



Universidade dos Açores

Departamento de Ciências Agrárias

Development of decision support tools for implementing mitigation strategies of Medfly populations

Reinaldo Macedo Soares Pimentel
Doutoramento em Ciências Agrárias

Orientador

Professor com Agregação Doutor David João Horta Lopes

Coorientadores

Professor Catedrático Doutor António Maria Marques Mexia (ISA/UTL)

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Abstract

The Medfly *Ceratitis capitata* Wiedemann is one of the most important threats to the trade of fresh fruits in the world due to its high potential damage to fruits and ability to survive in a wide range of geographic and climatic conditions.

Considering all the research already done in the Azores, mainly in Terceira Island, about the Medfly, it is with the perspective of addressing area-wide control program problems, related to Medfly aggregation and spatial distribution, that the main goals of the present work are to analyse the population dynamics of Medfly in Terceira and São Jorge Islands (Azores archipelago); evaluate the impact of long term attractive baits (1 year) against populations of Medfly; evaluate if there is any significant relation between the abundance of wild adults of Medfly and the spatial characteristics of a target location; and evaluate the efficiency of three geospatial methods to estimate flies trapped per day (FTD) values for non-sampled areas in Terceira Island.

The population dynamics studies revealed a sex ratio of males/females in São Jorge Island significantly lower than observed on Terceira Island and also revealed population dynamics are generally linked with host fruit availability and abundance. But the scale of population peaks or trap records are not reliable. In Terceira Island, under certain conditions, there was Medfly infestations in some fruits (e.g. Tobacco Weed fruit) while there were no trap captures at the same time.

The evaluation of long term bait stations showed a statistically significant difference between having and not having the long term attractive baits during all seasons when controlling Medfly females. Also fruit infestation levels show a significant (p-value of 0.02) difference between treated and control plots, and in general the results indicate that attractive baits are promising for wide application on Terceira Island. A challenge for area-wide control in the Azores is the small size of orchard parcels and the presence of numerous wild host reservoirs near orchards.

The evaluation of a relationship between the abundance of wild adults of Medfly and the spatial characteristics revealed that the Ordinary Least Square (OLS) method cannot

provide a satisfactory general explanation for abundance of both genders of wild adults, yet it might generate some hypotheses about the fruit seeking behaviour of wild adult females of *C. capitata*. Only the GWR analysis provided a satisfactory general explanation for abundance of both genders of Medfly. However, both methods suggest that males are more widely scattered than females, and because of that they might play an important role in scouting the surroundings for fruit hosts.

As for the evaluation of the efficiency of three geospatial methods to estimate FTD values for non-sampled areas, results demonstrated that the GWR method is capable of predicting hotspots for the next season and can be used to identify ecological corridors over a non-sampled area. The high spatial heterogeneity and topographical conditions present on Terceira Island may explain why a more mathematically complex method is more reliable than simpler methods for use in possible future wide-area control programs for Medfly control.

Keywords: *Ceratitis capitata*; medfly; population dynamics; sex-ratio; fruit infestation levels, Lufenuron, spatial regression, Geographic Weighted Regression, Inverse Distance Weighted, Ordinary Kriging;

Resumo

A Mosca-do-Mediterrâneo (*Ceratitis capitata* Wiedemann) é uma das principais ameaças para o comércio de frutas frescas por todo o mundo devido ao impacto económico negativo na qualidade dos frutos e à capacidade de adaptação desta a uma ampla variedade de condições geográficas e climáticas.

Considerando-se todos os trabalhos já realizados nos Açores, principalmente na Ilha Terceira sobre esta problemática, com a perspetiva de resolver problemas operacionais que normalmente surgem nos programas de controlo integrado da mosca-do-Mediterrâneo, nomeadamente os relacionados com a agregação desta praga e sua distribuição espacial, que os principais objetivos do presente trabalho foram: analisar a dinâmica populacional da Mosca-do-Mediterrâneo nas Ilhas Terceira e São Jorge (Açores); avaliar o impacto de estações esterilizadoras de longa duração (1 ano) contra as suas populações adultas; avaliar se existia qualquer relação significativa entre a abundância de adultos selvagens de mosca-do-Mediterrâneo e as características espaciais de um determinado local a tratar; e avaliar a eficiência de três métodos de análise geomática para estimar valores de moscas por armadilha, por dia, para áreas não fora do âmbito de amostragem e para as condições da Ilha Terceira e da ilha de S. Jorge.

Os estudos de dinâmica populacional revelaram, na Ilha de São Jorge, uma proporção sexual dos machos / fêmeas significativamente menor do que o observado na Ilha Terceira e em termos de dinâmica populacional revelou também que as dinâmicas populacionais nas duas Ilhas estudadas estão geralmente relacionadas com a disponibilidade e abundância de frutos nos seus hospedeiros. No entanto, o número de picos populacionais ou registos de capturas poderão não ser uma base fidedigna para a tomada de decisão por parte do produtor. Na Ilha Terceira, em determinadas condições, foram observadas infestações de mosca-do-Mediterrâneo em alguns frutos (por exemplo, frutos *Solanum mauritianum*), enquanto, em simultâneo neste hospedeiro não houve quaisquer registos de adultos.

A avaliação da utilização no campo das estações esterilizadoras de longa duração mostrou uma diferença estatisticamente significativa entre ter e não ter estes dispositivos

durante todo o período de estudo. O nível de infestação de frutos demonstra uma diferença significativa (valor-p de 0,02), entre as parcelas tratadas e as de controlo, sendo que, em geral, os resultados obtidos neste estudo indicaram que estes dispositivos parecem ser promissores para a sua aplicação a larga escala na ilha Terceira. No entanto, o recurso a estes dispositivos representa um desafio para o controlo em larga escala, pois as parcelas de pomares são pequenas e em menor quantidade quando comparadas com os numerosos reservatórios silvestres desta praga existentes na vizinhança das parcelas frutícolas. A avaliação de uma relação entre a abundância de adultos selvagens da mosca-do-Mediterrâneo e as características espaciais revelou que o método de mínimos quadrados ordinários não é capaz de fornecer uma explicação geral e satisfatória para a abundância populacional dos adultos de ambos os sexos. No entanto, pode gerar algumas hipóteses sobre o comportamento de fêmeas adultas selvagens na procura de alimento. Só a análise Regressão Geograficamente Ponderada (GWR) forneceu uma explicação geral satisfatória para a abundância de ambos os sexos. No entanto, ambos os métodos sugerem que os machos se dispersam mais amplamente do que as fêmeas, e que eles poderão desempenhar um papel importante na deteção de hospedeiros.

No que diz respeito à avaliação da eficiência de três métodos geográficos utilizados neste estudo para estimar os valores mosca por armadilha por dia para áreas não incluídas na amostragem, os resultados demonstraram que o método da Regressão Geograficamente Ponderada (GWR) foi o único capaz de prever a localização de focos de elevada densidade populacional na época seguinte. Por isso, poderá mesmo ser utilizado para identificar corredores ecológicos sobre uma área onde não se realizaram quaisquer amostragens. A alta heterogeneidade espacial e condições topográficas presentes na ilha Terceira poderão estar na base da explicação pelo qual um método matematicamente mais complexo é mais confiável, do que os métodos mais simples, e como tal deverá ser utilizado em futuros programas de larga escala de controlo da mosca-do-Mediterrâneo.

Palavras-chave: *Ceratitis capitata*; mosca do Mediterrâneo; Dinâmica populacional; rácio sexual; taxas de infestação de frutos, Lufenurão, Regressão espacial, Regressão Geograficamente Ponderada, Ponderação pelo Inverso da Distância, Kriging Ordinário;

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General introduction

The family Tephritidae is best known for its status as a pest group that attacks a wide array of commercial and wild fruits and vegetables in many different plant families. But just a few species of this family are actually commercial fruit infesters (Capinera 2008).

The Mediterranean fruit fly, or Medfly, (*Ceratitis capitata* Wiedemann (Diptera: Tephritidae)) is one of the major fruit fly pest species of quarantine importance. It is the most polyphagous tephritid and it infests fruits of plants belonging to more than 67 families. This condition of polyphagy makes this insect one of the most serious pests worldwide, with infestation levels reaching 100% in some of its hosts and that is why it is a quarantine pest in countries such as the United States of America and Japan (Papadopoulos 2008; Joint FAO/IAEA Programme 2013).

The Medfly adult is smaller than the domestic house fly. It measures 4–6 mm in length and 1–2 mm in width and unlike the domestic house fly, it has a very colorful appearance. The males are easily distinguished from females and by a pair of stalky, cornea like, pointed, black-gray expansions at the apex of the anterior part of their head. The wings are transparent with yellow, brown, and red stripes and each wing measures about 4.5 mm in length.

Genetic studies suggest that this insect is native to tropical Africa (Gasperi et al. 1991; Gasperi et al. 1997; Bohonak et al. 2001; Gasperi et al. 2002; Karsten et al. 2013), but due to the world fruit trade expansion, since the first ships transporting fruits worldwide, and its ability to adapt to new environments (Rossler 1986; Liquido & Shinoda 1991; Carvalho & Aguiar 1997; Pierre 2007; International Atomic Energy Agency & Joint FAO/IAEA Programme of Nuclear Techniques in Food and Agriculture 2008; Papadopoulos 2008; Sciarretta & Trematerra 2011; Radonjic et al. 2013), it has managed to spread invasively first to the Mediterranean area and then to many other parts of the world, including Australasia and North and South America (Joint FAO/IAEA Programme 2013).

Its high economic impact has made this pest one of the most studied in field and laboratory conditions around the world (Papadopoulos 2008). Because it's biological ability to adapt to new conditions, it is difficult to extrapolate the results from studies done in one country/island/locale to another, with different conditions.

However, population dynamics reported in some studies can be very similar even under different conditions. For instance, Radonjic et al. (2013) studied the seasonal occurrence of *Medfly* on the island of Chios (Greece) (approximately 38° North latitude) and their results also show certain similarities with patterns presented by Pimentel (2010) for Terceira Island (38° 37'N). Both authors found that the peak populations of Medfly are normally observed from mid-September to the end of November each year and normally an absence of captures of flies is observed from February through May/June.

Until recent years, area-wide control programs used the argument that insect pests had to be controlled using an unreliable degree of broad-spectrum insecticides. However, the indiscriminate use of these chemicals as a control tactic is no longer sustainable in view of increased development of resistance, pollution of soils and surface water, residues in food and the environment, representing risks to human health and biodiversity (Hendrichs et al. 2005; Nagel & Peveling 2005; Hendrichs et al. 2007; Vreysen et al. 2007).

With the publication of the European Directive 2009/128/EC, agrochemical manufactures were stimulated to develop new products that are more environmentally friendly or alternative non-chemical approaches or techniques. One of these new approaches is the use of bait stations containing phagostimulant bait that incorporates a chemosterilant. According to laboratory tests of Casaña-Giner et al (1999), lufenuron showed good potential for the control of *C. capitata* because it caused failure of larva to hatch from eggs laid by females either from ingesting it or getting it sexually transmitted by males that have eaten this insect growth regulator agent.

The promising success of this new technique (Navarro-Llopis et al. 2004; Navarro-Llopis et al. 2007; Bachrouch et al. 2008; Di Franco et al. 2008; Navarro-Llopis et al. 2010; Moya et al. 2010) has led Navarro-Llopis et al. (2011) to try this method along with the sterile insect technique (SIT) and found that plots exposed to this combination got a significantly greater reduction of pest population compared to other plots only exposed to SIT or control plots.

The SIT is based on a Trojan-horse technique which consists of a mass release of sterile males, of the target specie, over the area to be treated, and according to Enkerlin (2005) the SIT is most widely applied against tephritid fruit flies. The concept of this

technique was first introduced by Knippling (1955) as a means of controlling and/or eradicating insect populations described as agricultural pests. Since its introduction it has been widely applied against many species of Lepidoptera and Diptera.

This technique has three obvious advantages. The first advantage has to do with the fact that no chemical or exotic species are being introduced in the ecological system, and therefore it is environmentally friendly. The second advantage has to do with the species specificity of the targeted species and the third advantage has to do with the intrinsic characteristics of the species which will make the released insects track and aggregate with the wild populations. However, the SIT is only effective when integrated on an area-wide basis, addressing the total population of the pest, irrespective of its distribution (Hendrichs et al. 2002). Also SIT depends largely on the ability of sterile flies to spread in the target area and compete with the wild males for wild females. This necessity could be well improved if the sterile insects were to be released at the locations where the wild population is most abundant. But, according to Hendrichs et al. (2007) the ecological heterogeneity within field, within farm, and at broader spatial scales profoundly affects the population dynamics of pests.

There are several geostatistical methods usable for area-wide integrated pest management (AW-IPM) programs, but entomologists frequently use spatial interpolation methods to characterize the spatial distribution of pests. Two of the most commonly used methods in insect studies are Inverse Distance Weighted (IDW) (Lopes et al. 2005; Lopes, Pimentel, Dantas, et al. 2006; Pimentel et al. 2006; Bonsignore et al. 2008; Lopes et al. 2008; Lopes, Macedo, et al. 2009; Lopes et al. 2010; Pimentel 2010; Sciarretta & Trematerra 2011; Rhodes et al. 2011) and Ordinary Kriging (OK) (Alemany et al. 2006; Sciarretta et al. 2008; Tabilio et al. 2014). Both methods rely on Tobler's first law of geography, which states that things that are close are more related than things that are further away (Tobler 1970). But both methods are limited and while the trap information is the foundation of the spatial analyses and population dynamic studies of most fruit fly programs, it is equally important to have key ecological or geographic layers of information that may explain the presence, population dynamics, and movement of fruit flies (Nestel et al. 2004). According to Midgarden et al. (2014), the specific layers that are important for a given program depend on the geography and climate of the area, the tephritid species, and the objectives of the

program. Layers potentially important for tephritid fruit flies include host presence, land use, organic waste sites, altitude, transportation, soil type, temperature, and rainfall as well as areas that might presumably preclude fly presence (e.g., deserts, bodies of water, large areas of field crops, or pastureland). For instance, by methodically analysing Medfly metadata catches in California, Carey (1996) concluded that mountain topography, valleys, rivers, and shorelines may channel dispersal of an established population through paths of least resistance.

One of the biggest challenges when trying to forecast Medfly spatial distribution, over any area, is its natural condition to aggregate in several patches over an area. This aggregation could come from numerous conditions. One of these conditions, for example, could be related to the green leaf volatiles that can enhance the attraction responses of male pheromone and therefore attract more of the females. Other example as a potential inducer of pest aggregation could be related to plant kairomones (Tan 2000; Nestel et al. 2004; Quilici et al. 2014).

Since the settlement of the Azorean Islands horticulture has been an engine of economic development for the region, enhancing its relationship with foreign markets, for example through cultivation of oranges (Lopes, Pereira, et al. 2006).

The Azorean Archipelago is composed of 9 Islands and the impact of *C. capitata* on Azorean fruit production goes back to 1829, when it was first reported in the Azorean Archipelago (Quaintance 1912). The impact of this insect, at that time, on the Azorean orange production was such that at least one third of the shipped oranges reaching the British market were in bad condition. The successive losses of the orange fruit merchandise, condemned, among other reasons, the shipment to the United Kingdom and therefore the Azorean orange production economic sustainability (Quaintance 1912; Meneses 2006).

Forjaz et al. (2003) did the first, reported, study about Medfly in the Azorean Archipelago, and it was done on Terceira Island. Afterwards, within the activities and objectives of the research projects (MAC/3.1/A1) Interfruta (2004-2006), (MAC/3.1/A4) Interfruta II (2006-2009), more elaborated studies about *C. capitata* including high density locations, dispersion and fruit infestation were carried out (Pimentel et al. 2005; Lopes, Pimentel, Nunes, et al. 2006; Lopes, Pimentel, Dantas, et al. 2006; Pimentel 2010; Pimentel

et al. 2014). The research project (MAC/3/A163) CABMEDMAC (2010-2014) was initiated as a direct result of these projects that were all developed on Terceira Island. This research project had a principal goal to get better knowledge about fruit flies throughout Macaronesia. In the Archipelago of the Azores, the CABMEDMAC project developed its activities on São Jorge and Terceira Islands.

The (MAC/3.1/A1) Interfruta (2004-2006) research project encouraged other technicians in other Islands, mainly in the biggest island of the Azores Archipelago (São Miguel) to also study (Medeiros et al. 2007; Medeiros & Oliveira 2009) and try to find biological ways to control the Medfly (Oliveira et al. 2008; Tavares 2012).

Considering all the research work already done in the Azores, mainly on Terceira Island, about the Medfly, and the perspective of addressing area-wide control program problems related to Medfly aggregation and spatial distribution, the main goals of the present work are to:

1. Analyse the population dynamics of Medfly on Terceira and Sao Jorge Islands (Azores archipelago).
2. Evaluate the impact of long term attractive baits (1 year) as a featured protection method against populations of Medfly.
3. Evaluate if there is any significant relation between the abundance of Medfly adults and the spatial characteristics of a target location.
4. Evaluate the efficiency of three geospatial methods to estimate FTD values for non-sampled areas in Terceira Island.