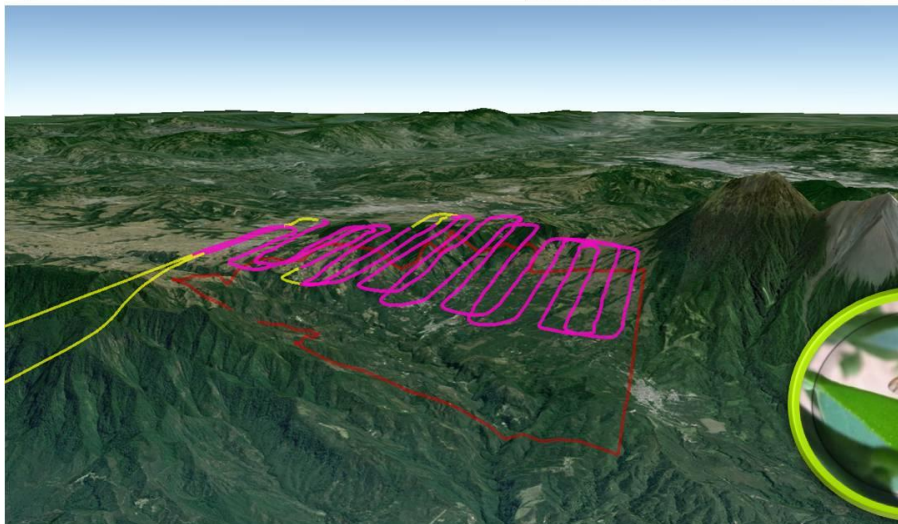




**IAEA**

**Joint FAO/IAEA Programme**  
Nuclear Techniques in Food and Agriculture

# STERILE INSECT RELEASE DENSITY CALCULATIONS SPREADSHEET



Food and Agriculture Organization of the United Nations  
International Atomic Energy Agency  
Vienna, 2019

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The proper citation for this document is:

**FAO/IAEA. 2019.** Sterile Insect Release Density Calculations Spreadsheet, Rendón P.A, Enkerlin W.R. and Cáceres C. (eds.), Food and Agriculture Organization of the United Nations/International Atomic Energy Agency. Vienna, Austria. 30 pp.

# **STERILE INSECT RELEASE DENSITY CALCULATIONS SPREADSHEET**

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Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture

Food and Agriculture Organization of the United Nations  
International Atomic Energy Agency  
Vienna, 2019

## **FOREWORD**

Sterile insects have been defined as beneficial organisms by the International Plant Protection Convention (FAO 2017). The sterile insect technique (SIT) has been applied in more than 30 countries worldwide for pest suppression, eradication, containment and prevention. Current sterile insect production supporting pest control programmes is over 3 billion insects per week. The investment that programmes conduct in SIT application (including production and release of sterile insects) is very substantial; therefore, an efficient use of the sterile insects is fundamental to provide economic feasibility to SIT programmes.

This spreadsheet model is a valuable tool for optimization of sterile insect release programmes. The calculations presented in this spreadsheet can be used for pests other than fruit flies because the same principles apply. The introduction of pest specific parameters will be required to adapt its use to other SIT pest control activities. The use of Excel software allows all action programmes worldwide to have easy access to the spreadsheet calculations since its use is global nowadays. This spreadsheet model has been validated and it is being used routinely in large-scale operational field programmes.

If users need to consult more details of this or other processes do not hesitate in contacting the editors of this manual.

The FAO/IAEA Officers responsible for the publication were Pedro A. Rendón, Walther R. Enkerlin and Carlos Cáceres.

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## 1. INTRODUCTION

The use of sterile insects to control their own species is an innovative practice that reduces the use of pesticides (and generates an alternative for pesticide resistance management), improves food security and safety and reduces environmental impact. These aspects have favored the use and expansion of sterile insects as a control tool, which includes pests with an impact on agriculture and livestock production, as well as pest that have a direct impact on human health.

The production of sterile insects and their subsequent emergence and release in the field are highly specialized activities that require appropriate protocols for the efficient application of the sterile insect technique (SIT) to control the target pests. The procedures for sterile insect production and the emergence and release, are reported in other procedures manuals (FAO/IAEA 2017). This document is more specific for the dynamic determination of sterile insect release density and their capacity of induction of sterility in a given wild population. In order to ensure the effectiveness of sterile insects, it is necessary to consider a number of factors that influence their efficiency in the field. These factors and their impact will be presented and discussed briefly.

The objective of this manual is to present a systematized form of (1) Measuring pest population growth through a network trapping system and the determination fly per trap per day (FTD) and by direct measurement inside field cages, (2) Determine the percent of induction of sterility introduced by sterile insects and the sterile: fertile (wild) ratio at which it achieves the greatest effectiveness, (3) The calculations that allow carrying out sterile insect releases with desired densities and sterile: fertile ratios to obtain predictable results, (4) Present the computations in the form of an electronic spreadsheet which facilitates calculations, analysis and record keeping, as well as (5) the graphical representation of the conditions under which insect control activities are carried out in the field to facilitates decision making by action programs managers.

## 2. CONTROLLING WILD INSECT POPULATIONS

### 2.1. Determining population growth through a trapping detection network

Fruit fly control programmes use a trapping array based on the deployment of different types of traps and attractants to determine the presence of a particular pest species (FAO/IAEA 2018). A practical procedure to establish and manage trapping networks based on risk factors is available in “Manual and spreadsheet for the application of the risk factors criteria for fruit fly trapping” (FAO/IAEA *in press*). After traps are collected, scored and data processed, usually on a weekly basis, a graphical display of the pest presence, in actual number of individuals collected, can be produced. Since traps are maintained in the field for the whole year, then it is possible to summarize the results monthly and determine periods which have a higher population growth as well as when limiting factors reduce population presence during the monitoring period (see Fig. 1). Another way of representing/estimating the pest presence is by using indices such as the number of flies/trap/day (FTD) which considers the number of flies, numbers of traps and the numbers of days between trap inspections or services.

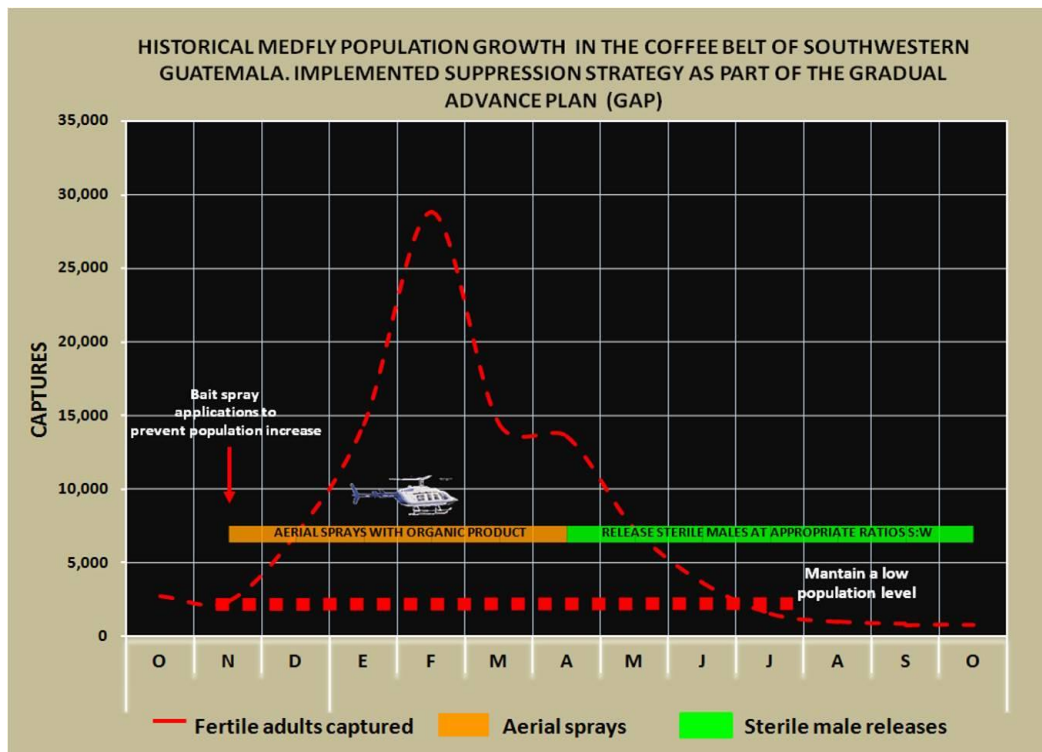
#### **Fly/trap/day (FTD) value for the fertile (wild) population:**

$$FTD\ wild = \frac{\text{Total captured wild flies}}{(\text{Total number of traps}) \times (\text{Number of days in the field})}$$

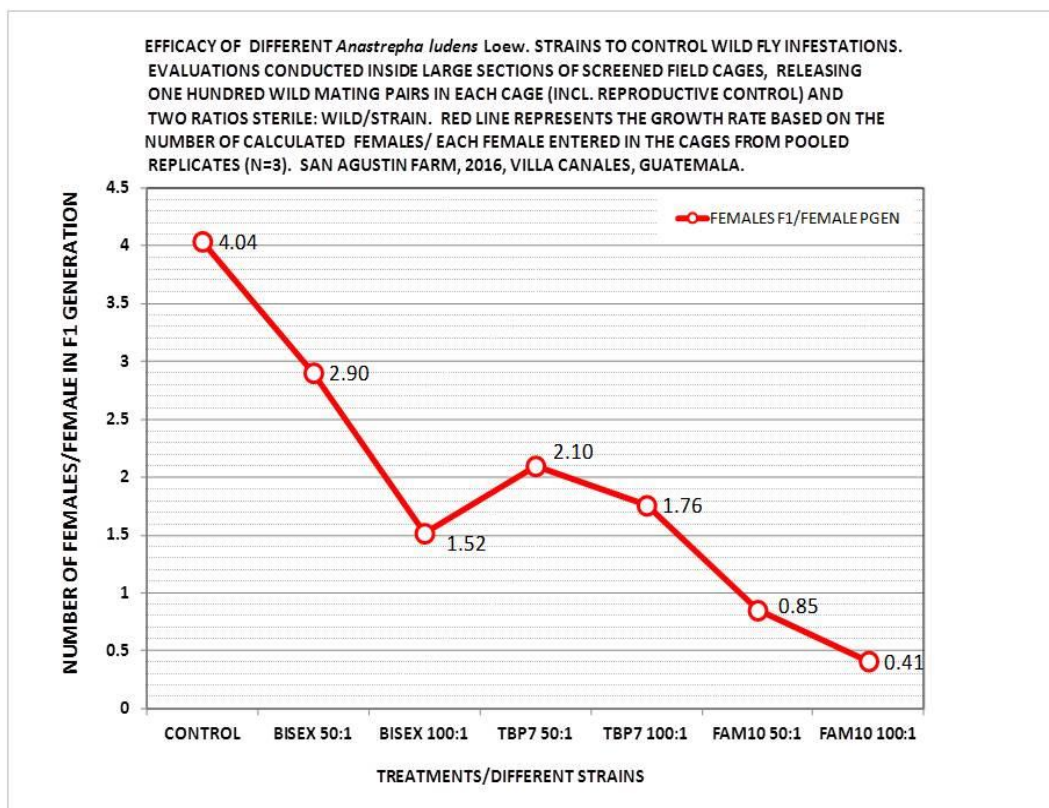
The FTD measures the size of a population in space and time in relative not absolute numbers. The FTD is used as an action threshold for the application of control activities including bait sprays for population suppression and sterile insects for population reduction to low pest prevalence or eradication levels. Once the location and size of the resident population based on FTD have been determined, then the control activities are planned. In tropical and subtropical areas, the current strategy is to reduce the resident population with the use of an insecticide-bait product that applied in small/localized areas at the beginning of the population growth, will not only minimize the use of insecticides but also prevent population increase and its dispersion. These insecticide-bait applications once the resident population has been reduced to an acceptable level (0.05 FTD) are stopped to allow the release of sterile insects (Fig. 1). The acceptable level FTD value that triggers each control activity may vary according to the climate and ecological conditions of the areas. Due to the residuality effect of insecticide/bait spray products the actual number of applications during the year is low.

### 2.2. Determining population growth through direct measurement inside field cages

A direct way of measuring population growth rate (R value) is to introduce a known number of females into a cage allowing their reproduction and then, at the following generation (F1) determine the number/survivorship of females produced from the original number introduced at the parental generation ( $P_{gen}$ ) to determine their growth (i.e.  $F1/P_{gen} = 404/100 = 4.04$  times or “4.04x” per generation). In the example given below (Fig. 2), equal number of wild females and males (100 mating pairs) were released inside large screened cages. A control treatment allowed to determine a growth rate of 4.04x, while treatments in which sterile insects from three different strains (*BSEX* = standard strain in which the two sexes have to be released due to the lack of a sex separation mechanism; *TBP7* = *Tapachula black pupae* strain # 7, strain from which only males were released due to the presence of a dimorphic trait or genetic sexing mechanism that allows pupae separation into males and females; females have a black color of puparium while males have brown, and Family 10 or “FAM10” strain, in which only males were release due to the presence of the same sexing system as in *TBP7*) were released at two ratios (50:1 or 100:1) in combination with wild insects, reduced population growth when compared to the control. The most efficacious treatment (strain/ and ratio sterile: wild) should be chosen for use in field control activities.



**Figure 1.** Control strategy once the pest population numbers and their location have been established by means of the use of a trapping network. Note the initial use of bait spray applications and the deployment of the sterile insect technique (SIT) for an extended period of the year compared to the number of bait spray applications.



**Figure 2.** Population growth and reduction (per treatment), based on the numbers of females produced in the following generation from an original number in the parental P<sub>gen</sub>. The growth rate is related to the host and other existing conditions at the time of reproduction.



With the aim of exemplifying the impact of the population growth by itself as well as the effect of the induction of sterility produced by the release of good quality sterile insects, a basic population growth equation is proposed:

$$P_f = P_i \times R (1 - C)$$

WHERE:

$P_f$  = Final population number

$P_i$  = Initial population number

R = Population growth (Number of females in F1 compared to the number of females in the parental generation, see 2.2. above).

C = value of the control activity (i.e. induction of sterility from SIT deployment or other control measures such as bait sprays, parasitoids and SIT combined, etc.)

Final population size without control → C=0, initial population number  $P_i=5$  (see detailed calculations on Table 1).

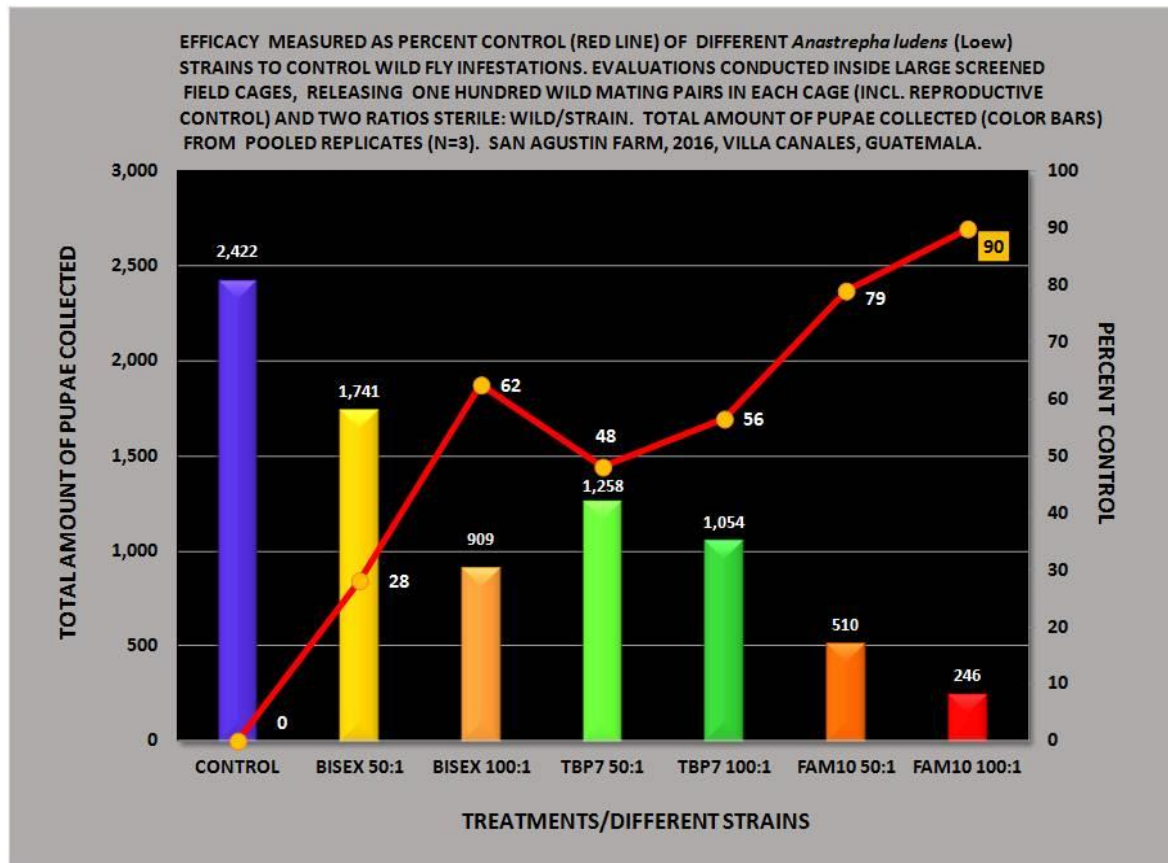
**Table 1.** Population increase based on fixed growth rate without control measures.

| Generation       | Final population size $P_f$<br>without control C = 0 | Initial population<br>number $P_i$ | From Fig.2, above<br>Fixed growth rate "R" | (1 - Control index)<br>When C is = 0 |
|------------------|--|------------------------------------|--|--------------------------------------|
| P <sub>Gen</sub> | 5  | 5                                  | 4.04                                       | 1                                    |
| F1               | 20   | 20                                 | 4.04                                       | 1                                    |
| F2               | 82   | 82                                 | 4.04                                       | 1                                    |
| F3               | 330  | 330                                | 4.04                                       | 1                                    |
| F4               | 1,332  | 1,332                              | 4.04                                       | 1                                    |
| F5               | 5,381  | 5,381                              | 4.04                                       | 1                                    |
| F6               | 21,740   | 21,740                             | 4.04                                       | 1                                    |

### 2.3. Determining the percent induction of sterility introduced by a sterile insect strain and the sterile: fertile ratio at which it achieves its best effectiveness

Induction of sterility is the degree of interference (percent control) that sterile insects achieve when released into a wild resident population (Knipling 1955). Large field screened cages (15 m x 7.5 m x 2.5 m) were used to release wild mating pairs along with sterile insects at different sterile: wild ratios in order to determine the degree of interference that reared insects achieve (field performance quality control). Insects used for these types of evaluations are left in the field for one week inside cage enclosures (mimicking action Programme weekly releases). Tests are replicated both in time and space. Sterile and wild insects are subjected to existing climatic conditions, mating and remating as well as predation. The resulting female egg laying is collected in fruit during the one-week-test and brought into a fruit processing facility where larvae are allowed to feed, leave the fruit and pupate. Collected stages are recorded for statistical analysis. The resulting data sets allow analyzing the performance of sterile insects at the given proportions. In the example presented, sterile to wild ratios of 50 and 100 were used and the induction of sterility was measured for one (P to F1) generation (Fig. 3).

Common biotic and abiotic factors that can affect the efficiency of SIT are presented in Annex 1.



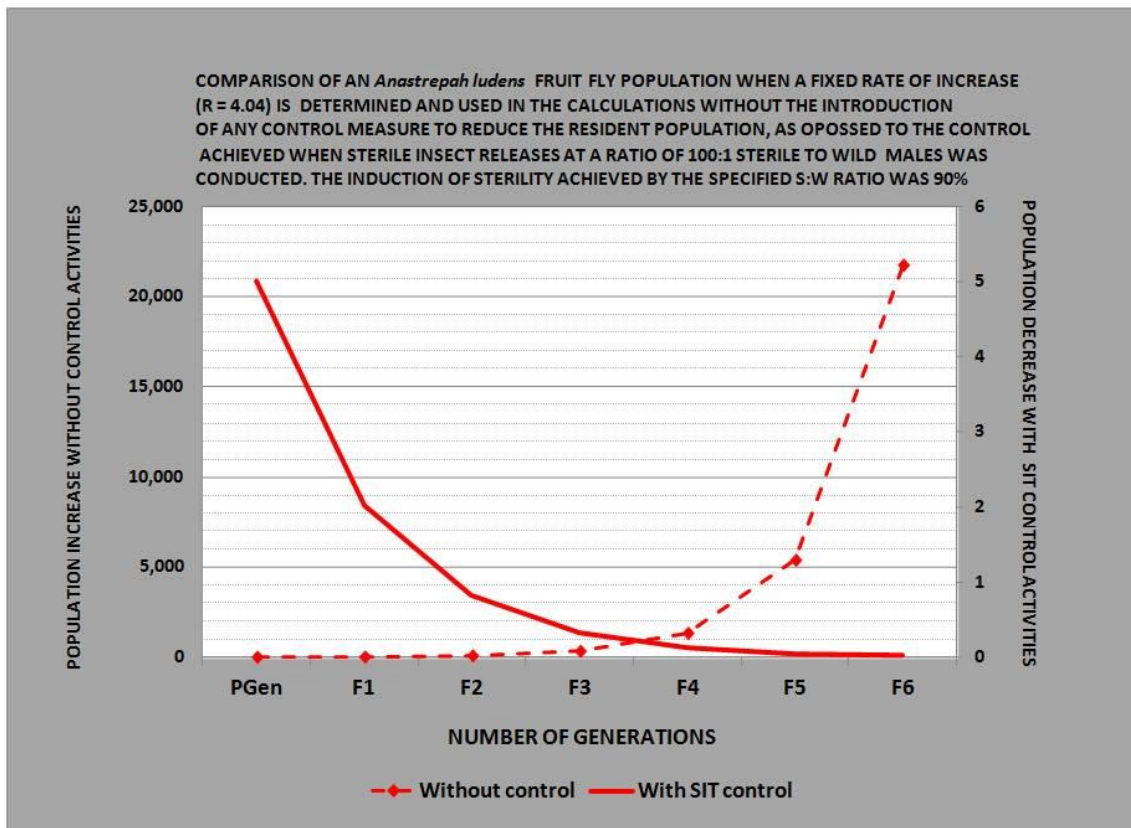
**Figure 3.** Percent control achieved in the comparison of different strains and ratios sterile male to wild male.

From Fig. 3, it can be determined that the highest percent control (90%) was achieved by strain Fam10 at a ratio of 100:1 sterile: wild males. This control value ( $C = 0.9$ ) can be used to determine the impact of successive good quality sterile insect releases into the wild population (Table 2).

**Table 2.** Population decrease based on fixed growth rate with control achieved by a proportion of 100:1 ratio of sterile: wild males from a genetic sexing strain of *Anastrepha ludens* Loew., black pupae strain Fam10.

| Generation | Final population size $P_f$ with control<br>$C = 0.9$ , Figure 3, above. | Initial population number $P_i$ | From Figure 2, above<br>Fixed growth rate "R" | (1-Control index)<br>$C = 0.9$ |
|------------|--|---------------------------------|---|--------------------------------|
| PGen       | 5  | 5                               | 4.04  | 0.1                            |
| F1         | 2.02   | 2.02                            | 4.04  | 0.1                            |
| F2         | 0.82   | 0.82                            | 4.04  | 0.1                            |
| F3         | 0.33   | 0.33                            | 4.04  | 0.1                            |
| F4         | 0.13   | 0.13                            | 4.04  | 0.1                            |
| F5         | 0.05   | 0.05                            | 4.04  | 0.1                            |
| F6         | 0.02   | 0.02                            | 4.04  | 0.1                            |

A theoretical graphical comparison of the results achieved with and without control activities is shown in Fig. 4.



**Figure 4.** Comparison of the control achieved with and without the release of sterile flies within a wild population

### 3. CALCULATING RELEASE DENSITIES AND THEIR RELATED STERILE: WILD RATIOS

The basic operational procedure shown in the following example is the same automated procedure used by the Spreadsheet Calculator presented in Section 4.4. To establish sterile insect release densities for action programmes that work in fruit fly infested areas, it is important to determine, first, the level of the wild population. As mentioned in Section 2.1., it can be estimated in relative terms by using a trapping scheme and the FTD index as described in FAO/IAEA (2017).

The procedure is as follows:

This procedure considers that the response of the sterile released flies and the wild flies to traps is equal.

**Determine the fly/trap/day (FTD) value for the fertile (wild) population:**

$$FTD\ wild = \frac{\text{Total captured wild flies}}{(\text{Total number of traps}) \times (\text{Number of days in the field})}$$

**Determine FTD value for released sterile flies, as follows:**

$$FTD_{sterile} = \frac{\text{Total recaptured sterile flies}}{(\text{Total number of traps}) \times (\text{Number of days in the field})}$$

Using the figures calculated from the 2 formulas above, we can calculate the sterile: wild ratio present in the field.

$$Ratio (S:W) = \frac{FTD_{sterile}}{FTD_{wild}}$$

Managers should determine an appropriate S:W ratio according to the action programme objective, the minimum initial release ratios recommended are summarized in Table 3.

**Table 3:** Minimum recommended initial release ratios depending on the action programme objective and ecological conditions including hosts in the target area (the ratios in the table correspond to Mediterranean fruit fly in tropical/subtropical conditions in coffee plantations).

| Programme objective  | Avg. Ratios* |
|----------------------|--------------|
| Suppression          | 25-100 :1    |
| Eradication          | 100-150: 1   |
| Containment          | 50-150: 1    |
| Preventive Release** | 25-50:1      |

\*Minimal S:W ratio. This ratio will continue to increase as  $FTD_{wild}$  is reduced due to suppression and SIT application/control.

\*\*Suggested ratio to ensure a minimum number of sterile flies required to outnumber potential entry. Based on the assumption that one wild fly is caught per trap per cycle, irrespective of whether a wild fly is caught or not.

If the calculated ratio S:W does not meet the objective of the action Programme (Table 3) additional non-SIT suppression measures need to be implemented before sterile insects can be released (i.e. bait sprays – See Fig. 1) or additional sterile flies have to be released per unit surface area to increase the over-flooding ratio. In the case of sterile fly release if the S:W ratios are above the recommended ratio, then the release density can be decreased.

In this case, only when the target  $FTD_{wild}$  of 0.05 has been achieved, should sterile fly releases be initiated. The 0.05 value is an  $FTD_{wilde}$  value above which it is normally not recommended to use sterile insects, except for hotspots situations.

**Example:**

Assuming that 5 traps are placed in  $1\text{km}^2$  (100 ha) and are exposed in the field for 7 days, and that these traps captured 3 wild flies, then:

$$FTD_{wild (W)} = 3 \text{ flies}/(5 \text{ traps} \times 7 \text{ days}) = 0.085$$

The same calculation using  $FTD_{sterile}$ :

Assuming 1,000,000 sterile flies were released in the same 1 km<sup>2</sup> area (release density of 10,000 sterile insects per hectare) and that a total of 3,000 flies were recaptured, then:

$$\text{FTD}_{\text{sterile (S)}} = 3,000 \text{ flies} / (5 \text{ traps} \times 7 \text{ days}) = 85.71$$

The current sterile:wild ratio would be:

$$\text{FTD}_W / \text{FTD}_F = 85.71 / 0.085 = 1008 \text{ (1008}_w\text{:1}_F\text{)}$$

The required number of sterile flies for a 50:1 ratio would be:

1,000,000 released sterile flies  
1008 current sterile: wild ratio  
50 required sterile: wild ratio

**New release rate in Km<sup>2</sup> = (required s: w ratio x previous release rate)/current s: w ratio.**

$$(1,000,000 \times 50) / 1008 = 49,600 \text{ sterile flies in 100 ha (1 km}^2\text{)}$$

**Number of sterile flies per hectare**

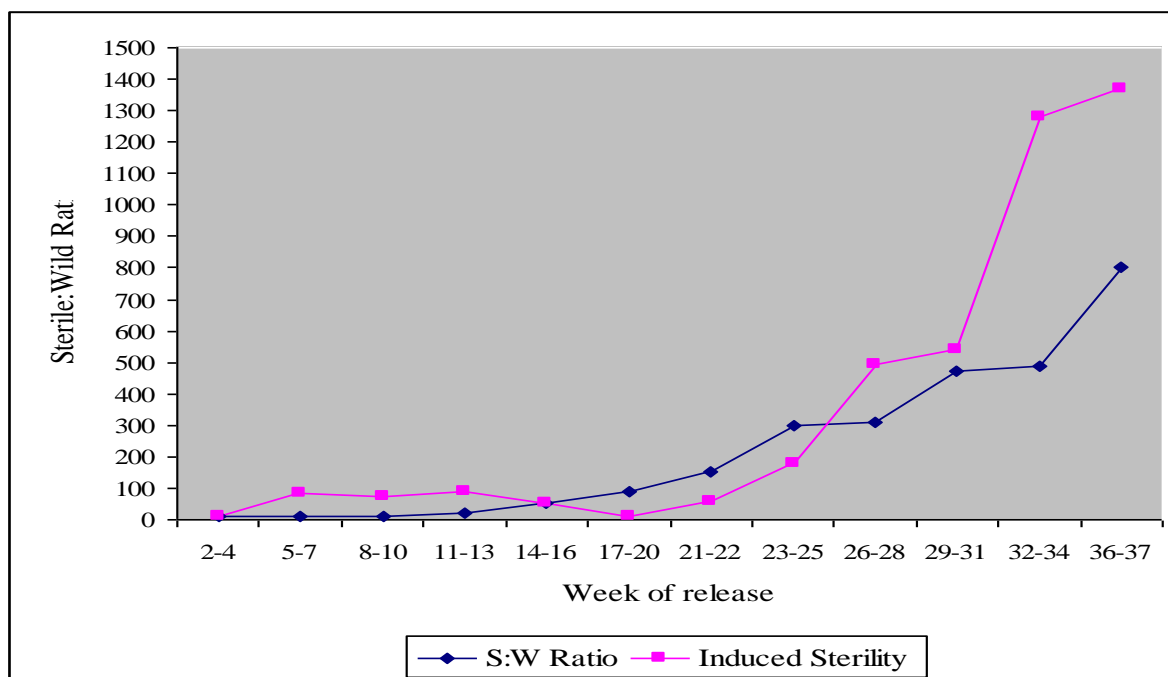
$$(49,600 / 100) = 496 \text{ sterile flies/ha}$$

If the ratio S:W needs to be increased there are two options to achieve the desired ratio:

- 1.) Additional suppression measures (i.e. bait sprays) can reduce  $\text{FTD}_{\text{wild}}$  from 0.085 to  $\text{FTD}_{\text{wild}} = 0.03$ , therefore the new S:W ratio is, 142:1 ( $0.085 / 0.03 * 50$ )
- 2.) Increase the sterile fly numbers to achieve the required ratio of steriles (ie. 142); to calculate the new release numbers substitute the new ratio in d) above.

$$(1,000,000 * 142) / 1008 = 140,873 \text{ sterile flies in one km}^2 \text{ or 1,408 in one hectare}$$

As the control process progresses the initial S: W ratio will increase (Fig. 5). This ratio will continue to increase as long as the  $\text{FTD}_{\text{wild}}$  continuous to decrease due to suppression measures and release of good quality sterile flies is kept constant (Rendón, et al 2004).



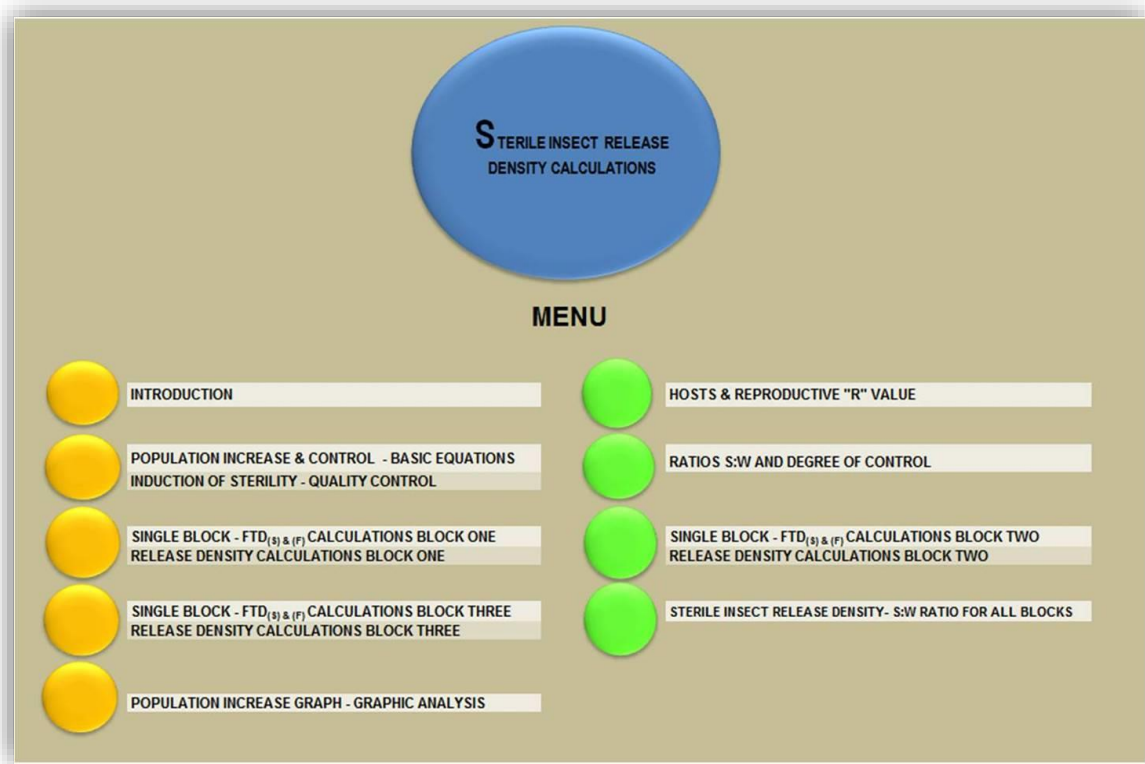
**Figure 5.** Increased S:W ratio as result of the sterile insect technique (SIT) application.

Programmes that are initiating area-wide SIT operations should determine their required release densities considering the conditions under which activities will be conducted such as assessed wild FTDs, objectives of the programme (suppression, eradication, etc.) and established over-flooding ratios. The existing SIT programmes, their objective and actual sterile insect release densities are shown in Annex 2.

#### **4. SPREADSHEET FOR STERILE INSECT RELEASE DENSITY CALCULATIONS**

This excel spreadsheet calculator allows determining sterile insect release ratios sterile: wild (S:W) based on actual field trapping data, it also enables, based on the desired S:W ratio calculating the required release densities for aerial release blocks (i.e. a release block is an arbitrary size area, an acre, a hectare, a square mile or square kilometer, often composed of hundreds or thousands of such unit areas), defined by the user of the SIT technology and is the surface area over which sterile insects are released using the most cost-effective or the available method, which can be by air or by ground. This spreadsheet has been validated and it is being routinely used in large-scale operational field programmes.

The spreadsheet has an initial menu as displayed in Fig. 6.



**Figure 6.** Initial page of the spreadsheet for the sterile insect release density calculations.

Each of the yellow and green buttons directs the user to a different page, where operational information needs to be entered and calculations are performed. The basic content of each of the pages in the menu is shown in Table 4.

**Table 4.** Page content description of the spreadsheet for the sterile insect release density calculations.

| SUBJECT   | CONTENTS   |
|---|--|
| INTRODUCTION  | General statements about the use of the spreadsheet.   |
| HOST & REPRODUCTIVE “R” VALUE                                     | Shows population growth equation and documented “R” values for different hosts. More values can be added based on user available information.  |
| POPULATION INCREASE & CONTROL (BASIC EQUATIONS)                   | Allows calculating population growth without and with control based on “R” values and the degree of control “C” that the user can input. Graphical display of the results can be viewed through an internal page link.   |
| RATIOS S:W AND DEGREE OF CONTROL                                  | Graphs showing results of different evaluations to determine appropriate ratios S: W and the control achieved. This page also shows ratios and release densities according to Programme working area as well as examples of block design and monitoring.   |
| SINGLE BLOCK - FTD <sub>(S)</sub> & (F)<br>CALCULATIONS BLOCK ONE | This page is in its design and operation similar to the pages for single block calculations two and three (see Figure 6). <i>These pages were prepared to simulate an action Programme operation with a minimum of three blocks but more can be added, as needed, and linked as a standard Excel file operation.</i> The information entered here allows calculating for the whole year (on a weekly basis) the fly/trap/day index based on the size of the user Programme blocks, the number of traps deployed, number of insects captured and so on. It also calculates weekly field ratios S: W, this information is transferred to the page named release density calculations |
| RELEASE DENSITY CALCULATIONS BLOCK ONE                            | This page is in its design and operation similar to the pages for release density calculations blocks two and three (see Figure 6). <i>These pages were prepared to simulate an action Programme operation with a minimum of three blocks but more can be added, as needed, and linked as a standard Excel file operation.</i> This page allows calculating the required weekly release density per release block. The information required is the previous week release rate as well as the ratio S: W that the operational Programme aims at.  |
| STERILE INSECT RELEASE DENSITY S:W RATIO FOR ALL BLOCKS           | This page allows adding up the amounts of insects required/block to generate a total quantity of sterile insects required/weekly by an operational Programme.  |
| POPULATION INCREASE GRAPH - GRAPHIC ANALYSIS                      | Summary of all the graphs generated by each of the pages of the spreadsheet. It shows population growth with and without control activities, graphs that show sterile insect performance by release block and the summary of insect needs for all blocks. This summary can help managers to move sterile insects between blocks as required.   |



## 4.1. Hosts and reproductive "R" value

The spreadsheet pages (4.1 and 4.2) conduct an automatic demonstration/calculation of the subject presented in sections 2.2 and 2.3 above. These pages can be used to validate the control technology being used to reduce a population growing in a characterized growth rate host.

The page 4.1 shows population growth equation and documented “R” values for different hosts (Fig. 7). The information presented here for host growth rate, can be used in the population growth equation. Additional information generated by the user/action programme can be reported/filed here for future modeling/use.

INSECT PESTS USUALLY HAVE SEVERAL HOSTS IN WHICH THEY CAN GROW THEIR IMMATURE/LARVAL STAGES. FRUIT FLIES GENERALLY HAVE MORE THAN ONE HOST, WITH SPECIES SUCH AS THE MEDITERRANEAN FRUIT FLY HAVING MORE THAN 579 SPECIES OF FRUITS AND VEGETABLES REPORTED (Liquido, et al 2017)

FEMALES OF THE DIFFERENT INSECT SPECIES LAY THEIR EGGS IN AVAILABLE HOSTS AND PRODUCE AN INCREASED NUMBER OF INDIVIDUALS IN THE FOLLOWING GENERATION.

A BASIC MATHEMATICAL FORMULA THAT DESCRIBES POPULATION GROWTH IS:

$$P_f = P_i \times R (1-C)$$

WHERE:

Pf = FINAL POPULATION NUMBER  
Pi = INITIAL POPULATION NUMBER

R = POPULATION GROWTH (NUMBER OF FEMALES IN F1 COMPARED TO THE NUMBER OF FEMALES IN PARENTAL GENERATION).  
C = CONTROL VALUE.

"R" VALUE FOR : MEDITERRANEAN FRUIT FLY

| HOST LIST | SCIENTIFIC NAME           | GROWTH RATE |
|-----------|---------------------------|-------------|
| COFFEE    | <i>Coffea arabica</i> (L) | 9           |
| SPONDIAS  | <i>Spondias purpurea</i>  | 6.5         |
|           |                           |             |
|           |                           |             |

GO TO POPULATION INCREASE FORMULA

← → | MENU | INTRO | **HOSTS & REPRODUCTIVE "R" VALUE** | POP. INCREASE FORMULA | POP. INCREASE GRAPH | RATIOS SW AND DEGREE OF CONTROL | SINGLE BLOCK F

**Figure 7.** appearance of the page: Host and reproductive value “R”.

Each host will provide specific conditions to the insect pest and that will generate a particular growth rate for the host species. If sterile insect releases are conducted in areas that are planted with particular host/vegetation it would be important to know the growth potential of the pest under those conditions to direct and assess control activities.

## 4.2. Population increase & control – basic equations

The page 4. 2 together with the previously described allows programme managers to assess the effect of control actions over an insect population. This page in particular, allows the user to compute population growth with and without control activities. Population growth is based on “R” values and it can be combined to the degree of control “C” value that the user could input. A graphical display of the results can be viewed through an internal page link that transfers to the page named “graphic analysis”.

The page is divided into two sections; the first section allows the user, based on the population growth formula presented in 2.2, to calculate the increase of the population without control. To automatically conduct this calculation, the desired initial population and the chosen increase rate need to be entered. The numbers in red allow entering the desired user or action programme information to calculate the population growth without control. The user can decide on: (1) the size of the initial population (Pi) and (2) if the calculations are going to be performed using a fix or a variable growth rate (see red numbers in Fig. 8).

$P_f = P_i \times R (1-C)$   
WITHOUT CONTROL ACTIVITIES

| GENERATION | FINAL POPULATION SIZE (P <sub>f</sub> ) W/O CONTROL | P <sub>i</sub> | FIXED/VARIABLE GROWTH RATE** "R" | (1 - CONTROL INDEX "C")<br>"C" = 0 |
|------------|---|----------------|----------------------------------|------------------------------------|
| PG         | 5   |                | 9                                | 1                                  |
| F1         | 45  | 45             | 9                                | 1                                  |
| F2         | 405   | 405            | 9                                | 1                                  |
| F3         | 3,645   | 3,645          | 9                                | 1                                  |
| F4         | 32,805  | 32,805         | 9                                | 1                                  |
| F5         | 295,245   | 295,245        | 9                                | 1                                  |
| F6         | 2,657,205   | 2,657,205      | 9                                | 1                                  |
| F7         | 23,914,845  | 23,914,845     | 9                                | 1                                  |
| F8         | 215,233,605   | 215,233,605    | 9                                | 1                                  |
| F9         | 1,937,102,445                                       | 1,937,102,445  |                                  | 1                                  |
| F10        | #VALUE!   | #VALUE!        |                                  | 1                                  |
| F11        | #VALUE!   | #VALUE!        |                                  | 1                                  |
| F12        | #VALUE!   | #VALUE!        |                                  | 1                                  |

**Figure 8.** Final population and population growth based on a user defined “R” value. Numbers in red allow entering the desired user or action programme information to calculate the population growth without control.

The second section of this page, deals with a population over which a control activity has been conducted and a measurable effect of this action (percentage control) can be introduced as the “C” value (see red number in Fig. 9). Introducing this control value allows calculations on the population reduction that can be observed in the column FINAL POPULATION SIZE WITH CONTROL ACTIVITIES (Fig. 9) and that are also graphically displayed in the page POPULATION INCREASE GRAPH - GRAPHIC ANALYSIS.

As it can be deduced from this calculations and graphical results, programme managers need to document the degree of control achieved by each of the control measures performed in field whether those actions are alone (bait sprays, bait stations, mechanical control, release of sterile insects, or a combination with others such as SIT and parasitoids) and then with that information be able to model the efficacy of their activities.

$P_f = P_i \times R (1-C)$   
WITH CONTROL ACTIVITIES

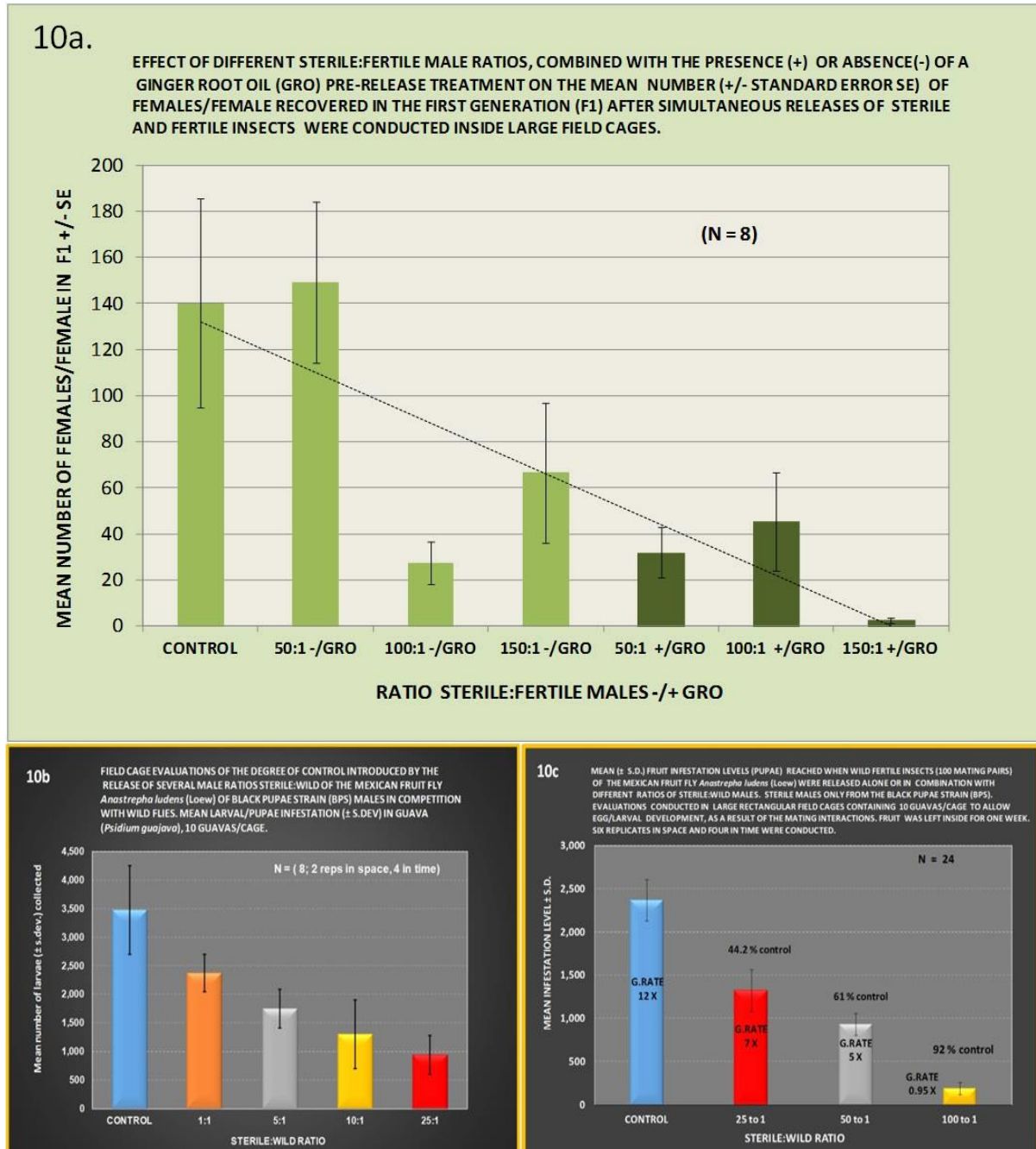
| CONTROL INDEX ("C") USING STERILE INSECTS* | R x (1-CONTROL INDEX) | P <sub>f</sub> | FINAL POPULATION SIZE (P <sub>f</sub> )<br>WITH CONTROL ACTIVITIES |
|--|-----------------------|----------------|--|
| "C" = 0.92                                 |                       |                |  |
| 0.92                                       | 0.72                  | 5.00           | 5.00   |
| 0.92                                       | 0.72                  | 3.60           | 3.60   |
| 0.92                                       | 0.72                  | 2.59           | 2.59   |
| 0.92                                       | 0.72                  | 1.87           | 1.87   |
| 0.92                                       | 0.72                  | 1.34           | 1.34   |
| 0.92                                       | 0.72                  | 0.97           | 0.97   |
| 0.92                                       | 0.72                  | 0.70           | 0.70   |
| 0.92                                       | 0.72                  | 0.50           | 0.50   |
| 0.92                                       | 0.72                  | 0.36           | 0.36   |
| 0.92                                       | #VALUE!               | 0.26           | 0.26   |
| 0.92                                       | #VALUE!               |                | #VALUE!  |
| 0.92                                       | #VALUE!               |                | #VALUE!  |
| 0.92                                       | #VALUE!               |                | #VALUE!  |
| 0.92                                       | #VALUE!               |                | #VALUE!  |

**Figure 9.** Final population based on a known growth rate “R” and a degree of control “C”.

For a comparison of SIT releases with a known induction of sterility or control “C” value, see Fig. 4 in Section 2.3.

### 4.3. Ratios S:W and degree of control

This page contains several graphical examples, showing the user results of different evaluations that allowed determining ratios S:W and their respective degree of control achieved (Fig. 10).



**Figure 10.** Field evaluations of sterile insects to assess their impact on the reduction of wild populations. In (10a), Using sexual preconditioning, (10b & c), densities as resources to improve field efficacy of the SIT.

This page also shows the suggested release densities and ratios sterile: wild according to Programme working area and block objective (Fig. 11) as well as examples of block designs, with recommended release densities, monitoring of sterile: wild ratios achieved by release block (Fig. 12) and the sterile insect aerial release patterns (Fig. 13).

**PROPOSED RELEASE RATES ACCORDING TO WORKING AREA AND AIMED RATIOS  
STERILE TO WILD FOR THE MEDITERRANEAN FRUIT FLY *Ceratitis capitata* (Wied.)**

| Working Area   | SIT Block Objective | Lower Densities | Max. Densities | Sterile / Wild Ratio * |
|----------------|---------------------|-----------------|----------------|------------------------|
| Free           | Prevention          | 500             | 500            | Min. 200 a 1           |
|                | Erradication        | 1,000           | 5,000          | Min. 200 a 1           |
| Low Prevalence | Prevention          | 500             | 500            | Min. 200 a 1           |
|                | Erradication        | 1,000           | 5,000          | Min. 200 a 1           |
| Supression     | Supression          | 1,000           | 6,000          | Min. 150 a 1           |
|                | Contention          | 1,000           | 6,000          | Min. 50 a 1            |
| Monitoring     | NA                  | NA              | NA             | NA                     |

\* The Sterile : Wild ratio is calculated first, and then the targeted density is defined within the range specified for each working area and SIT block objective.  
 \*\* The use of a 200:1 ratio is to allow for mortality, predation and/or quality issues and still ensure appropriate field results (see graph)

Figure 11. Recommended release densities, for Mediterranean fruit fly action programmes.

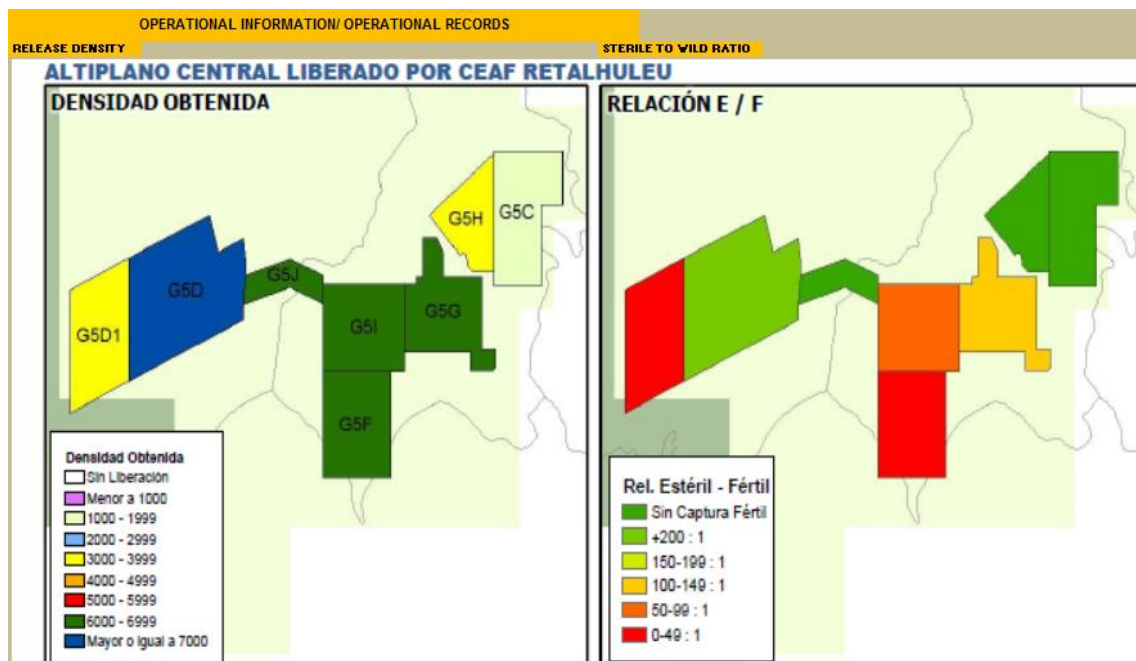


Figure 12. Left: recommended release densities by block and Right: monitoring of sterile: wild ratios at each release block. Recommend release densities and monitoring by blocks can be achieved through the use of this spreadsheet calculator



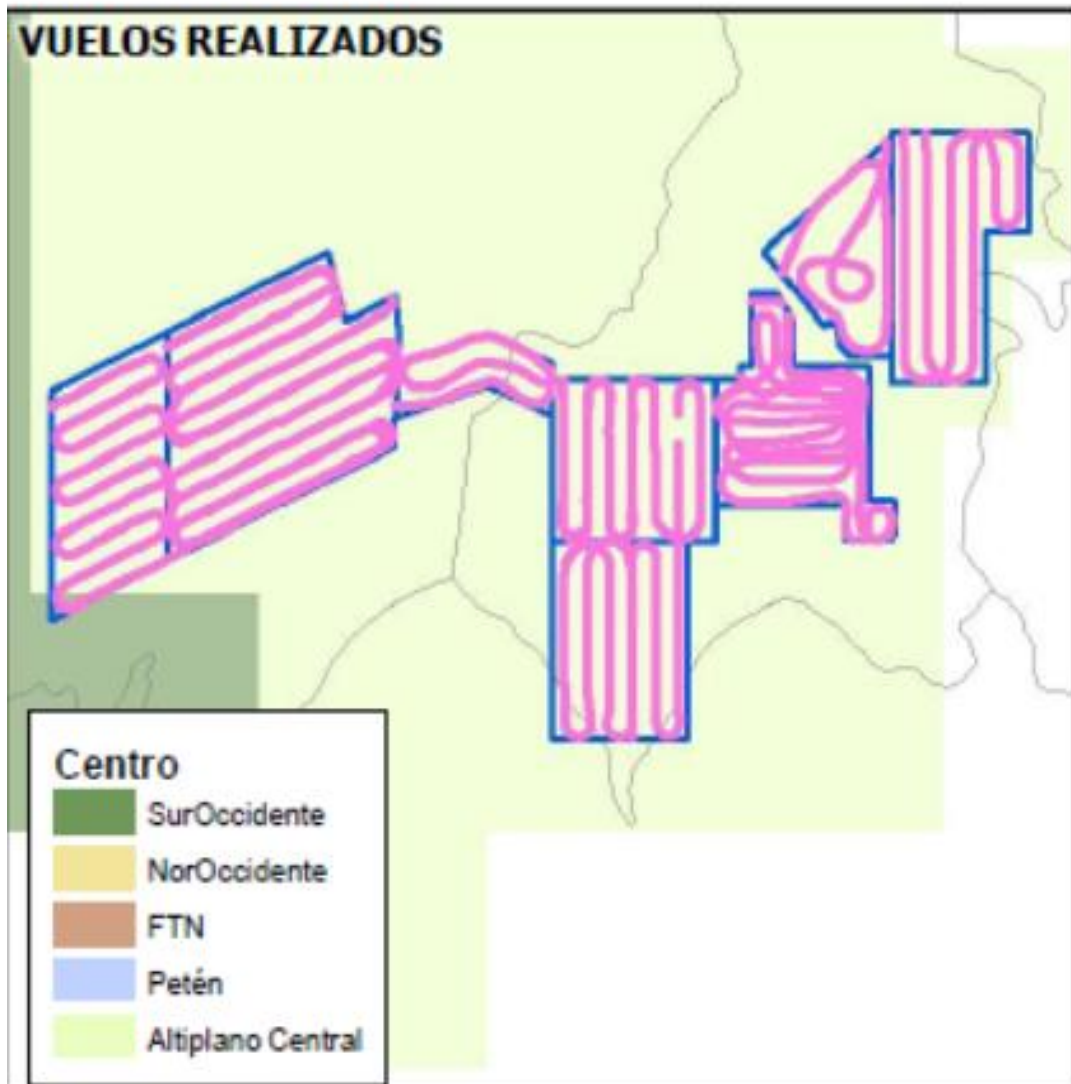


Figure 13. Monitoring of aerial release flights at each block as well as their release pattern.

#### 4.4. Spreadsheet calculator

##### 4.4.1. Single block - $FTD_{(s)}$ & $(w)$ calculations block one (similar to single block calculations two and three)

This page is in its design and operation similar to the pages for single block FTD calculations two and three (see Fig. 6). *These pages were prepared to simulate an action programme operation with a minimum of three blocks but more can be added, as needed, and linked as a standard excel file operation.* The information entered here allows calculating for the whole year (on a weekly basis) the fly/trap/day index for the wild and sterile adult populations based on the size of the user Programme release blocks (as defined in Fig. 14) the number of traps deployed, number of insects captured. Based on the sterile and wild FTD ( $FTD_s/FTD_w$ ) it also calculates weekly field ratios S:W achieved by the programme operations. The spreadsheet transfers the information to the page named “Release density calculations”. Field data needs to be entered in those cells that are showing numbers in red (Fig. 14).

| RELEASE DENSITY CALCULATIONS BLOCK ONE |                                       |                        |                             |                 |                       |             |                     |             |         |      |
|--|---------------------------------------|------------------------|-----------------------------|-----------------|-----------------------|-------------|---------------------|-------------|---------|------|
| RELEASE BLOCK # ONE                    |                                       |                        |                             |                 |                       |             |                     |             |         |      |
| WEEK #                                 | RELEASE BLOCK SIZE (km <sup>2</sup> ) | STERILE FLIES CAPTURED | # TRAPS PER km <sup>2</sup> | NUMBER OF TRAPS | DAYS OF TRAP EXPOSURE | FTD STERILE | WILD FLIES CAPTURED | FTD WILD    | RATIO   |      |
|  |                                       |                        |                             |                 |                       |             |                     |             | STERILE | WILD |
| 1                                      | 100                                   | 640                    | 5                           | 500             | 7                     | 0.182857143 | 2                   | 0.000571429 | 320     | 1    |
| 2                                      | 100                                   | 70                     | 5                           | 500             | 7                     | 0.02        | 1                   | 0.000285714 | 70      | 1    |
| 3                                      | 100                                   | 82                     | 5                           | 500             | 7                     | 0.023428571 | 1                   | 0.000285714 | 82      | 1    |
| 4                                      | 100                                   | 45                     | 5                           | 500             | 7                     | 0.012857143 | 0.25                | 7.14285E-05 | 180     | 1    |
| 5                                      | 100                                   | 52                     | 5                           | 500             | 7                     | 0.014857143 | 1                   | 0.000285714 | 52      | 1    |
| 6                                      | 100                                   | 47                     | 5                           | 500             | 7                     | 0.013428571 | 1                   | 0.000285714 | 47      | 1    |
| 7                                      | 100                                   | 32                     | 5                           | 500             | 7                     | 0.009142857 | 1                   | 0.000285714 | 32      | 1    |
| 8                                      | 100                                   | 55                     | 5                           | 500             | 7                     | 0.015714286 | 0.25                | 7.14285E-05 | 220     | 1    |
| 9                                      | 100                                   | 60                     | 5                           | 500             | 7                     | 0.017142857 | 0.25                | 7.14285E-05 | 240     | 1    |
| 10                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 0.25                | 7.14285E-05 | 192     | 1    |
| 11                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 1.25                | 0.000357143 | 38      | 2    |
| 12                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 2.25                | 0.000642857 | 21      | 3    |
| 13                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 3.25                | 0.000928571 | 15      | 4    |
| 14                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 4.25                | 0.001214286 | 11      | 5    |
| 15                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 5.25                | 0.0015      | 9       | 6    |
| 16                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 6.25                | 0.001785714 | 8       | 7    |
| 17                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 7.25                | 0.002071429 | 7       | 8    |
| 18                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 8.25                | 0.002357143 | 6       | 9    |
| 19                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 9.25                | 0.002642857 | 5       | 10   |
| 20                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 10.25               | 0.002928571 | 5       | 11   |
| 21                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 11.25               | 0.003214286 | 4       | 12   |
| 22                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 12.25               | 0.0035      | 4       | 13   |
| 23                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 13.25               | 0.003785714 | 4       | 14   |
| 24                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 14.25               | 0.004071429 | 3       | 15   |
| 25                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 15.25               | 0.004357143 | 3       | 16   |
| 26                                     | 100                                   | 48                     | 5                           | 500             | 7                     | 0.013714286 | 16.25               | 0.004642857 | 1       | 17   |

**Figure 14.** Single block fly/trap/day (FTD) calculation page. The numbers entered allow determining field effectiveness in terms of ratios sterile to wild (S:W).

If no wild flies are caught, a wild capture of 0.25, 0.50, and 1.00 flies per release block is assumed for free, low prevalence and suppression areas, respectively. A zero value for wild flies will prevent the Excel from computing a recommended sterile fly density. In scenarios such as the preventative release programmes (PRP's) where no wild flies are captured most of the time, it is convenient to assume a capture based on the "rate of approach". For specific areas a number of detections is conveniently assumed in order to continuously maintain an effective release of sterile males over the target area. The assumed capture "rate of approach" of 1 fly/month is equivalent to 0.25 flies/week in the free area; for low prevalence 2 flies/month (0.5 flies/week) and 4 flies/month (1 fly/week) in the suppression area.

#### 4.4.2. Release density calculations block one (similar to two and three)

This page is in its design and operation similar to the pages for release density calculations blocks two and three (see Fig. 6). *These pages were prepared to simulate an action programme operation with a minimum of three blocks but more can be added, as needed, and linked as a standard excel file operation.* This page allows calculating the required weekly release density per release block. The information required is the previous week release rate (sterile male flies released per unit area - ha, km<sup>2</sup>, miles<sup>2</sup>, acres) as well as the ratio S:W that the operational programme aims at (Fig. 15).

| RELEASE DENSITY CALC BLOCK ONE |                         |  |  |               |                    |         |      |                    |                                  |  |
|--------------------------------|-------------------------|--|--|---------------|--------------------|---------|------|--------------------|----------------------------------|--|
| RELEASE BLOCK # ONE            |                         |  |  |               |                    |         |      |                    |                                  |  |
| WEEKS                          | RELEASE RATE            |  | TOTAL INSECTS RELEASED/Km <sup>2</sup> | ♂ FTD sterile | ♂ FTD wild/fertile | RATIO   |      | REQUIRED S:W RATIO | NEW RELEASE RATE/Km <sup>2</sup> | NEW RELEASE RATE STERILE INSECTS/HECTARE |
|                                | STERILE INSECTS/HECTARE |  |  |               |                    | STERILE | WILD |                    |                                  |  |
| 1                              | 400                     |  | 40,000                                 | 0.183         | 0.006              | 320.0   | 1.0  | 100                | 12,500                           | 125                                      |
| 2                              | 500                     |  | 50,000                                 | 0.020         | 0.0003             | 70.0    | 1.0  | 100                | 71,429                           | 714                                      |
| 3                              | 500                     |  | 50,000                                 | 0.023         | 0.0003             | 82.0    | 1.0  | 100                | 60,976                           | 610                                      |
| 4                              | 500                     |  | 50,000                                 | 0.013         | 0.0001             | 180.0   | 1.0  | 100                | 27,778                           | 278                                      |
| 5                              | 500                     |  | 50,000                                 | 0.015         | 0.0003             | 52.0    | 1.0  | 100                | 96,154                           | 962                                      |
| 6                              | 500                     |  | 50,000                                 | 0.013         | 0.0003             | 47.0    | 1.0  | 100                | 106,383                          | 1,064                                    |
| 7                              | 500                     |  | 50,000                                 | 0.009         | 0.0003             | 32.0    | 1.0  | 100                | 156,250                          | 1,563                                    |
| 8                              | 500                     |  | 50,000                                 | 0.016         | 0.0001             | 220.0   | 1.0  | 100                | 22,727                           | 227                                      |
| 9                              | 500                     |  | 50,000                                 | 0.017         | 0.0001             | 240.0   | 1.0  | 100                | 20,833                           | 208                                      |
| 10                             | 500                     |  | 50,000                                 | 0.014         | 0.0001             | 192.0   | 1.0  | 100                | 26,042                           | 260                                      |
| 11                             | 500                     |  | 50,000                                 | 0.014         | 0.0004             | 38.4    | 1.0  | 100                | 130,208                          | 1,302                                    |
| 12                             | 500                     |  | 50,000                                 | 0.014         | 0.0006             | 21.3    | 1.0  | 100                | 234,375                          | 2,344                                    |
| 13                             | 500                     |  | 50,000                                 | 0.014         | 0.0009             | 14.8    | 1.0  | 100                | 338,542                          | 3,385                                    |
| 14                             | 500                     |  | 50,000                                 | 0.014         | 0.0012             | 11.3    | 1.0  | 100                | 442,708                          | 4,427                                    |
| 15                             | 500                     |  | 50,000                                 | 0.014         | 0.0015             | 9.1     | 1.0  | 100                | 546,875                          | 5,469                                    |
| 16                             | 500                     |  | 50,000                                 | 0.014         | 0.0018             | 7.7     | 1.0  | 100                | 651,042                          | 6,510                                    |
| 17                             | 500                     |  | 50,000                                 | 0.014         | 0.0021             | 6.6     | 1.0  | 100                | 755,208                          | 7,552                                    |

**Figure 15.** Release density calculations page. Operational data need to be entered in cells with red numbers.

#### 4.4.3. Sterile insect release density S: W ratio for all blocks

This page allows adding up the amounts of sterile insects required in each of the release blocks (Fig. 16). It also generates a total quantity of sterile insects required/week by the action programme field operations. This spreadsheet has three blocks calculation pages but more can be added by simply duplicating the pages and changing the block names as required by the user.

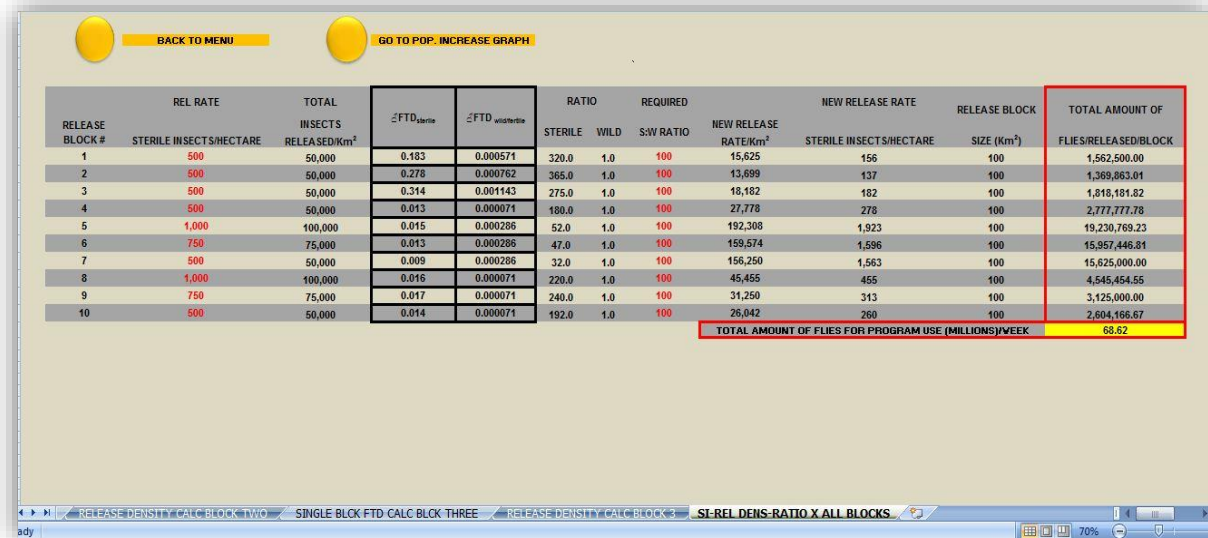


Figure 16. Total amount of insects required/week by an action programme, based on the number of release blocks in operation.

#### 4.5. Population increase graph - graphic analysis

Summary of all the graphs generated in the pages of the spreadsheet (Fig. 17 and Fig. 18). It shows population growth with and without control activities, graphs that show sterile insect performance/release block and the summary of insect needs for all blocks which can help managers to move sterile insects between blocks as required.

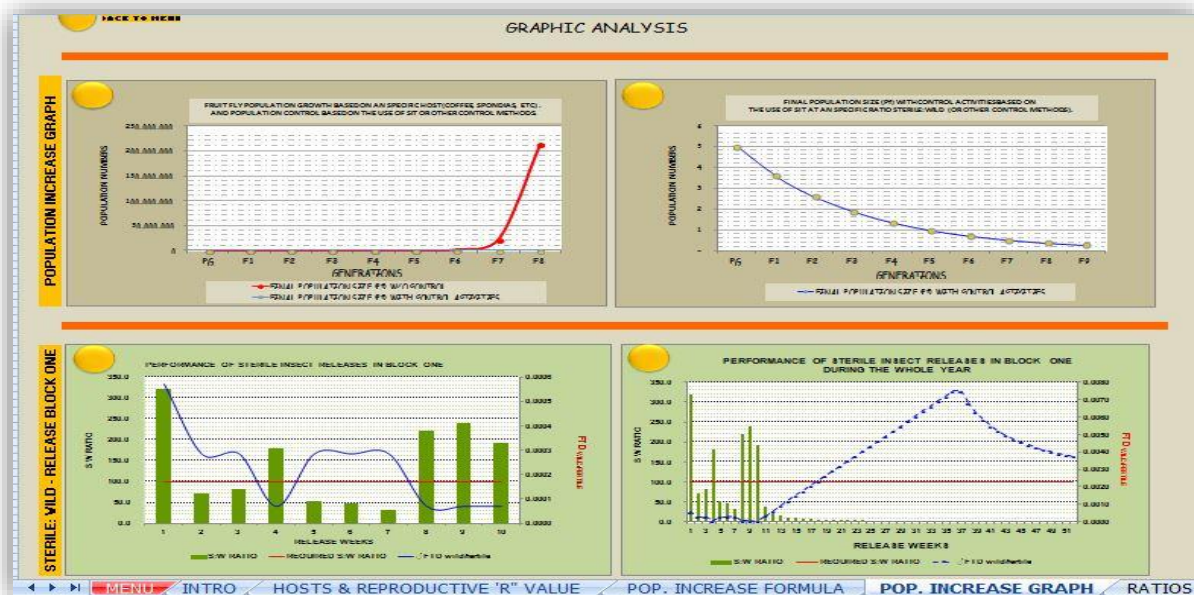
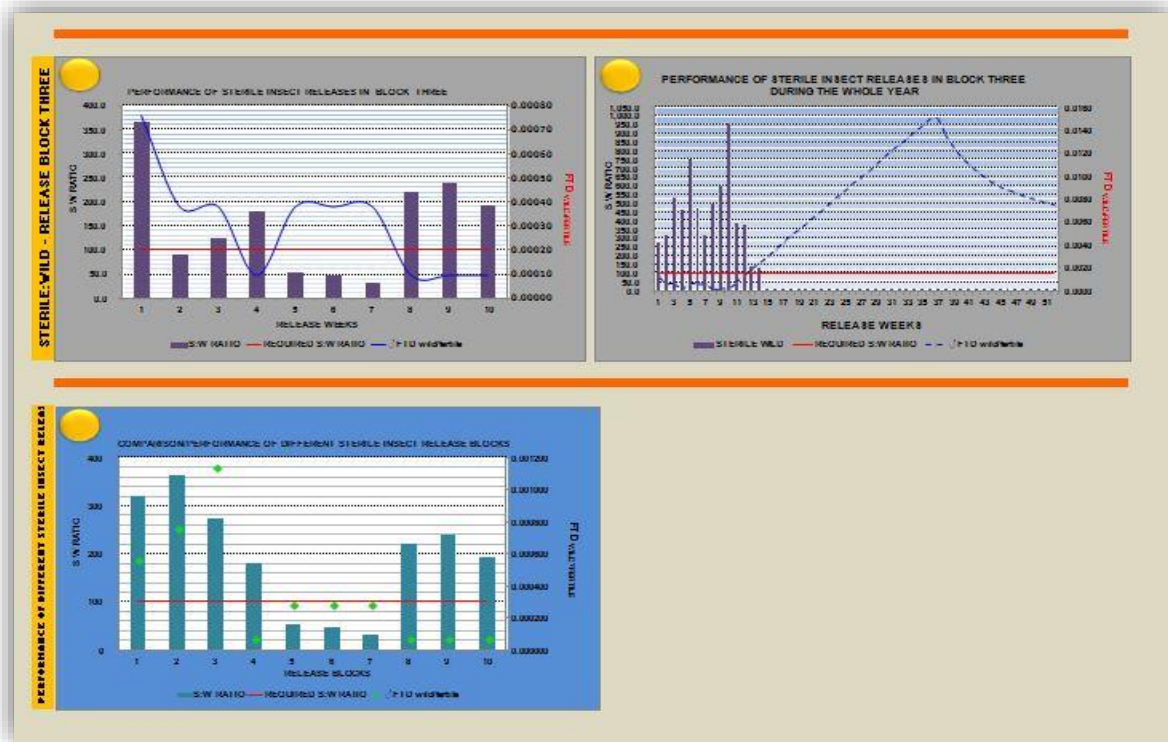


Figure 17. Graphical display of sterile insect release impact and performance when controlling pest populations.





**Figure 18.** Graphical display of the performance of sterile insect in each block and the analysis of total insects required/week for all blocks under action programme release.

**ACKNOWLEDGEMENTS:**

To USDA/APHIS/PPQ/CPHST and USDA/APHIS/IS for their support and the opportunity provided to validate the use of this “Sterile Insect Release density calculations” spreadsheet in its operational/action “Moscamed Regional Programme”.



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**Annex 1.** Factors that could affect sterile insect field presence, and their control effectiveness.

| Biotic conditions                    | Comments  |
|--------------------------------------|---|
| Insect Age at release time           | Ideally insects should be released as close as possible to their mating age.  |
| Feeding status at release time       | Ideally insects should be well fed and hydrated at the time of their field release.   |
| Insect longevity                     | Ideally, insects should have a good field survival to perform their control activity.   |
| Sexual preconditioning               | If available, this practice has proved to be advantageous.  |
| Insect/Strain fitness                | This should be of interest to the rearing facility to deliver competitive insects to release facilities/field operations.                   |
| Insect predation                     | Needs to be assessed in particular in relation to the release methodology and strain fitness.   |
| Abiotic conditions                   |   |
| Sterile insect release density       | Calculated based on actual sterile insect performance or empirical.   |
| Insect distribution – release method | Ground or aerial releases vary in their insect field placement /distribution. Wild populations are clumped as opposed to evenly distributed |
| Insect ratio sterile: wild           | Different insect species vary in their sterile: wild ratios for control.  |
| Vegetation type/density              | High/low host density affects overall wild population presence and sterile requirements and distribution.                                   |
| Environmental conditions             | Temperature, humidity, rain, snow, have effects on insect population  |
| Topographic condition                | Conditions will dictate release methods. Irregular topography vs large flat surface areas   |
| Release speed and altitude           | Fast/slow, high or low release conditions affect insects landing on the target area.  |
| Insect release equipment             | Appropriate equipment should be used to avoid damage to the insects while conducting release operations.                                    |
| Insect recapture                     | Low or high recapture affect sterile insect release calculations.   |

**Annex 2.** The existing SIT programmes, their objective and actual sterile insect release densities are shown in the table below. Programmes that are initiating area-wide SIT operations should determine their required release densities considering the conditions under which activities will be conducted such as assessed wild FTDs, objectives of the programme (suppression, eradication, etc.) and established over-flooding ratios. Release densities for different fruit fly SIT programmes and their respective programme objectives

| Country            | Fruit fly species                           | Objective                  | Aerial Release Density (Male Flies*/Ha) | Main Host and Area Characteristics   |
|--------------------|---|----------------------------|---|--|
| Argentina          | Medfly ( <i>C. capitata</i> )               | Eradication<br>Prevention  | 500-3,000<br>250-1,500                  | Stone and soft fruit (peaches, plums, apples and others)/Oasis-Valleys with extreme high/low temperatures. |
| Australia          | Qfly ( <i>B. tryoni</i> )                   | Prevention<br>Eradication  | 1,000<br>Not available                  | Soft fruit (tomatoes)/stone (peaches, plums)/Flat and dry area.  |
| Brazil             | Medfly                                      | Suppression                | 1,000-2,000                             | Mango and grapes subtropical conditions in a valley  |
| Chile              | Medfly                                      | Prevention<br>Eradication  | 1,500-2,500<br>>3,000                   | Guava, mangoes/isolated valleys surrounded by mountains and desert.  |
| Guatemala          | Medfly                                      | Containment<br>Eradication | 5,000                                   | Continuous coffee, mixed host rural areas/coastal, valley and mountainous area.                            |
| Israel             | Medfly                                      | Eradication<br>Suppression | 1,000                                   | Citrus and urban backyard hosts  |
| Japan (Okinawa)    | Melon fly ( <i>B. cucurbitae</i> )          | Prevention                 | Not available                           | Garden crops and urban backyard hosts  |
| Jordan             | Medfly                                      | Eradication                | 1,000                                   | Citrus and urban backyard hosts  |
| Mexico             | Medfly                                      | Eradication                | 2,000- 5,000                            | Continuous coffee, mixed host rural areas/coastal, valley and mountainous area.                            |
| Mexico             | Mexfly ( <i>A. ludens</i> )                 | Suppression                | 2,500                                   | Citrus, Guava, mangoes production areas/coast, oasis, mountainous area.                                    |
| Mexico             | West Indian fruit fly ( <i>A. obliqua</i> ) | Suppression                | 2,500                                   | Mangoes, coast and mountainous areas.  |
| Peru               | Medfly                                      | Eradication                | 1,000-2,000                             | Olives/oasis   |
| Portugal (Madeira) | Medfly                                      | Suppression                | 3,000-5,000                             | Mixed fruits and vegetables  |
| Spain              | Medfly                                      | Suppression                | 1,000-2,000                             | Citrus   |
| South Africa       | Medfly                                      | Suppression                | 1,200                                   | Grapes/isolated valleys – dry with irrigation  |

\*Adjusted for percent emergence, however, not for flying males.

|                |   |                           |                    |   |
|----------------|---|---------------------------|--------------------|---|
| Thailand       | Oriental fruit fly ( <i>B. dorsalis</i> ) | Suppression               | 5,000              | Pilot areas of mango orchards with no isolation                       |
|                | Guava fruit fly ( <i>B. correcta</i> )    | Suppression               | 5,000              |   |
| USA California | Medfly                                    | Prevention<br>Eradication | 250<br>1,000       | Urban (Jungle) fruit and vegetables. Variable climate and topography. |
| USA Florida    | Medfly                                    | Prevention<br>Eradication | 500<br>1,000-1,400 | Citrus and urban host/Coastal area, tropical.                         |
| USA Hawaii     | Melon fly                                 | Suppression               | Not available      | Experimental- Tropical, melon, squash.                                |
| USA Texas      | Mexfly                                    | Suppression               | 650                | Citrus and urban host/semi-arid with irrigation                       |