Microwave treatment for pest control: the case of *Rhynchophorus ferrugineus* in *Phoenix canariensis*

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Rhynchophorus ferrugineus (red palm weevil) is currently one of the major threats to palms in the Mediterranean area. No single technique developed up to now seems able to completely eradicate this pest. This paper introduces microwave treatment as an effective tool to help the control of this insect; the effect of microwave radiation on palm tissues and on the curculionid is described and discussed. The main advantage of microwaves is their eco-compatibility, and preliminary results of their application to *Rhynchophorus ferrugineus* control are very promising.

Introduction

The invasive red palm weevil, *Rhynchophorus ferrugineus* (Olivier 1790) (Coleoptera: Curculionidae) (listed on the EPPO A2 list of pests recommended for regulation, and subject to an EPPO Diagnostic protocol PM 7/83, EPPO Code RHYCFE), has become the major pest of palms in the Mediterranean area, where it has spread slowly during the mid-1990s and very quickly during the last 5 years. The infested countries in the Mediterranean and the year of first record are reported in Table 1.

This pest is currently widely distributed in Oceania, Asia (Yuezhong *et al.*, 2009), Africa and Europe. On 2010-10-18, the detection of *R. ferrugineus* in California (USA) was officially confirmed (EPPO 2010).

Red palm weevil in Italy

Rhynchophorus ferrugineus was found in Italy for the first time in 2004, on some plants of *Phoenix canariensis* located in a nursery next to Pistoia (Tuscany). These plants were immediately destroyed and since then official records have not been reported in this region. At the end of 2005, the insect was reported in Sicily and in Campania. Since then it has quickly spread in areas where *P. canariensis* is present, resulting in rapid death of thousands of plants. In Table 2 data about the year of the first record and the number of palms destroyed due to this pest in the Italian regions are reported. These data are likely to underestimate the total number of specimens compromised by the curculionid. At present, although notification of the presence of attacked plants to the competent Phytosanitary Services is required, reports were often not made by the plant owners in order to avoid official measures required to eradicate the pest. Some owners preferred less expensive actions which do not prevent the spread of the *R. ferrugineus*.

This reluctance of plant owners to comply with the notification process and collaborate with the Phytosanitary Services has strongly contributed to the proliferation of the insect, but other reasons can be identified:

- the lack of precise knowledge of ethology of red palm weevil in these areas;
- the lack of adequate means to fight the pest, in particular in the first years of its presence;
- the particular aggressive behaviour of *R. ferrugineus* on *P. canariensis* compared to the other host plants in the areas of origin of the insect, where the pest can live within the host plant for some years;
- contiguity of susceptible plants;
- involvement of a large number of public and private owners whose feelings, regarding the problem has been (and still is) very heterogeneous.

Several control methods have been applied against this pest, within an integrated pest management strategy. Its main components are phytosanitation, which involves cutting down and burning infested palms, use of insecticides (Llácer & Jacas, 2010) and use of pheromone traps for adult monitoring and mass trapping (Guarino *et al.*, 2010).

Chemical control against *R. ferrugineus* is based mainly on the repeated application of large quantities of pesticides, which are applied in a range of preventive and curative procedures designed to limit and contain the spread of infestation by different applications [protecting wounds, soil application, spraying,

Table 1 Mediterranean basin countries and year of first record

Country	Year	EPPO Reporting Service first record
Albania	2009	2009/207
Cyprus	2006	2007/022
France	2006	2006/225
Jordan	1999	1999/078
Greece	2006	2006/226
Israel	1999	1999/119
Italy	2004	2006/001
Libya	2009	2010/205
Malta	2007	2010/204
Morocco	2008	2009/001
Portugal	2008	2008/022
Syria	2007	2007/002
Spain	1993	96/096
Turkey	2005	2007/001

localised direct injections into the palm trunk (stipe) and fumigation] (Faleiro, 2006).

As a consequence, there are major concerns about the environmental pollution caused by these treatments, especially in public areas where ornamental palms are grown.

In this context new research into and action plans concerning control are strongly recommended by EU commission (see Commission Decision, 2010). The development of a biocontrol strategy for *R. ferrugineus* (Esteban-Durán *et al.*, 1998) using a fungus (Güerri-Agulló *et al.*, 2010) or entomopathogenic nematodes (Atakan *et al.*, 2009), as well as the employment of physical methods (preventing entry of weevils through cut ends of petioles and wounds), use of attractants and other chemicals, have also been suggested (EPPO, 2008).

In the last few years a major effort has been made to find an efficient, cheap and eco-compatible method to control *R. ferrugineus* but, to date, a solution has not been found. The current emergency can only be managed by combining different techniques that need to be adapted and applied according to each particular case: video surveillance, preventive treatments with chemicals (sprays or injections), treatment by mechanical sanitation associated with chemicals or entomopathogens, destruction of the compromised plants etc.

In addition to the use of the aforementioned tools, research has focused on novel techniques and the use of microwaves is considered a particularly promising tool. The Campania Region, after observing good potential results obtained by the ‘‘Ecopalm Ring®’’ (Griffo *et al.*, 2009), a toroidal prototype machine proposed for this kind of treatment, has funded a specific research project to evaluate the effectiveness of the procedure. This project is strongly multi-disciplinarian involving agronomists, physicists, engineers and entomologists.

Background information on microwaves

Microwaves are electromagnetic fields that oscillate in the 300 MHz–300 GHz frequency range. The thermal effect of microwaves (microwave dielectric heating) is largely employed for the heating and processing of food and in production of industrial materials.

The idea of using microwaves against *R. ferrugineus* is based on the fact that microwave radiation should be able to induce a thermal increase in the pest, heating it to a lethal temperature, without harming the plant tissues.

The ability of microwaves to inducing heating in certain materials is due to the interaction mechanisms between the electromagnetic field and the molecules constituting the materials. In particular two parameters are commonly used for describing the electromagnetic properties of the materials of interest: the relative permittivity (ε), and the equivalent electric conductivity (σ). Relative permittivity is related to the ability of materials to ‘store’ electromagnetic energy and it is reported as a value relative to that of free space ($\varepsilon_0 = 8.86*10^{-12}$ F/m), equivalent electric conductivity measures the capacity of a material to dissipate the electromagnetic energy, transforming it into heat. For instance, the air has a σ close to zero, whereas distilled water, at 20°C and at 2.45 GHz, has $\sigma = 1.3$ S/m. Gabriel *et al.* (1998) and Nelson (1996) provide more detail on the basic theory underlying microwave dielectric heating. The current paper summarizes the main differences between conventional and microwave-induced heating.

Table 2 First record and the number of palms destroyed by Italian region

Regions	First record	2004	2005	2006	2007	2008	2009/2010	Totals
Abruzzo	2007	–	–	–	30	120	350	500
Calabria	2007	–	–	–	6	35	200	241
Campania	2005	–	8	241	5069	4272	3421	13011
Lazio	2006	–	–	30	120	204	713	1067
Liguria	2007	–	–	–	3	21	87	111
Marche	2007	–	–	–	20	63	439	522
Puglia	2006	–	–	34	725	867	2579	4205
Sardegna	2007	–	–	–	30	418	309	757
Sicilia	2005	–	73	120	4811	7506	6464	18974
Molise	2007	–	–	–	–	22	97	119
Basilicata	2010	–	–	–	–	–	1	1
Toscana	2004	3	–	–	–	–	–	3
Overall Italy	2004	3	81	425	10 814	13 528	14 660	39 511

In conventional heating (with an external heating source) the thermal increase is related to the heat flux into the material from the surface. The heat flux is proportional to the gradient of the temperature through the thermal conductivity; the thermal conductivity can be very low for non-metallic materials and very high surface temperatures are required to achieve the desired internal temperature.

In microwave dielectric heating, the mechanism involved for transforming the electromagnetic energy into heat is related to the nature of the molecules constituting the materials that, in presence of the electromagnetic field, can move, oscillate and/or rotate. In the Debye interpretation (Debye, 1929), the heat is generated by frictional forces occurring between a polar molecule, whose rotational velocity has been increased by coupling with the microwave radiation, and neighbouring molecules. To summarize, an electromagnetic wave impinging on a material will be partially reflected, but part of the radiation will penetrate in the material, transferring a percentage of its energy, that will be dissipated and converted into heat according to the electrical conductivity (σ).

The dissipation process subtracts power from the electromagnetic wave, that will attenuate more rapidly with greater electric conductivity. This behaviour corresponds to a heating source that is inside the material, instead of being outside it, and electromagnetic radiation can produce direct heating in the area where the electromagnetic field is able to propagate (volumetric heating), but the heat flux would successively spread in the rest of the material by means of thermal conduction. In this way heating of the material is similar to that from conventional heating, but with lower surface temperature and a more uniform temperature distribution. In addition, microwave energy can be extremely efficient in the selective heating of materials as no energy is wasted in 'bulk heating' the sample, if it has a different and lower conductivity.

Microwave applications have been designed for the volumetric heating of rubber, wood, paper, agricultural products, for food-processing and disinfestations (wheat, rice, fruit) (Fleurat-Lessard, 2001; Lewandowski, 2001; Wang & Tang, 2001; Vincent *et al.*, 2003). In recent years this technique has attracted particular interest, and is to be evaluated, by the International Plant Protection Convention (IPPC) for the treatment of wood packaging materials, according to ISPM15 *Regulation of Wood Packing Material in International Trade* of the FAO.

Microwave treatment of palms

This research aimed to obtain, by means of microwave heating, temperatures that are lethal for the different life stages of *R. ferrugineus,* without damaging the host plants. The basis for these studies is the knowledge of the electromagnetic characteristics (ε, σ) of the materials involved in the process (Nelson, 1996). These properties depend on several parameters, including frequency, temperature, humidity and, for the insect, its developmental stage (egg, larva, adult), as well as the modifications induced by the insect on the palm (damaged tissues). To our knowledge, these properties are not reported in literature, thus measurements were carried out on several *P. canariensis* specimens and on weevils (either larva or adult). Then, on the basis of the results obtained, a set of treatments of *P. canariensis* opportunely infested with *R. ferrugineus* was executed in order to test the effectiveness of microwaves for killing the pest.

Material and methods

Permittivity measurements

Wide band swept frequency permittivity measurements of the different tissues under investigation were carried out by means of the open-ended coaxial cable technique, a technique widely used for the characterization of biological materials (Gabriel & Peyman, 2006) and agricultural products (Wang *et al.*, 2003).

Measurements were carried out in the 0.5–20 GHz frequency range by using a coaxial truncated probe (RGU316, 3.5 mm diameter) connected to the vector network analyzer Anritsu 37347C. The probe, in direct contact with the material under test (Fig. 1), was employed for measurements of the reflection coefficient at the probe–material interface after a standard calibration procedure (Gabriel & Peyman, 2006). The complex permittivity was then obtained using a rational function model (Anderson *et al.*, 1994).

Fig. 1 Experimental set-up for permittivity measurements with the open ended coaxial cable technique.

Lethal temperature estimation

Data on the thermal death kinetics of target insects is required for the estimation of lethal temperature and time combinations. Different methods can be used to evaluate and analyse thermal resistance of insects. The application of hot air was here used to increase the ambient temperature to obtain a preliminary indication of the thermal limits of survival of the pest.

Rhynchophorus ferrugineus of various stages were reared in a controlled rearing room at the Department of Entomology and Agrarian Zoology, University of Naples. The room was maintained at 25 ± 2°C and 60–70% RH. The photoperiod was approximately 12:12 L:D. The room was also used as a media room for handling and preparing materials for artificial diets.

In order to identify the lethal temperature and time ranges, tests on 200 specimens were carried out at the Phytopathology laboratories of Campania Region in an electrically heated incubator. At least three replicates for each stage and for each temperature were carried out. Five stage groups were identified: four groups of larvae divided into different categories according to their weight (total range 2–6 g) plus one group of adults weighing about 1 g. Two specimens from each group were transferred into 9 cm petri dishes and then placed in an incubator (Fig. 2A,B) where the temperature was monitored with a thermocouple connected to an external thermometer. Observations taken each minute determined the time needed for the apparent death of the insect at fixed temperatures (at 60% R.H.). After exposure, adults and larvae were observed for 24 h to determine whether they were actually dead before mortality was recorded.

Microwave treatments of *P. canariensis*

After the preliminary analysis, tests were carried out treating some artificially infested plants with microwaves. The treatment was done in the greenhouses of Phytopathology laboratories of Campania Region on four *P. canariensis* (stipe diameter about 20 cm) 20 days after a controlled infection performed with four young larvae per plant (Fig. 3A,B).

The plants were exposed to microwaves, generated by a magnetron (2.45 GHz, 1 kW power), and the induced heating was evaluated by means of an infrared thermal camera (A40M IR Thermo Vision; Flir Systems, Wilsonville, Oregon, United States) connected to a computer for data acquisition both during and after microwave power application. The experimental set-up is shown in Fig. 4A; this facilitates microwave application in the field.

In order to be able to rapidly acquire the images of the internal temperature distribution, prior to the experiment the palms were cut on a horizontal section and the two halves were joined together again to reconstruct the original plant (Fig. 4B). This technique is typically used in order to evaluate the efficacy of a microwave applicators employed for hyperthermia (d'Ambrosio & Migliore, 2010) or material (Andreuccetti *et al.*, 1994) treatments. The same procedure was applied to three non-infested palms as a control.

During the testing the environmental temperature was about 10°C, and the palm was exposed for variable time intervals, ranging from 2 to 30 min. Great care was taken for the security of the operators and, regarding the exposure limits to the radiation, a value of the effective electric field <20 V/m, in the area where operators were present, was verified by means of the PMM30 radiation monitor and EP330 probe.

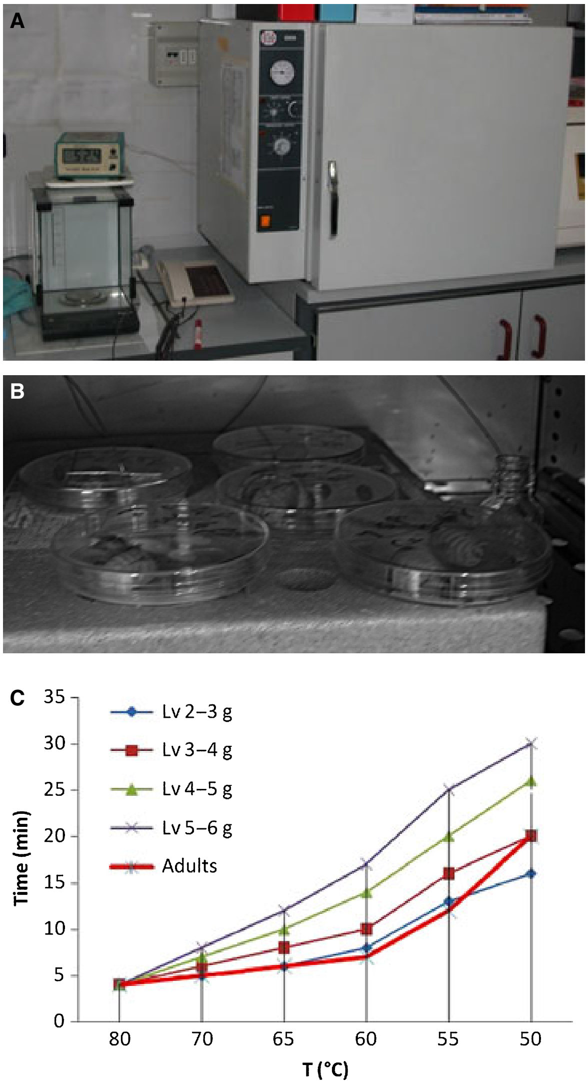

Fig. 2 (A) Incubator with thermometer; (B) Larvae in the incubator; (C) Time for 100% kill of larvae and adults at different temperatures LT100 (minutes).

Results

The estimation of the time to 100% kill of larvae and adults at a fixed temperature (in an incubator as described in Lethal temperature estimation) is shown in Fig. 2C. The adult insects were much more sensitive to heat than the larger larvae with 20 min at 50°C and only 4 min at 80°C causing adult death.

Lethal time for the larvae varies with weight and the most resistant were those who weighed between 4 and 6 g. The smallest larvae had a similar sensitivity to adults. The results are

Fig. 3 Greenhouses of Campania region; (A) plants under controlled infestation; (B) example of controlled infestation.

Fig. 4 Microwave treatment of palm; (A) experimental setup; (B) preliminary cut of the palm; (C) section of the palm with larva inside.

consistent with those observed for eggs by (Li *et al.*, 2010) that failed to survive at 40°C, being evidently more sensitive than the youngest larvae that was tested.

The average values of the electromagnetic parameters, measured at 2.45 GHz (the frequency typically used by high power microwave sources as magnetron) and at ambient temperature, are shown in Table 3. This presents the permittivity, equivalent conductivity and skin depth of the palm tissues, both healthy and damaged by insect (larva and adult) and, for comparison, values for distilled water (Nyshadham *et al.*, 1992) and pine wood (Andreuccetti *et al.*, 1994). It should be noticed that, in spite of the data reported for other kind of woods, both healthy and damaged palm tissues have high conductivities (σ), due to their high water content; consequently, the electromagnetic field would

Table 3 Electromagnetic properties of materials at 2.45 GHz

	Relative permittivity	Equivalent electric conductivity σ (S/m)	Penetration depth δ (cm)
Healthy palm wood (high density of fibres)	30.3	1.17	2.5
Healthy palm wood (low density of fibres)	46.0	1.40	2.6
Damaged tissue	50.7	2.93	1.3
Pine wood	3.1	0.068	13.4
Distilled water	73.6	1.29	3.5
Larva	35.3	1.04	3.1
Adult insect	9.3	0.38	4.3

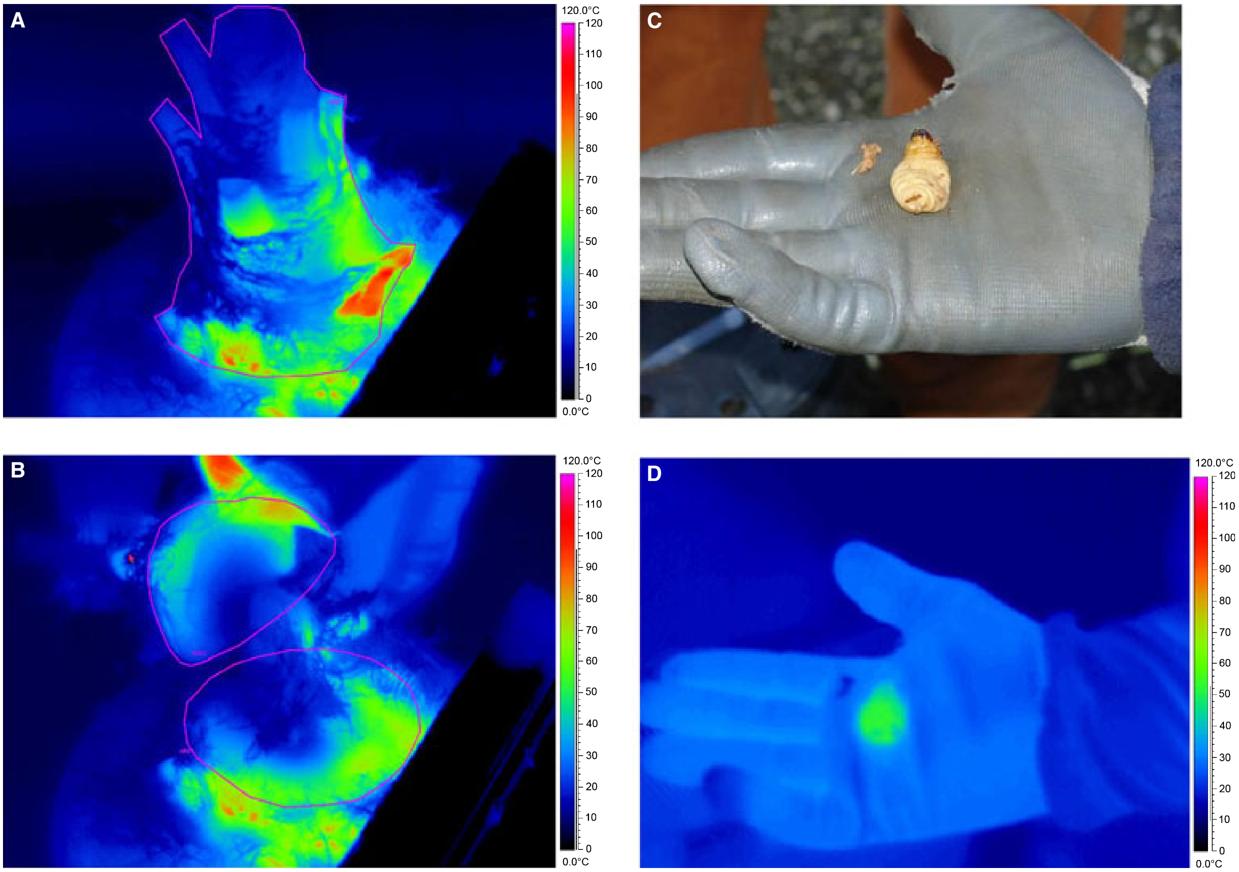

Fig. 5 (A) Thermogram of the external temperatures for the palm; (B) thermogram of the section of the palm; (C,D) photo and thermogram of a dead larva (temperature about 50°C) following microwave treatment.

rapidly decay once penetrated inside the palm tissue, and only the external section of the palm is heated directly by the microwaves.

Regarding the insects, both permittivity and conductivity are higher for larvae than adults and similar to that of palm healthy tissues.

In conclusion, the heating induced by microwave treatment will directly affect only the external part of the palm, while the inner zones will be subject to heating via thermal conduction. Therefore, the insects that are reached directly by the microwaves on the external part of the palm will rapidly increase their temperature, whereas the ones inside the tree will be subjected to a temperature increase by the thermal conduction from the surrounding environment. A transient region will occur where insects will be affected by both mechanisms.

The analysis of the data was confirmed by the results obtained on the infested palm which underwent microwave exposure. Figure 5A,B shows the thermograms obtained after 30 min of treatment and the histograms of the percentages of the temperature values measured in the marked areas. In spite of the relatively low power level employed (1 kW), the brief duration of the exposures (30 min) and the reduced radiated surface, it is possible to see that some areas of the external tissues have reached temperatures greater than 80°C, while in the inner tissues of the palm temperatures in the range 40–60°C have been observed in the exposed side and the remaining zone was at ambient temperature (10–20°C). It is also necessary to underline that the larva which were alive before the treatment (Fig. 5A) died after having reached a temperature of the order of 50°C (Fig. 5B,C).

Conclusions

Microwave heating applications are increasingly proposed in environmental engineering (Jones *et al.*, 2002). A number of areas are assessed, including contaminated soil remediation, waste processing, mineral processing and activated carbon regeneration. Benefits such as reduced energy consumption, process time savings, increased process yield and environmental compatibility are exploited and in this context microwaves could be a potential candidate for limiting the spread of *R. ferrugineus.* In this preliminary study microwaves are tested to evaluate the potential technique which could treat infested palms directly in the field and the results are very promising for controlling red palm weevil. The irradiation of a palm with a 2.45 GHz magnetron produces a rapid increase of the temperature in the tissues, that can reach up to 70°C in some regions just 2 min after the radiation being switched on; longer treatment increases the heated area, but the high temperature spots do not seem to increase excessively. Probably, this behaviour is due to the fact that the heating of wood produces the concurrent evaporation of part of the water contained in its tissues. These results show that microwave heating could induce a thermal increase capable of killing the insects without damaging the palm tissues. In spite of the high temperatures reached in some 'spot zones', palm functions seem not to be damaged; only the external layer treated is slightly dehydrated due to the microwave irradiation (Griffo *et al.*, 2009). At present Campania Region is supporting a project in order to evaluate the long-term effects of the microwave exposures on treated palms.

In the last few years an important effort has been made in the search of an efficient, cheap and eco-compatible method for fighting the *R. ferrugineus* but, to date, a solution has not yet been found. The current emergency can be dealt with only by combining different techniques that need to be chosen and applied according to each particular case. The technique the authors propose in this work should be considered as a valid additional tool for the fight against the curculionid.

Considering that the external symptoms of the infestation are rarely visible at the very early stages, when smaller numbers of larvae are present, and become obvious only in the advanced stage of infestation, the method, based on thermal effect of microwave treatment, can be used as a curative treatment of infected palm trees. It can also be used as a preventive treatment in the production of healthy plants for planting, as well as for disposal of unrecoverable palm trees that are in advanced stage of infestation. In this latter case the infested tissues can be overheated with microwaves so they are totally disinfested and can be safely disposed of as standard green waste. This method could be an alternative disposal to that currently used in many countries such as grinding or burning the plants in an incinerator (EU, 2010).

Acknowledgements

The authors would like to thank Regione Campania, CERICT and MIPAAF (DIPROPALM) for supporting their research. The authors would also like to express their gratitude in memory of Adriano Isotton, whose enthusiasm was an inspiration for their work.

Traitement par microondes pour la lutte phytosanitaire: le cas de *Rhynchophorus ferrugineus* dans *Phoenix canariensis*

Rhynchophorus ferrugineus (Charançon rouge du palmier) est actuellement une des menaces principales pour les palmiers de la zone méditerranéenne. Aucune technique isolée développée jusqu'à présent ne semble pourvoir éradiquer complètement ce ravageur. Cet article présente le traitement par microonde comme un outil efficace pour aider au contrôle de cet insecte; l'effet des radiations des microondes sur les tissus des palmiers et sur le charançon est décrit et discuté. Le principal avantage des microondes est leur éco-compatibilité, et les résultats préliminaires de leur application à la lutte contre *Rhynchophorus ferrugineus* sont très prometteurs.

Микроволновые обработки для борьбы с вредными организмами: случай *Rhynchophorus ferrugineus* в пальмах *Phoenix canariensis*

Rhynchophorus ferrugineus (красный пальмовый долгоносик) в настоящее время представляет собой одну из наибольших угроз для пальмовых деревьев в районе Средиземного моря. Создается впечатление, что ни одна из методик, разработанных до настоящего времени, не в состоянии обеспечить полное уничтожение этого вредителя. В данной работе представлена микроволновая обработка, рассматриваемая в качестве эффективного инструмента, помогающего бороться с этим вредителем; описывается и обсуждается воздействие микроволнового облучения на пальмовые ткани и долгоносиков *Curculionidae*. Главное преимущество микроволновых установок заключается в их экологической совместимости, а предварительные результаты их применения в борьбе с *Rhynchophorus ferrugineus* были весьма многообещающими.

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