June 2010 in Dar es Salaam.

7.3.7 Seasonal population fluctuation of different predator species

In Kibaha, a total of nineteen species of predators were found preying on *R. iceryoides* throughout the study period. The major predators were six species of Coccinelidae: *Cryptolaemus montrouzieri* Mulsant (1.42%), *Hyperaspis amurensis* Weise (1.27%), *Hyperaspis bigeminata* Randall (12.11%), *Exochomus nigromaculatus* Goeze (3.47%), *Chilocorus renipustulatus* Scriba (15.72%), *Chilocorus nigrita* Fabricius (31.23%); one Lycaenidae (*Spalgis lemolea* Druce) (9.07%); one Drosophilidae (*Cacoxenus perspicax* Knab) (20.89%). Other minor predators included *Pyroderces badia* Hodges (Lepidoptera: Cosmopterigidae), *Hemerobius* sp. (Neuroptera: Hemerobiidae), *Cheiracanthium virescens* Sundevall (Arachnida: Clubionidae) and the rest were other coccinelids: *Rodolia limbata* Motschulsky, *Rodolia pumila* Weise, *Micraspis* sp., *Propylea dissecta* Mulsant, *Propylea 14-punctata* Schachbrett-Marienkäfer, *Telsimia nitida* Chapin, *Harmonia dimidiata* Fabricius and *Hyperaspis* sp. The population of the major predators was found to increase with the population build-up of *R. iceryoides*. The highest level of predator activity was recorded between December and February for each year (Figure 7. 8).

In Dar es Salaam, a total of 11 species of predators were recorded. In the order of their importance they were *C. nigrita* (30.32%), *H. bigeminata* (18.47%), *C. renipustulatus* (15.65%), *C. perspicax* (15.08%), *E. nigromaculatus*, *C. montrouzieri*, *H. amurensis*, *R. pumila*, *S. lemolea*, *Hyperaspis* sp. and *R. limbata*. Like in Kibaha, the population of the major predators increased in a density dependant manner together with that of their prey, *R. iceryoides*. The highest level of activities by the predators was observed between December and February for both years (Figure 7. 9).

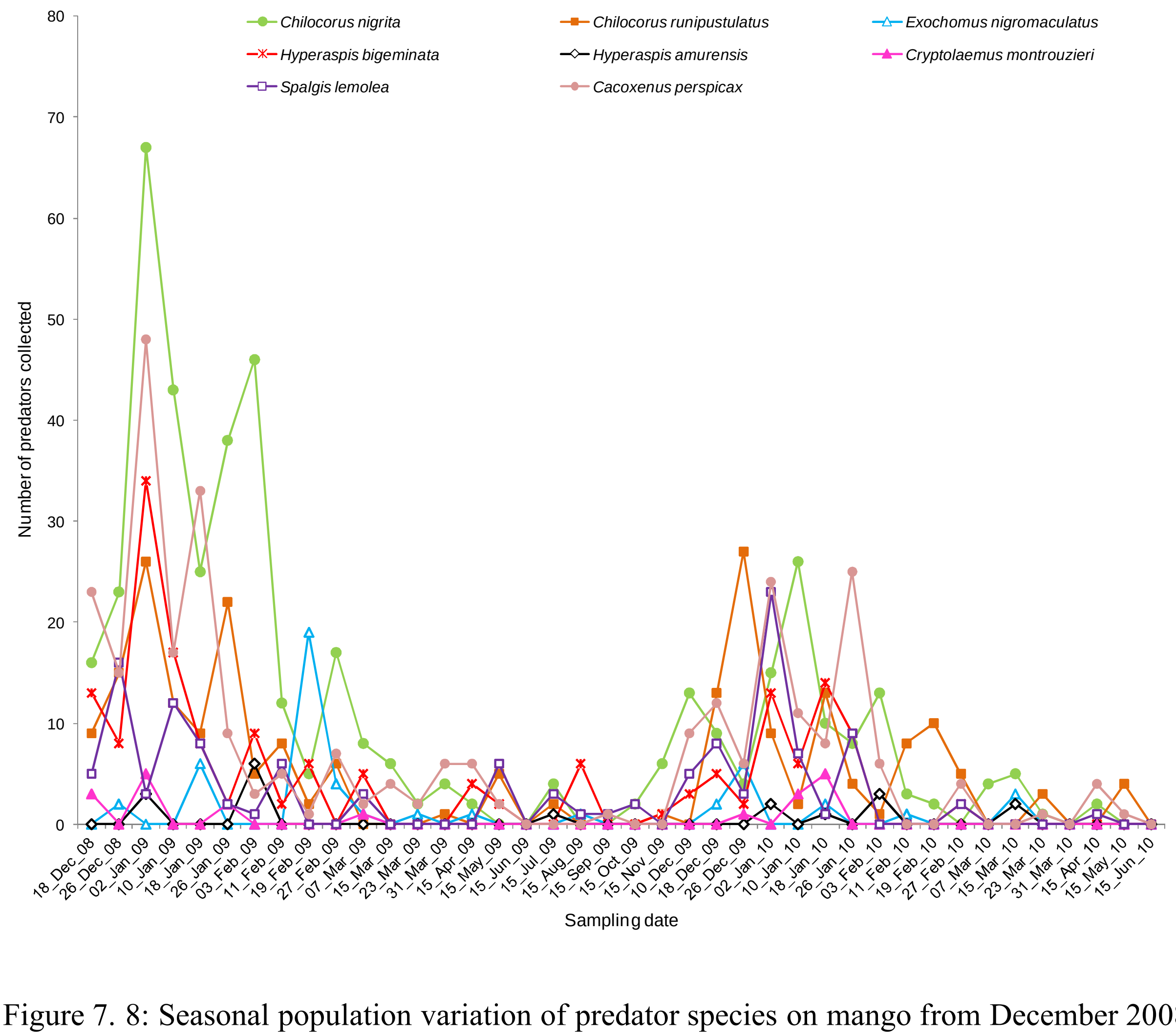

Figure 7. 8: Seasonal population variation of predator species on mango from December 2008 to June 2010 in Kibaha.

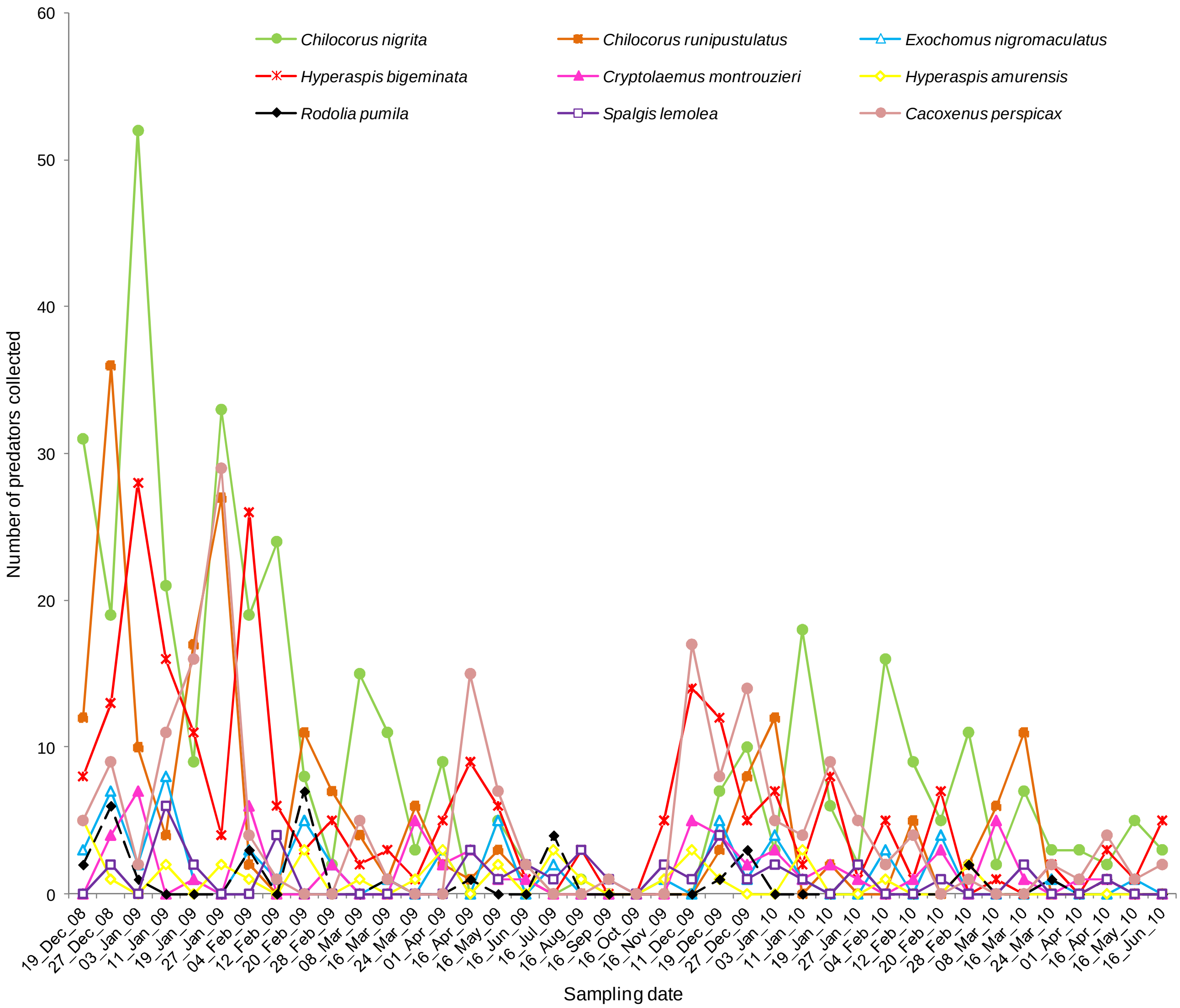

Figure 7. 9: Seasonal population variation of predator species on mango from December 2008 to June 2010 in Dar es Salaam.

7.3.8 Seasonal fluctuation of hyperparasitoid species population

In Kibaha, a total of 14 species of hyperparasitoids were found parasitizing the primary parasitoids of *R. iceryoides*. These hyperparasitoids belong to four families, namely Encrytidae (*Achrysopophagus aegyptiacus* Mercet, *Cheiloneurus carinatus* sp.nov, *Cheiloneurus angustifrons* sp.nov, *Cheiloneurus cyanonotus* Waterston and *Cheiloneurus latiscapus* Girault), Aphelinidae (*Coccophagus gilvus* Hayat, *Coccophagus pseudococci* Compere, *Coccophagus* *bivittatus* Compere, *Marietta leopardina* Motschulsky, *Coccophagus lycimnia* Walker and *Coccophagus nigricorpus* Shafee), Signiphoridae (*Chartocerus conjugalis* Mercet and *Chartocerus* sp.) and Eulophidae (*Tetrastichus* sp.). The most abundant hyperparasitoids were *C. conjugalis* (8.72%), *C. carinatus* (4.37%), *M. leopardina* (2.21%), *C. cyanonotus* (1.37%) and *C. latiscapus* (1.32%). The remaining nine hyperparasitoid species accounted for only 6% of the total wasps recovered. The highest number (n = 64) of the key hyperparasitoid, *C. conjugalis* was recorded on the 10th of January 2009 (mean temp. of 27.9°C, mean RH of 66%, and no rainfall) (Figure 7. 10). The number of the hyperparasitoid was positively correlated with that of the primary parasitoid ($r = 0.8343$; $P < 0.0001$).

In the Dar es Salaam study site, a total of 12 species of hyperparasitoids were recovered from the *R. iceryoides* mummies. These were from three families, Encrytidae (*A. aegyptiacus*, *C. carinatus*, *C. angustifrons*, *C. cyanonotus* and *C. latiscapus*); Aphelinidae (*C. gilvus*, *C. pseudococci*, *C. bivittatus*, *M. leopardina*, *C. lycimnia* and *C. nigricorpus*) and Signiphoridae (*C. conjugalis*). The most abundant hyperparasitoids were *C. conjugalis* (9.39%), followed by *C. carinatus* (2.66%), *M. leopardina* (2.63%), *C. pseudococci* (0.92%) and *C. angustifrons* (1.01%). The remaining seven hyperparasitoid species accounted for 3% of the total wasps recovered. The highest number of the dominant species, *C. conjugalis* (n = 46) was recorded on the 03rd of January 2009 (mean temp.of 28.3°C, mean RH of 68.5% and total rainfall of 0.2 mm (Figure 7. 11). Similar to the Kibaha scenario, the number of the hyperparasitoids were found to positively correlated with that of the primary parasitoid ($r = 0.6654$; $P < 0.0001$).

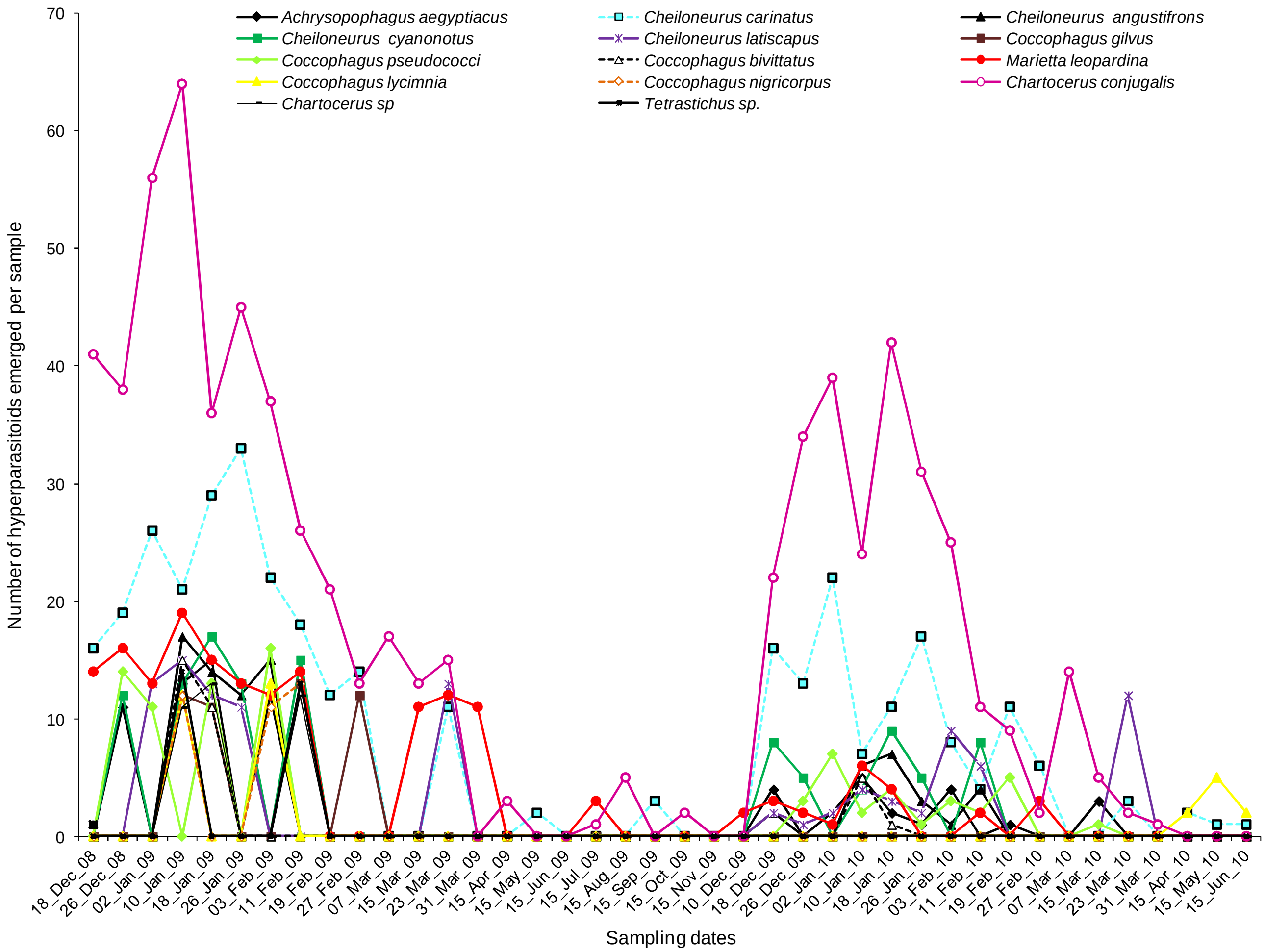

Figure 7. 10: Relative abundance of hyperparasitoids during December 2008 to June 2010 survey in Kibaha.

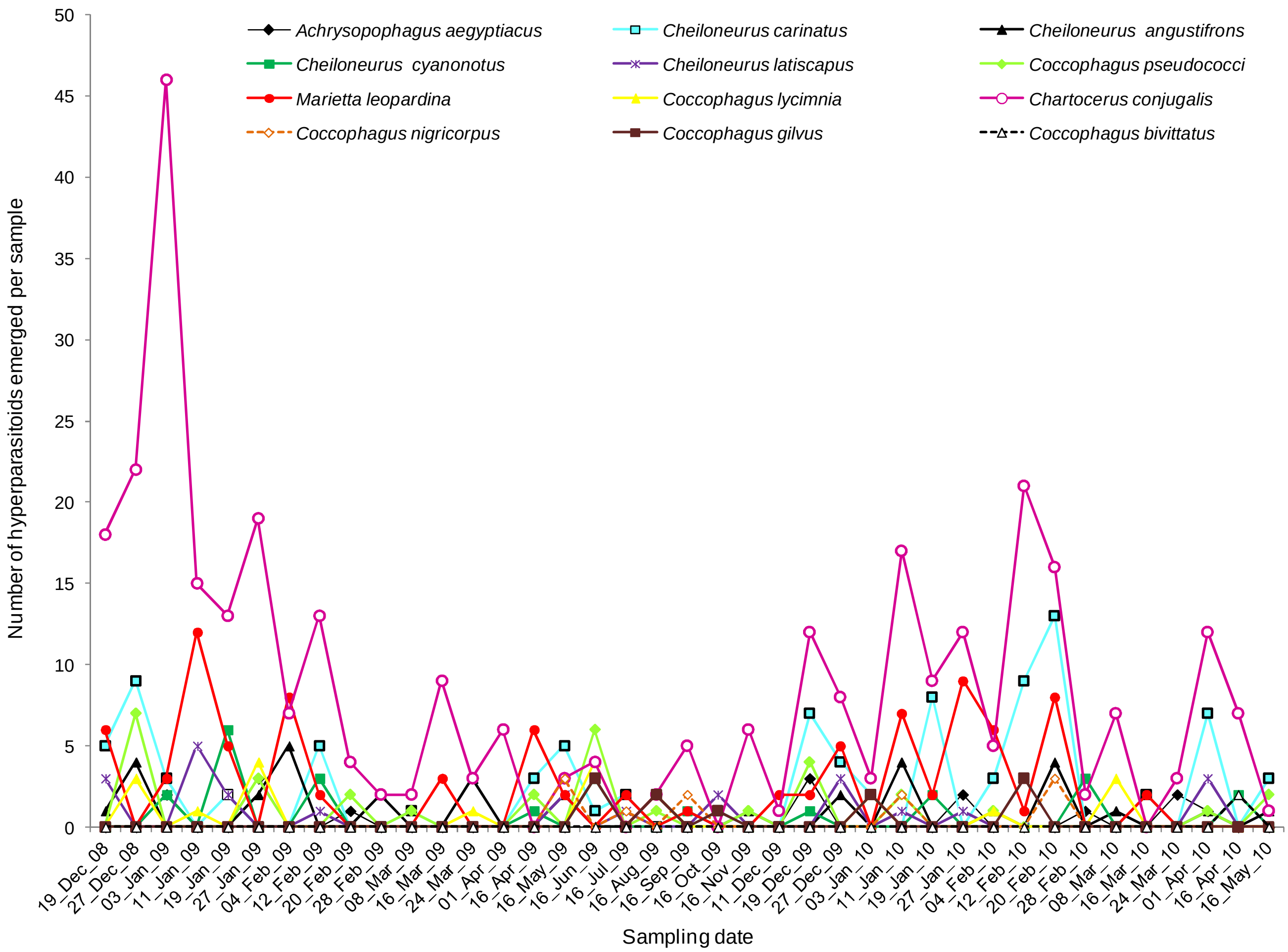

Figure 7. 11: Relative abundance of hyperparasitoids during December 2008 to June 2010 survey in Dar es Salaam.

7.3.10 Effect of weather variables on the infestation level and the number of mummified mealybugs on the different plant parts in Kibaha

The population dynamics of *R. iceryoides* on twigs and leaves was significantly and positively affected by the mean temperature (*Wald* = 4.21; *P* = 0.0402 and *Wald* = 15.33; *P* < 0.0001, for twigs and leaves, respectively). However, on the fruit the mealybug population dynamics was not affected by the temperature (*Wald* = 0.02; *P* = 0.8816). Similarly, rainfall had significant but negative effect on the population dynamics of *R. iceryoides* on twigs (*Wald* = 11.99; *P* = 0.0005) and leaves (*Wald* = 9.32; *P* = 0.0023), whereas the population dynamics on the fruit was not influenced by rainfall (*Wald* = 0.16; *P* = 0.6913). On the other hand mean relative humidity had no effect on population dynamics of the pest on the twigs (Wald = 0.01; P = 0.9218) and the leaves (*Wald* = 0.11; *P* = 0.7429). While, the pest population on the fruit was significantly and positively affected by the mean relative humidity (*Wald* = 11.33; *P* = 0.0008).

Like that of the host, the population dynamics of the parasitoids was significantly and positively influenced by the temperature on both twig (*Wald* = 9.99; *P* = 0.0016) and leaves (*Wald* = 11.02; *P* = 0.0009), while temperature had no effect on parasitoid population dynamics on fruit (*Wald* = 1.12; *P* = 0.2900). Likewise, rainfall had significant but negative effect on the population dynamics on the parasitoid on twigs (*Wald* = 11.57; *P* = 0.0007) and leaves (*Wald* = 4.48; *P* = 0.0342). However, the population dynamics of the parasitoid on the fruit was not influenced by rainfall (*Wald* = 0.07; *P* = 0.7900). On the other had mean relative humidity had no effect on population dynamics of the parasitoids on all plant part assessed (*Wald* = 0.07; *P* = 0.7893, *Wald* = 1.03; *P* = 0.3091 and *Wald* = 1.67; *P* = 0.2000 for twigs, leaves and fruit, respectively).

7.3.11 Effect of weather variables on the infestation level and the number of mummified mealybugs on the different plant parts in Dar es Salaam

Similar to that in Kibaha, the population dynamics of *R. iceryoides* on twigs and leaves was significantly and positively affected by the mean temperature (*Wald* = 4.81; *P* = 0.0280 and *Wald* = 10.31; *P* = 0.0013, for twigs and leaves, respectively). However, on the fruit the mealybug population dynamics was not affected by the temperature (*Wald* = 3.68; *P* = 0.0550). Also rainfall had significant but negative effect on the population dynamics of *R. iceryoides* on twigs (*Wald* = 5.51; *P* = 0.0190) and leaves (*Wald* = 5.51; *P* = 0.0189), while, the population dynamics on the fruit was not influenced by rainfall (*Wald* = 2.46; *P* = 0.1170). Mean relative humidity had no effect on population dynamics of the pest on the twigs, leaves and fruit (*Wald* = 0.01; *P* = 0.9210, *Wald* = 0.10; *P* = 0.7490, and *Wald* = 0.01; *P* = 0.9370, respectively).

The population dynamics of the parasitoids, like that of its host was also significantly and positively affected by the temperature on twig (*Wald* = 8.95; *P* = 0.0028) and leaves (*Wald* = 5.67; *P* = 0.0173), while temperature had no effect on parasitoid population dynamics on fruit (*Wald* = 1.19; *P* = 0.2762). Rainfall had a significant but negative effect on the population dynamics of the parasitoid on the twigs (*Wald* = 8.53; *P* = 0.0035), leaves (*Wald* = 8.49; *P* =

0.0035), and fruits (*Wald* = 10.14; *P* = 0.0015). Similar to that in Kibaha, relative humidity had no effect on population dynamics of the parasitoids on all plant part assessed (*Wald* = 0.75; *P* = 0.3851, *Wald* = 0.49; *P* = 0.4843 and *Wald* = 0.88; *P* = 0.3475, for twigs, leaves and fruits, respectively).

7.4 Discussion

The results of this study demonstrated that there was a clear and distinct seasonal pattern in population dynamics of the mango mealybug at the coastal area of Tanzania. The seasonal and annual fluctuations of *R. iceryoides* were very closely associated with the mango fruiting season. The population of *R. iceryoides* was found to be very low at the beginning of the mango season. Subsequently, the population built up as the mango season advanced, reaching its peak towards the mango harvesting season, which coincided with the dry season (northeast monsoon). Thereafter, the population declined to its lowest during the long rains (March to May), and cold and dry southwest monsoon (June-October). Boavida and Neuenschwander (1995) also reported a similar population dynamics pattern in relation to the rainfall for the related mealybug *R. invadens* in Benin. Further, the cassava mealybug, *Phenacoccus manihoti* Matile-Ferrero (Homoptera: Pseudococcidae), was also found to increase ten-fold during the dry season compared to the rainy season (Le Rü et al., 1991). Calatayud et al. (1994), attributed the increase of *P. manihoti* populations during the dry season to changes in the levels of secondary compounds which enhanced the plant resistance and that had a positive influence on the population dynamics of the pest. This argument may also explain the high mango infestation with *R. iceryoides* during the dry season, although, no chemical analysis was carried out to confirm this. However, in general, water stressed plants were more susceptible to mealybug infestations (Fabres and Le Rü, 1988; Gutierrez et al., 1993; Koricheva et al., 1998; Lunderstadt, 1998; Calatayud et al., 2000; Calatayud et al., 2002; Shrewsbury et al., 2004).

This study revealed that rainfall was significantly and negatively correlated, while temperature was significantly and positively correlated with *R. iceryoides* populations as well as its associated natural enemies on the various plant parts. Similar trends of association between these weather factors and *R. iceryoides* populations were reported by Suresh and Kavitha (2008). The authors found that relatively high temperatures and lack of rainfall were associated with an increase in mealybug populations. They also reported that for every unit increase in relative humidity and rainfall, there was a 0.05 unit population reduction in the *R. iceryoides* population on mango, in India. On the other hand, every unit of sunshine hours increased the population by 3.93 units. Heavy rains were observed to wash mealybugs off the mango plants down to the soil surface and led to considerable mealybug mortality; being one of the principal causes of the sharp decline of *R. iceryoides* population in that study. Similar findings were reported for other mealybug species such *Maconellicoccus hirsutus* (Green) (Mukherjee, 1919; Sriharan et al., 1979; Shree and Boraiah, 1988) and *Phenacoccus solenopsis* Tinsley (Suresh and Kavitha, 2008, Dhawan et al., 2009). In this study, besides rainfall, other factors such as availability of suitable host plants, and the action of natural enemies may have contributed to the population fluctuations of *R. iceryoides*. These factors were reported to have strong influences on the population dynamics of the congeneric pest, *R. invadens* (Pitan, 2000; Boavida and Neuenschwander, 1995). Besides its influence on the population dynamics, through washing down of the adults and crawlers, rainfall also promoted mango vegetative growth, which promoted new colonization sites for subsequent mealybug generations (Singh, 1968; Whiley, 1993; Boavida and Neuenschwander, 1995). For example, Boavida and Neuenschwander (1995) found that during flushing, large populations of young mealybug females, moved along different branches from older leaves or shoots to newer ones. This dispersal behaviour of mealybugs may explain the distinct mealybug population dynamics on the different plant parts (i.e., twigs, leaves and fruits). The highest mealybug infestation levels on the twigs and leaves was recorded on the 10th of January 2009 at a mean temperature of 27.97°C, mean relative humidity of 66% and no rainfall. This temperature has been reported to be within the optimal range for rapid proliferation of the population of the mealybug, *Paracoccus marginatus* Williams and Granara de Willink (Hemiptera: Pseudococcidae) (Chong et al., 2008). In an earlier study the author found that *P. marginatus* had the highest fecundity and longest life span at a temperature of 28.7°C and relative humidity of 65 ± 2% (Chong et al., 2003).

Parasitoid and predator populations were well synchronized with that of the host/prey, *R. iceryoides*. Despite the fact that these natural enemies seemed to be working in a density dependant manner in relation to the population of the pest, they played a minimal role in the seasonal dynamics of *R. iceryoides*. The individual and combined action of the parasitoid as well as that of predators were found to be very low on all plant parts in both Kibaha and Dar es Salam during the entire study period. This could be due to that fact that these parasitoid species, being indigenous to Africa, share no evolutionary history with that of the pest. On the other hand most predators encountered during the studies were generalists and therefore exerted very little pressure on the pest population; considering the fact that there were other sympatric mealybug species in the orchard (Tanga et al., unpuplished data), in addition to other pests.

Although several species of hyperparasitoids were recorded in this study, their abundance as well as percentage hyperparasitism was extremely low at both Kibaha and Dar es Salaam. Therefore, their impact on the population of the primary parasitoid was quite low. However, caution needs to be taken when introducing parasitoid species for biological control of the pest. Efforts are underway to introduce co-evolved parasitoids from the pest aboriginal home (India) but laboratory studies are needed to establish whether these hyperparasitoids might attack the introduced parasitoid species, and limit their efficiency in suppressing the pest. There is much evidence which showed that hyperparasitoids can influence the population dynamics and community structure of primary parasitoids enough to disrupt biological control of the host (Holler et al., 1993, Morris et al., 2001; Van Nouhuys and Hanski, 2000; Van Veen et al., 2001).

These results provide valuable information of the seasonal dynamics of the invasive pest *R. iceryoides*, and permits forecasting of possible outbreaks based on the prevailing weather conditions. Based on the light of the results of this study, it can also be concluded that the population of the indigenous parasitoids were not able to reach to a level that could suppress the exploding population of this pest. This calls for introduction of co-evoleved natural enemies from the pest's aboriginal home, India. Information generated during this study on the seasonal fluctuation pattern of *R. iceryoides*, could also be used to guide future augmentative releases of the most dominant indigenous parasitoid species, *A. pseudococci* or possible introduced parasitoid species. For example, the mass releases of the parasitoids during the off-mango season (March- Septemper), where the pest populations in mango orchards are quite low, but relatively high on the alternative host plants, such Jerusalem-thorn, *P. aculeata* and pigeon pea *C. cajan* (Tanga, unpublished data), will help parasitoid population build-up ahead of the heavy infestations by *R. iceryoides* during the mango fruiting season (October-February). Therefore, contributing to the suppression of the pest and enhancing the livelihood of the growers.