



**SOIL BIODESINFECTATION AND PARTICIPATIVE RESEARCH. MEDITERRANEAN
HORTICULTURAL CROPS**

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"Die Notwendigkeit zu entscheiden reicht weiter als die Möglichkeit zu erkennen"

(The need to decide surpasses the possibilities of knowledge)

I. Kant in Barres et al. (2008)

"In a moment of decision the best thing you can do is the right thing.

The worst thing you can do is nothing"

Theodore Roosevelt (1903)

Summary

The complexities involved in the use of organic matter for the management of soilborne pathogens through soil biodesinfection highlights the need for participatory research, where scientific "know-how" is combined with the experience of technicians and producers. Horticultural crops in the Mediterranean region are analysed, highlighting that production techniques should be optimized to efficiently manage fungi and phytoparasitic nematodes. The root-knot nematodes *Meloidogyne arenaria*, *M. incognita* and *M. javanica* are localized in thermophilic areas and in greenhouses. Other quarantine organisms are *M. chitwoodi* and *M. fallax* from cooler climates, which should be studied to avoid their propagation in the Mediterranean region. The problems associated with cyst nematodes of the genera *Globodera* and *Heterodera* are also highlighted. However, these organisms have a restricted host range and can be easily controlled through the use of crop rotations, as well as nematodes of bulbs, aerial parts and seed of the genera *Ditylenchus*, *Aphelenchoides* and *Anguina*. The virus vectors, the endoparasites and a large number of ectoparasitic species, without forgetting the mycophagous nematodes of mushroom, are also analyzed. Soil biodesinfection is described not only from a plant health perspective but also from that of its role in soil improvement, although these techniques should be combined with other production methods to increase self-regulation capacity and to design suppressive production systems. The factors which have allowed the maintenance of crop viability in traditional systems are discussed, given that they could be fundamental in the management of some horticultural crop systems using agroecological criteria.

Keywords: agroecology, biodesinfection, crop rotation, fungi, nematodes, organic matter.

Introduction

In Agricultural Science, research and technology transfer for the design of crop management systems should, particularly in the agronomic field, should be based on Participatory Research (PR) models which set out to combine scientific knowledge and the experience of farmers and live stockbreeders. The knowledge acquired by the farmer is vital because, together with the agricultural entrepreneur, he can respond to citizens' demands for quality agricultural products at a fair price. This, in its turn, leads to the promotion of knowledge for the activation of the economy.

Agricultural systems are so complex that they require the involvement, not only of scientists and technical experts, but also of farmers and live stockbreeders, as well as their agricultural organizations, through the creation of PR programmes based on agroecological criteria. At the same time, social, economic and commercial aspects have to be taken into account. PR programmes began with Rhoades & Booth (1982), taking as their reference point Rogers' innovation dissemination theory (1962). The methodology was recently analyzed in the case of potatoes by Ortiz *et al.* (2008), who also bear in mind the ideas suggested by Engel (1997) and Biggs & Matsuert (2004), in their attempt to identify the chief components of the system and their function, as well as to establish the interactions existing between them. It should be remembered, however, that the participation of the farmers themselves, along with their families and the agricultural communities, in the programme is essential. Local governments also have a role to play too. The PR method enables farmers to acquire new knowledge and techniques and, above all, to strengthen their organization at local level. There is, however, a striking lack of information and PR experts. There is also a lack of coordination amongst the institutions that take part and the relations existing between them are far from stable. This is largely the result of the usually precarious job situations of the people responsible for PR projects, who should – in

addition – be devoted full time to PR so that farmers and their organizations can make more effective use of the available resources, avoiding duplication of effort and undesirable competition between one organization and another.

The first step to be taken in a PR programme is the demystification of “science, which, like culture, should be the heritage of all citizens”. This means that scientists should work harder on how they transmit their knowledge, using terminology that technicians, farmers and live stockbreeders can understand. This will in its turn generate knowledge that can be used to develop easily applicable working methods. These methods should at the same time help to improve the economic situation of farmers through the development of viable knowledge transfer systems which include nutritional, social and environmental quality, aspects that people are concerned about.

In this paper the problem caused by plant parasitic nematodes in horticultural crops have been chosen as the model for study since, from the point of view of crop health, they are one of the chief causes of yield loss due to the presence of gall-forming nematodes *Meloidogyne arenaria*, *M. hapla* and *M. incognita* in Europe and America and *M. javanica* in Africa and Asia. Two highly pathogenic nematodes have been recently described within the *Meloidogyne* genus, *M. chiwoodi* and *M. fallax*, which cause very serious problems in northern Europe, where they are considered as quarantine nematodes. Their introduction in Spain should be averted at all costs (Bello *et al.* 2008), as should the cyst-forming nematodes *Globodera pallida* and *G. rostochiensis* (Greco *et al.* 1982, López-Cepero 1992, García Álvarez *et al.* 2005).

Among the endoparasitic nematodes the genus *Pratylenchus* is highlighted for the problems it causes in horticultural crops, fruit and banana trees. They are polyphagous migratory species and produce necrosis in the roots, and, in the case of *P. penetrans*, may be associated with fungi such as *Rhizoctonia solani*, *Verticillium* and *Fusarium* in horticultural crops. One should also emphasize the destructive role played by the virus-transmitting polyphagous migratory ectoparasitic nematodes, especially in the case of potatoes. *Paratrichodorus pachydermus* and *Trichodorus primitivus* have been found in potato crops in Europe, *P. minor* in North America and *P. minor* and *P. Teres* (López-Pérez *et al.* 2001) in the Canary Islands. Finally, various species of the genera *Helicotylenchus*, *Hemicycliophora*, *Nacobbus*, *Paratylenchus*, *Rotylenchulus*, *Rotylenchus*, *Tylechorhynchus* and *Xiphinema* have been found but they are not considered to present serious problems (Bello *et al.* 2008b).

It should not be forgotten that other saprophagous nematodes of the genera *Rhabditidis* and *Diplogaster* exist in the soil which can act as secondary pathogenic organisms in tuber crops already affected by other pathogens. And there are species of the genus *Ditylenchus* which, though not parasites, can favour the diffusion of bacteria and pathogenic fungi. It is also important to remember that when it comes to managing soil-borne organisms including nematodes one has to bear in mind that the *Rhabditidis* species decompose organic matter, the self-regulating mechanisms of the predatory Mononchidae and Discolaimidae species and the bioindicative value of the impact of cultivation practices of species of the Dorylaimidae order. Finally, one should not forget either the entomopathogenic nematodes and their contribution to the regulation of problems caused by pests (Kaya & Gaugler 1993).

One of the main problems with nematodes in horticultural crops is virulence selection when resistant varieties are used for the management of gall-forming nematodes including *Meloidogyne incognita* in peppers and tomatoes (Robertson *et al.* 2006, Torres *et al.* 2007)

and the management of cyst-forming of the genus *Globodera*, introduced principally through seed potatoes (Bello *et al.* 2008b).

Crop rotation is a solution for specific nematodes of the *Globodera* genus (*G. pallida* and *G. rostochiensis*) but it is not effective against polyphagous species. The possible introduction of *M. chitwoodi* and *M. fallax* could cause problems because they are polyphagous and affect vegetable species in natural areas. In the use of substrates, it should be pointed out that those of natural origin including the "enarenados" (sandy soils) of Almería, the "jables" and "sorribas" of the Canary Islands have been the chief explanation for the continued cultivation of vegetables in Almería and the Canary Islands but, owing to a lack of sanitary control of the seeds and especially to the prevailing monoculture, these substrates have been affected by the introduction of pathogenic organisms (López-Gálvez & Naredo 1996, Bello 2008b).

Soil solarization is ineffective in the control of nematodes. It has been shown that as soil temperature increases large-scale migration of nematodes takes place (Table 3) and in the case of "jables" isothermic substrates where there is no in-depth temperature increase. Farmers have for this reason developed the alternative known as "minado", which is a possible solution to the problem when combined with other cultivation techniques. "minado" is based on the combination of solarization with biofumigation (biosolarization), which serves to biodisinfest soils and, at the same time, reduce the amount of organic matter and its negative impact on tuberization (Katan & de Vay 1991, Bello *et al.* 2008b).

Borruey Aznar & Mula Acosta (2008) applied biodisinfection techniques in the management of common scabies of the potato, caused by the bacterium *Streptomyces scabies*, in the municipality of Bello (Teruel), using 41 t · ha of fresh sheep manure, saturation irrigation and covering the soil with 400 galga- thick plastic, placed on 19th July and removed on 21st October 2005, three months after its application. Once the soil had been disinfected, the cv "Agrida" potatoes were sown in 5 litre pots. Temperature was maintained at 22°C and they were harvested 140 days after planting. With regard to the incidence of scabies in the treated plots, the tubers were healthy (index 0) or had no more than 5% of their surface affected (index 1) while in the control plots tubers appeared with indices of 2 and 3 – that is to say 10 and 25% respectively of scabies –affected surface. The first effect of biodisinfection was observed at the moment of withdrawing the plastic since, while the control plots were full of vegetation, the covered area was practically clean. Production increased by 36% in the biodisinfected plots (45,379 kg ha as opposed to 33,476 kg ha), while the incidence of scabies was 36.3% in the biodisinfected soil as against 76.2% in control soil. Bearing in mind that the tubers with index 1 of scabies are marketable, scabies impact is thus reduced to 5.8% in treated soil as opposed to 33.5% in control soil. As a result, 93.2% of production in biodisinfected soil was marketable (42,573 kg ha), as against 57.7% of production in control soil (19,316 kg ha). This circumstance would permit the reduction to one year of traditional rotation with grain crops, as opposed to periods of three to seven years of grain monoculture. Finally, the use of green manures is proposed for future studies.

On account of the seriousness of the problems caused by nematodes in vegetable crops and especially their great complexity, we have chosen as the watchword for our study I. Kant's statement in Barres *et al.* (2007): "The need to decide exceeds the possibilities of knowledge", trying to for this purpose design a PR project that combines scientific knowledge with that of technical experts and farmers. We take as our points of reference the studies of Díaz Jiménez (2007) and Torres *et al.* (2007). We have also enjoyed the cooperation of farmers José Ballester (El Perelló, Valencia), Miguel S. Benítez-Gil (northern Tenerife) and Juan Ramón Delgado Martín (southern Tenerife), who belong to families several generations of vegetable farmers.

Materials and methods

We begin with an examination of the paper by Bello *et al.* (2008b,c) with a view to establishing clearly the phytonematological problems posed by horticultural crops. Among the non-chemical management alternatives, we have analyzed those proposed by the Methyl Bromide Technical Options Committee (MBTOC 2007), which forms part of the Montreal Protocol for the search of alternatives to methyl bromide (MB), a soil fumigant whose emissions destroy the stratospheric ozone layer (Tables 1 & 2). Of special interest are sanitary measures in the use of seeds and vegetable material, irrigation water and pathogen-free machinery, but especially those designed to regulate the introduction of pathogenic organisms. Also to be included here are the biodisinfection of the soil through the management of organic material, substrates of natural origin such as the "enarenados" (sandy soils) of Almeria, the "jables" and "sorribas" systems in the Canary Islands, the use of resistant varieties, the rotation of crops, catch crops and above all, traditional practices which give rise to suppressive production systems where pest and disease problems are infrequent (Rodríguez-Kábana & Canullo 1992, Urbano & Moro 1992).

The biodisinfection of soils has been studied for the management of both nematodes and soil fungi, following the methodology described in Bello *et al.* (2003, 2008a). Examples were chosen which, based on the application of solid and liquid biodisinfectants, proved effective in the control of fungi and the pathogenic nematodes of vegetables (Bello 1998, Kirkegaard & Sarwar 1998, Blok *et al.* 2000, Bello *et al.* 2003, 2008a-d, Lazarovits *et al.* 2005).

In order to be able to design rotation systems for the management of gall-forming nematodes of the genus *Meloidogyne* and to improve the efficacy of biodisinfection treatment, we started by characterizing the biotypes, using the methods developed by Robertson *et al.* (2006) and Torres *et al.* (2007), taking as our term of reference the Hartman and Sasser's race characterization scheme (1985). One should also draw attention to recent studies on the application of plant improvement, through the selection of resistant varieties for the control of *Phytophthora infestans*, late potato blight, *Rhizoctonia*, *Synchytrium endobioticum*, *Nacobus aberrans*, *Globodera pallida*, *G. rostochiensis*, *Erwinia carotovora*, *Ralstonia solanacearum* and virus (Hernández *et al.* 2008).

Once the necessary information had been gathered from the experts and researchers, the field teams were asked which of the selected alternatives were viable, following the method established by Díaz-Jiménez (2007) and Torres *et al.* (2007), using an open and flexible questionnaire in the north and south of the island of Tenerife on different crops, at different times and with different cultivation systems. They were asked about possible interactions among the pest and disease problems found, as also the economic feasibility of the alternatives and the possibility of applying a management model based on agroecological criteria in each area.

Results

We then proceeded to analyze the phytonematological problems involved according to their order of magnitude, indicating the results of applying different soil biodisinfection techniques, and finally establishing their viability in the existing production systems with the help of farmers from Almería, the island of Tenerife and Valencia.

The gall-forming *Meloidogyne* stand out among the phytoparasitic nematodes, represented fundamentally by *M. arenaria*, *M. incognita* and *M. javanica*, which are found in thermophilic areas or greenhouse crops. In Spain, however, *M. hapla* predominates in temperate areas. *Meloidogyne* cause highly characteristic root nodules and, in terms of management, it should be remembered that they are polyphagous, which means that crop rotation is not always the answer. The production of deformities in the roots and tubers makes the presence of these nematodes extremely easy to detect. *M. fallax* has been found in northern Tenerife. This and *M. chitwoodi* are species normally found in colder climates of northern Europe and, if they come to flourish in horticultural crops, can constitute a serious problem.

Cyst-forming nematodes of the genus *Globdera* are represented by *G. pallida* and *G. rostochiensis*, which are parasites specific to potato, although *G. pallida* can also parasitize tomatoes. Potato cvs "Cara" and "Red Cara" present resistance to *G. rostochiensis* but not to *G. pallida*. Since they are specific nematodes, the most effective way of managing them is crop rotation, although it should be pointed out that these nematodes have been introduced by tubers used as seeds. One has to remember that potato growing has been maintained over the last few years in the Canary Islands thanks, essentially, to the design of substrates of natural origin: the "jables" of southern Tenerife and the "sorribas" to be found in all the islands. Among the "jables", the management technique known as "el minado" is of special importance. This consists in applying to the "jable" substrates intensive irrigation, to the point of saturation, although this is only effective when organic matter, which acts as a biodisinfecant, is applied at the same time (Bello *et al.* 2008b).

Other species of nematodes are represented by pathogens of aerial parts and the bulbs of the genus *Ditylenchus*. We have already spoken of *D. dipsaci*, but one should not forget the genera *Aphlenchoides* and *Anguina*. Also deserving of a mention are the endoparasitic species of the genus *Pratylenchus* which cause necrosis in the roots. Various species and genera of ectoparasitic nematodes including *Criconemoides mutabile*, *Helicotylenchus digonicus*, *H. dihystra*, *H. erythrinae*, *Quinisulcius acti* and *Rotylenchus hopper* have also been found but, in spite of being polyphagous, they are not known to have caused problems in horticultural crops. It is worth stressing that, among the nematodes associated with horticultural crops, there has been no evidence in Spain of *Ditylenchos destructor* or *M. chitwoodi*, while *M. fallax* is found only in the northern region of the island of Tenerife. Virus-transmitting species of the genera *Paratrichodorus* and *Trichodorus* are present. *Paratrichodorus minor* and *P. teres* (López-Pérez *et al.* 2001), along with tropical species including *Nacobus*, *Radopholus* and *Rotylenchulus*, have not been observed in the Canary Islands.

The biodisinfection of soils is carried out using gases from the decomposition of organic matter to regulate the presence in the soil of populations of pathogenic organisms in plants without this having any negative impact on edaphic diversity. Biodisinfection is based on the biological activity of the soil, which is why it tends to be ineffective when applied for the first time to conventional farming soils where chemical fumigants have been used continuously, although its efficacy improves with time and repeated application. One has to bear in mind that there are different kinds of biodisinfecants, solid and liquid, but their decomposition gives rise to gases, principally ammonium, which all act as soil biodisinfecants. The biodisinfection of soils should form part of a production management system (PMS), being combined with other alternatives, in particular crop rotation or resistant varieties to ensure its effectiveness in time. We pass on now to some of the results obtained from our working group's research.

The first assays in biodisinfection began in El Perelló (Valencia) using organic matter, mainly animal manure, in vegetable crops (Table 3). Efficacy was similar to that of the

chemical soil fumigant MB, especially in the management of gall-forming nematodes of the genus *Meloidogyne*. The results figure in the work of Bello *et al.* (2003). Several farmers have adopted soil biodisinfection using sheep manure at doses of from 4 to 5 kg/m² as an alternative to traditional fumigants. A recent PR project involved using 2 l/m² doses of industrial citrus and covered sugar beet vinasses waste without plastic covering. The first-phase application of vinasses injected into the soil, with abundant localized irrigation, proved highly efficient in the management of nematodes, with an average galling index of 2.2, while citrus and control wastes produced indices of 5.1 and 5.3 respectively. The production figure was 4.5 kg/m² in the case of treatment with injected beet vinasses and production was similar with citrus waste without plastic. Although the Cooperative's average stands at 7 kg/m² (Table 4), one has to bear in mind that the experiment corresponds to a late – and shorter - cultivation cycle in which production is usually much lower.

In view of the need to eliminate MB, a powerful destroyer of the ozone layer, the former Ministry of the Environment launched an alternatives project, selecting biodisinfection principally for sweet pepper crops in El Campo de Cartagena (Murcia), since this was one of the areas where the critical use of MB was permitted. Work has been going forward in this area for over seven years in the use of mixes of fresh sheep and chicken manure, combined with resistant plants for the control of *Meloidogyne incognita*, and this has proved effective not only against nematodes but also against fungi and weeds (Tables 5 & 6). One can progressively reduce the doses of organic matter, after repeated use during the summer, reaching a dose of 2 kg/m² of fresh sheep manure combined with 0.5 kg/m² of chicken manure as from the fifth year.

Research has been on going at the Centro Agrario of Marchamalo (Guadalajara) - under the auspices of the regional government of Castilla-La Mancha – into the effect of beet vinasses at a dose of 1.5 kg./m², using plastic covering. This has been effective in the control of *M. incognita* (Table 7) in a soil based on river-borne silt and clay sediments, the result of the materials through which the Henares river passes. Most of these are relatively unconsolidated – that is to say, made up of loam, clay and sand (Jimeno *et al.* 1987). It is, as a result, a deep soil, of medium to strong texture (open sand) with a high water-retention capacity and without surface stones, with a moderately basic pH, abundant organic material and nutrients. Its capacity for change is good and saturation of the colloidal complex is very high. Calcium carbonate is present from a depth of 45 cm.

In view of the importance of nematodes in agriculture, one should emphasize the presence in the experiment of, in addition to phytoparasites, the groups of saprophytes and predators which have a considerable impact on the decomposition of organic matter and increased soil self-regulation capacity, as well as other groups of free living nematodes, especially the Dorylaimids, which are valuable bioindicators. Among the Mononchid predators the predominant species is *Mylonchulus sigmaturus* and among the Dorylaimids *Aporcelaimellus obtudicaudatus* and *Ecumenicus monohystera*, while *Mesodorylaimus litoralis* and *Nygolaimus* sp. also appear in the control plots. These nematodes are not generally abundant and may belong to the remains of nematofauna associated with the original vegetation of the zone. Being highly sensitive to cultivation techniques, they could be extremely useful as *bioindicators*. The saprophytic nematodes belong to the order Rhabditid and the Mesorhabditidae family, are responsible for the decomposition of organic matter. The number of individuals is low, which suggests that higher levels of organic matter should be incorporated into the soil to increase the efficacy of biodisinfection. *Cruzinema tripartitum* was also found as a predominant species among the Mesorhabditidae in previous studies. These results for free living nematodes and saprophytic nematodes show the importance of organic farming methods from the point of view of soil

functionality, since these species are not abundant or fail to appear in soils where chemical fumigants are applied (Bello *et al.* 2008b).

The University of Burgos is carrying out experiments in the management at laboratory level of populations of the potato cyst-forming nematode *Globodera rostochiensis*, using pig slurry and *Raphanus sativus* green manures. One part of an experiment was carried out without cover and the other with plastic to retain the gases. The results are extremely promising, especially when plastic covering is used (Table 8).

González-López (2007) has studied the effect of different concentrations of vinasses from byproducts of the wine industry used in obtaining grape remains in cultivated soils, in the search for greater agronomic viability and improved application techniques. The following goals were set: 1, to study their impact on nematode populations in the soil, in particular the control of *Xiphinema index*, the nematode vector of "grape fanleaf virus" (GFLV), one of the main reasons why the disinfection of soils for the replanting of vineyards is obligatory in EU countries, and also examine their effect on gall-forming nematodes of the genus *Meloidogyne*, principally *M. arenaria* and *M. incognita* species, which, apart from being parasites of grapevine, are also among the main causes of horticultural crop losses; 2, to establish their impact on soil fertility both in vineyards and horticultural crops and the possible phytotoxic and environmental effects of the selected concentrations; and 3, to determine the optimal doses, along with the season, methods and machinery suitable for their application, bearing in mind the scientific and phytotechnical ground rules for the use of agroindustrial byproducts. It was found that *in vitro* control of the *X. index* nematode is effective with wine industry vinasses at a concentration of 5% and that this is also effective in the control of *Meloidogyne* populations. It has been found that the saprophagous nematodes of the rhabditids and enquitreids of the oligochetes group, which are fundamental in the dynamics of organic matter, are increased when biodisinfection is carried out with vinasses from the wine industry, especially when solid byproducts are applied. The conclusion is that biodisinfection should be applied to the whole plot to avert the appearance of areas infected with populations of phytoparasitic nematodes. The use of biodisinfection could reduce the cost of soil disinfection in the replanting of vineyards and improve soil characteristics by increasing fertility and biodiversity, while also improving its physical properties. It at the same contributes to the search for solutions to the large quantities of waste generated by the wine industry.

Santos *et al.* (2008) have studied the biocidal effect of three agroindustrial byproducts from the obtaining of alcohol: sugar cane, beet and wine vinasses. Their behaviour *in vitro* was studied at different dilutions - 1,3,5,7, 10 and 15% - in combatting six phytopathogenic fungi: *Fusarium oxysporum* f.sp. *melonis* strains 0 and 1, *F. oxysporum* f.sp. *radiciscucumerinum*, *Phytophthora parasitica*, *Pythium_aphanidermatum* and *Sclerotinia sclerotiorum*. The next step was the study of the antagonistic capacity of the dilutions tested *in vitro* on bean-growing soil in laboratory conditions to analyze the behaviour of the byproducts with fusarium populations. *In vitro* assays showed that wine vinasses presented a 100% fungic growth suppression capacity at dilutions of between 5 and 7% for *F. oxysporum* f.sp. *melonis* strains 0 and 1, *P. aphanidermatum*, *P. parasitica* and *S. sclerotiorum* and of between 10 and 15% for *F.oxysporum* f.sp. *radicis- cucumerinum*. Beet vinasses also have an approximately 100% suppressive impact on fungic growth in the case of some of the phytopathogenic fungi assayed: *P.aphanydermatum* at a 7% dilution and *F. oxysporum* f.sp.*radicis-cucumerinum* when exposed to concentrations of 15%, while no 100% inhibition was observed in the control of *F.oxysporum* f.sp. *melonis* strains 0 and 1 at any dilution. Although maximum inhibition occurs at high dilutions, there are statistically significant differences from controls at lower concentrations, as is the case of the 1% for *F. oxysporum* f.sp. *radicis-cucumerinum*, the 3% registered for *P. aphanidermatum* and *P. parasitica* and the 7% found for *S. sclerotiorum*. With sugar cane

vinasses, on the other hand, no inhibition of mycelial development was achieved in any of the fungi assayed and they had no effect on *P. aphanidermatum*, which showed maximum growth at all the dilutions assayed. This study demonstrated that, without a doubt, wine vinasses give the best results, with inhibition close to 100% in all the fungi assayed. On the other hand, when the three vinasses extracts are applied to samples of bean-growing soil with no pathogenesis present, there is an increase in the fusarium populations. It is thought that they may act as soil improvers by favouring biodiversity. Diáñez *et al.* (2007) obtained results similar to those registered with wine vinasses when they used grape remains compost, observing its suppressive impact on nine phytopathogenic fungi using the aereated "extracts" of this compost in *in vitro* studies. A high level of mycelial growth inhibition was obtained, in all cases above 80% and at all the concentration used.

The town council of Tenerife is working on the biodisinfection of soils for the cultivation of potatoes in northern Tenerife. It has obtained effective results with cow and green manures in the control of *Rhizoctonia solani* and these are evident in crop yields (Tascón *et al.* 2007). Biodisinfection with manure is the only treatment that reaches an hour-long temperature of 47° C, this being the critical temperature and the exposition time required to kill 90% of propagated *R. solani*. There are seen to be significant differences between the production of control crops and those treated with solarization, biodisinfection with cauliflower and with manure. In the case of *R. solani* sclerotinia in tubers, the best results are obtained with cauliflower biodisinfection (63.7%), followed by solarization (19.6%) and manure biodisinfection (9.6%). One can thus conclude that the effect is due, not only to the temperature needed to kill 90% of the propagated *R. solani*, but also to other factors. The largest difference between income and expenditure, bearing in mind disinfection only, is obtained with solarization (2,188 Euros/ 1,000 m²), followed by biodisinfection with cauliflower (2,020 Euros/ 1,000m²), with manure (1,834 Euros/ 1,000 m²) and finally the controls (1.430 Euros/ 1,000 m²).

The biodisinfectant effect of the application of organic matter combined with solarization in representative sandy soils has been studied in Almería, where chemical soil disinfection methods have been the norm (Torres *et al.* 2007). The general aim of the study was the management of *Meloidogyne* gall-forming nematodes with non-chemical alternatives. The specific goals were to recover and improve traditional techniques for the application of organic matter in the sandy soils of Almería based on scientific principles regulating the processes of soil biodisinfection. These systems are also combined with solarization (biodisinfection). For this purpose, research has centred on evaluation of the biodisinfectant effect of organic matter as traditionally applied in the management of edaphic pathogenic organisms in vegetables. Also examined was the effect of using tomato crop remains, contributing in this way to closing the nutrient cycles in the cultivation system. Biodisinfection combined with other management alternatives was proposed. These alternatives included the use of resistant varieties, after studying their behavior with different nematode biotypes of *Meloidogyne*, to increase their efficacy, and other traditional practices.

The efficacy of biodisinfection depends fundamentally on the method applied, not on isolated factors such as dose, composition or organic matter distribution in the soil. Costs depend on the method chosen, which should set out to reduce dose and transport, using local byproducts. The traditional use of sandy soils is recommended in the control of nematodes, and it is to be pointed out that this also modifies the distribution in depth of the crops' root systems, a limiting factor since it requires the establishment of an agronomic management system adapted to each individual circumstance. The presence of complexes of species or biotypes of *Meloidogyne* in the sandy soils of Almería frequently means that their control requires the implementation of a management system based on

agroecological criteria that takes into account the experience of farmers and traditional cultivation techniques.

Discussion

The management of agricultural systems based on organic farming criteria requires the consideration of ecological questions that will enable us to understand how the systems work, taking the concepts of structure and function as points of departure, not with a view to describing all the elements and processes that make up the system, but to defining those key elements and processes that govern its working. To start with, one can define the aerial subsystem, a simple and directly observable phenosystem which is therefore easy to manage, the main difficulty lying in the fact that, being open, it depends on the activities of neighbouring farmers. This complicates management, especially in the case of pests and diseases, and this is the chief limiting factor. The second element to be considered is the edaphic subsystem, which, unlike the aerial one, is a cryptosystem by no means easy to observe and characterized by its diversity. It is therefore viewed as a complex and closed system the management of which depends on the management ability of each individual farmer. These agroecological criteria served as the basis for the establishment of traditional systems that have maintained their feasibility, in spite of the many limiting factors. Standing out among these traditional systems are the "jables" of the district of Chasna (southern Tenerife) which, if proper sanitary regulations had been in place, should not have had problems with soil diseases and pests, since we are talking about substrates of natural origin. The same is true of the "sorribas", where the basalt walls and sunrise orientation subject the borders to solarization, permitting the cultivation of vegetables highly sensitive to soil diseases such as sweet potato, cucurbit, cabbage and pepper.

One's attention is struck by the appearance of problems caused by the gall-forming nematodes *Meloidogyne arenaria*, *M. incognita* and *M. javanica* especially in horticultural crops. The optimization of soil biodisinfection techniques is proposed as a management measure, not only for pathogens, but also to increase the chemical, physical and biological fertility of the soil, using animal manure, green manures, especially brassicas, agroindustrial and livestock waste, thus closing, in addition, matter cycles in order to increase the self-regulation capacity of the crop.

More attention should be paid to learning about the processes regulating soil biodisinfection, using local resources as biodisinfectants, which can be of various kinds, both solid and liquid. Through a process of decomposition, these give rise to gases with high ammonia content which act as biodisinfectants. Biodisinfection techniques should form part of a harmonious crop-management programme to ensure the crop a self-regulation capacity and give rise to suppressive systems, making pests and diseases less frequent.

Crop varieties should be selected not only for their nutritive quality but also their resistance to, or tolerance of, pathogenic and parasitic organisms, their efficacy as catch crops, antagonists or vegetable cover to regulate soil temperature, which affects the duration of the pathogens' cycle, and their contribution to soil protection against erosion phenomena. Also, by means of mixed farming and associated crops, complemented with hedges to create barriers, where fruit trees can play an important role, one is contributing to the diversification of the agrarian landscape and preventing, in particular, the propagation of pests and diseases.

The use of criteria based on functional biodiversity allows us to maintain the agrosystem's self-regulatory capacity by promoting the increase of micorrizas, rizobacteria and endophytic micro-organisms, which play an important role in the enhancement of soil fertility, which in turn increases plant nutrition. At the same time, they can act as naturally induced agents of biological control. With this agroecological focus, one should also analyze the functional aspects of traditional practices, planting periods, farming techniques, organic amendments and compost additions, as well as water management. Specific local agrosystems have also an important place in agroecology and in this context the functional aspects of natural substrates are examined. These, in the case of the Canary Islands, take the shape of the "jables" of southern Tenerife, the "enarenados" (sandy soils) of Lanzarote and the "sorribas" to be found all over the islands, as well as the sandy soil systems of Almería, where every crop element has a noteworthy function in the system's sustainability. PR programmes are indispensable because they permit the combination of scientific knowledge with the experience of hands-on technicians, farmers and live stockbreeders. In this way, on the basis of agroecological criteria, one can design rotation systems appropriate to every circumstance which will bear in mind, not only the productive aspects, but also functional biodiversity criteria, without forgetting the complementary role of livestock breeding to achieve greater economic profitability from the productive system and the re-use of agricultural byproducts, thus closing the matter and energy cycles.

The difficulties observed in the implementation of PR programmes have been, in the first place, the organization and selection of scientific researchers since, in our country's present scientific situation, most of them are primarily concerned about the level of their publications on the international stage and are hard to convince of the potential value of their experiences in the task of changing the management model of agricultural systems at home, which in general have no apparent connection with their research work. The setting up of technical groups, on the other hand, has presented no serious difficulty because it means an opportunity to design new management methods and learn more about their working areas. The main problem in their case is that of combining the implementation of a project at experimental level with their technical work in the transfer of technology and, above all, with their administrative duties. For that reason, the Agricultural Advisory Service and the farmers' organizations should have at least one technical team doing experimental field work. Finally, farmers and stockbreeders are the key to a project's viability because, with the right working atmosphere, they provide information on agrosystem management, not only at district or regional level, but also as an on-going process in time. They are the depositaries of the knowledge of generations.

Conclusions

- For the management of agricultural systems with agroecological criteria we need an approach that allows us to understand the way they work, taking structure and function as our points of reference with a view to selecting the key elements and processes that govern the functioning of a given crop. These agroecological criteria served as the foundation for the design of traditional systems that are still viable in spite of the great number of limiting factors.
- Further studies should be made into the processes regulating soil biodisinfection, using local resources of various kinds, both solid and liquid, which through a dynamic decomposition process give rise to gases with a high ammonia content that act as biodisinfectants. Biodisinfection techniques should form part of a harmonious crop management programme which assures self-regulation and in turn suppressive systems where pests and diseases are infrequent.

- PR programmes are indispensable because they permit the combination of scientific knowledge with the experience of technical experts, farmers and stockbreeders, so that, with agroecological criteria, we can design rotation systems appropriate to every circumstance. These should bear in mind, not only the productive aspects, but also functional biodiversity, without forgetting the complementary role of livestock breeding, to achieve greater economic profitability from the productive system and the re-use of agricultural byproducts, closing matter and energy cycles and developing techniques for the management and application of organic matter.
- The difficulties observed in the implementation of a PR programme have been, in the first place, the selection and organization of a team of participating scientists since, in our country's present scientific situation, most of them are chiefly interested in the level of their publications in the international sphere and it is hard to convince them that their experience could help to change the management model in agricultural systems here. On the other hand, no serious difficulty has arisen with the setting up of a group of technical experts since this is an opportunity to plan new management methods, their main problem being combining work on the project with their administrative duties. The Agricultural Advisory Service and the Cooperatives should therefore assign at least one group of experts, free of office responsibilities, to field experiments. As to farmers and stockbreeders, it is clear that when the right working atmosphere is created there are no problems. It must be emphasized that they are the key to the project's viability, since they supply information on the management of agrosystems, not only at district or regional level, but as an on-going process in time, being depositaries of the accumulated knowledge of generations.

Acknowledgements

We wish to express our gratitude for the cooperation of Desiré Alfonso and Cristo Medina of the Centro de Conservación de la Biodiversidad Agrícola of the Cabildo Insular de Tenerife, and especially to the Canary Island farmers interviewed: Modesto Bernardo, Manuel Díaz, Gregorio Gil, Antonio González, Eusebio Hernández, Sandra Kober, Pedro Naranjo, Daniel Romero and Onésimo Santos. Our thanks go also to the staff of the Laboratorio de Canarias Explosivos, S.A., for their analyses; to the Cooperativa Unión Protectora of El Perelló (Valencia) and in particular to the farmer Concepción Puchares Miñana for her cooperation in organizing the experimental field work; to Dr Francisco Camacho of the Departamento de Producción Vegetal of the University of Almería; to the SAT "Costa de Nijar" ; to the farmers Jesús Ochoa of Santo Domingo de la Calzada, of the UAGR of La Rioja, and Jesús Sanchís de Liria (Valencia), of the ULLYR, for their cooperation. We would place special emphasis too on the contributions of Doctors Alfonso Navas of the Museo Nacional de Ciencias Naturales in Madrid, Rodrigo Rodríguez-Kábana of the University of Auburn, Alabama, and Pedro Urbano of the Departamento de Producción Vegetal, Fitotecnia, Universidad Politécnica de Madrid, who, with their ideas and moral support, have done so much to ensure that this study reached the light of day.

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Table 1.- Non-chemical alternatives to soil fumigants (MBTOC 2007)

1. Sanitary Measures: use of seeds, plants, irrigation and pathogen-free machinery.
2. Biodisinfection of soils: biofumigation, biosolarization.
3. Substrates, covering or mulching from local resources: enarenado (sandy soil), jables y sorribas.
4. Use of resistant varieties.
5. Crop rotation, catch crops, antagonistic plants.
6. Traditional practices:
 - 6.1. Planting period.
 - 6.2. Management of water and irrigation.
 - 6.3. Work.
 - 6.4. Organic amendments and compost.
7. Induced resistance:
 - 7.1. Endophytic microorganisms, micorrizas, rhizobacterias.
 - 7.2. Fertilization and nutrition of the plant.
8. Naturally induced biological control agents.

Design of production systems using agroecological criteria and local resources.

Table 2.- MB Consumption for critical use as a soil fumigant (MBTOC 2008)

Country / year	2009	2010
Australia		
- Strawberry greenhouses	29,790	29,790
Canada		
- Strawberry greenhouses	7,462	7,462
Israel		
1. Broomrape	125,000	-----
2. Greenhouse flowers and bulbs	85,431	-----
3. Wild flowers	34,698	-----
4. Greenhouse and wild melon	87,500	-----
5. Potato	75,500	-----
6. Greenhouse strawberry production	77,750	-----
7. Greenhouse strawberries	28,075	-----
8. Sweet potato	95,000	-----
Total	608,454	-----
Japan		
1. Cucumber	34,300	30,690
2. Wild ginger	63,056	53,400
3. Protected ginger	8,325	8,300
4. Melon	91,100	81,720
5. Pepper	81,149	72,990
6. Watermelon	21,650	14,500
Total	299,580	261,600
USA		
1. Cucurbits	407,091	302,974
2. Wild aubergine	48,691	32,820
3. Forestry	122,060	117,826
4. Fruit and flower greenhouses	25,326	17,363
5. Replanting of fruit trees and vineyards	292,756	215,800
6. Ornamentals	107,136	84,617
7. Wild pepper	548,984	463,282
8. Strawberry fields	1.269,321	1.007,477
9. Greenhouse strawberries	7,944	4,690
10. Sweet potato	18,144	14,515
11. Tomato field	1.003,876	737,584
Total	3.851,329	2.998,948

Table 3.- Biodisinfection experiment in "El Perelló" (Valencia), 5 kg m⁻² sheep manure (Indiv. 100 cc⁻¹ soil)

Depth	Sampling points (Indiv. 100 cc ⁻¹ soil)									Total	Average
	1	2	3	4	5	6	7	8(**)	9		
0-20 cm											
alive	0	0	0	0	0	2	0	2	0	2	0
death	28	14	12	6	220	8	12	8	6	306	38
index (*)	0	0	0	0	0	1	0	1	0	1	0.1
20-40 cm											
alive	0	0	0	0	0	4	0	28	0	4	1
death	290	118	6	24	210	0	48	92	16	712	89
index	4	2	1	0	0	1	3	1	1	12	1.5
> 40 cm											
alive	0	0	0	0	0	0	0	46	2	2	0
death	2	0	0	4	10	2	4	152	4	26	3
index	0	1	0	0	1	0	0	2	2	4	0.5

(*) Bridge & Page (1980) experimental index on tomato cv Marmande plant on 300 g of biofumigated soil after one month .

(**) Solarization.

Table 4.- Average nodulation rates by *Meloidogyne incognita* in cucumber cultivation, El Perelló (Valencia)*

	With plastic	Without plastic	Production kg m ⁻²
Control	-----	5,3 ± 1,8 a	3,7
Beet vinasses	3,5 ± 2,1 ab	5,1 ± 2,1 ab	3,2
Citric waste	5,1 ± 1,6 a	5,1 ± 2,7 ab	3,6
Injected vinasses	2,2 ± 1,9 b	4,5 ± 1,9 ab	4,5

* Non-parameter test of Kruskal-Wallis (significant difference = 0,004) Different letters indicate significant differences.

Table 5.- Effect of repeated biodisinfection (biosolarization, B+S) on production and other parameters for the cultivation of peppers (Campo de Cartagena, Murcia) compared to the use of MB. Biodisinfection lasted between 8 and 10 weeks with fresh sheep and chicken manure: 6 kg in the 3rd year and 2,5 kg m⁻² in the 5th, 6th and 7th (Table 6, Guerrero et al. 2008)

Treatment	Weed index	% infected plants <i>M. incognita</i>	Nodulation index	Height (cm)	Production (kg m ⁻²)
Control	1,58 b	83,33 d	3,97 d	158 b	6,34 b
B+S 3rd year	0,42 a	73,33 cd	2,33 c	185 a	8,70 a
B+S 5th year	0,67 a	13,33 ab	0,67 a	179 a	9,23 a
B+S 6th year	0,67 a	40,00 bc	1,67 b	188 a	8,40 a
B+S 7th year	0,17 a	6,67 a	0,13 a	188 a	8,49 a
BM 98:230 g m ⁻²	0,50 a	6,6 a	0,20 a	183 a	5,93 b

(*) Comparisons with ANOVA LSD (least significant difference). The same letter or letters in the same column do not indicate statistically significant differences (α= 0,05). The amounts show the average.

Table 6.- Amount of manure used in the repeated biodisinfection assay. Biodisinfection began between the 2nd and 3rd week of August and the experiment ended in the last week of October or the 1st of November. 0.05mm transparent polyethylene plastic was used. 98.2% methyl bromide was applied during the first 10 days of November, using 0.04mm VIF plastic (Guerrero *et al.* 2008)

Year of repeated biodisinfection	Fresh sheep manure (kg/m ²)	Chicken manure (kg/m ²)	Total (kg/m ²)
1st	7	3	10
2nd	5	2,5	7,5
3rd	4	2	6
4th	3	1,5	4,5
5th	2	0,5	2,5
6th	2	0,5	2,5
7th	2	0,5	2,5

Table 7.- Biodisinfection of soils with beet vinasses (1,5 kg m⁻²) 3^o year, Marchamalo (Guadalajara) Final experiment 07-08

Treatment	<i>Meloidogyne</i>	Dorilaimids	Rhabditids	Enquitreids	Mononquids
Control	218 ± 55,1 a	5 ± 4,2 a	73 ± 15,9 a	11 ± 3,5 a	0 ± 0 a
5% no plastic cover	143 ± 38,0 a	5 ± 2,5 a	84 ± 46,3 a	8 ± 1,9 a	0,5 ± 1 a
5% plastic cover	62 ± 39,6 b	3 ± 2,0 a	81 ± 88,8 a	8 ± 2,8 a	1 ± 2 a
A significance *	0,017	0,630	0,694	0,439	0,573

* Non-parameter trial by Kruskal-Wallis

Table 8.- Effect of the application of pig slurry (P), green manure *Raphanus sativus* (R) and control with water (A) on *Globodera rostochiensis* populations. The experiment was undertaken in open containers (a) and closed containers (c), following a period of 60 days of biodisinfection at 30 °C (viable and non-viable individuals / 100 cc)

		Treatments		Eggs & juveniles	
		1 ^o	2 ^o	Non-viable	Viabiles
P _a	Pig slurry (10 cc)	Open H ₂ O saturated		1653	125
P _c		Closed H ₂ O saturated		1715	16
A _a	H ₂ O (10 cc)	Open H ₂ O saturated		345	1263
A _c		Closed H ₂ O saturated		421	1317
R _a	<i>Raphanus sativus</i> (8 g)	Open H ₂ O saturated		895	643
R _c		Closed H ₂ O saturated		1689	12
T	Control		-----	18	1762



Figure 1.- Ecology of agrarian systems: structure and functions. Aerial and edafic subsystems characteristics

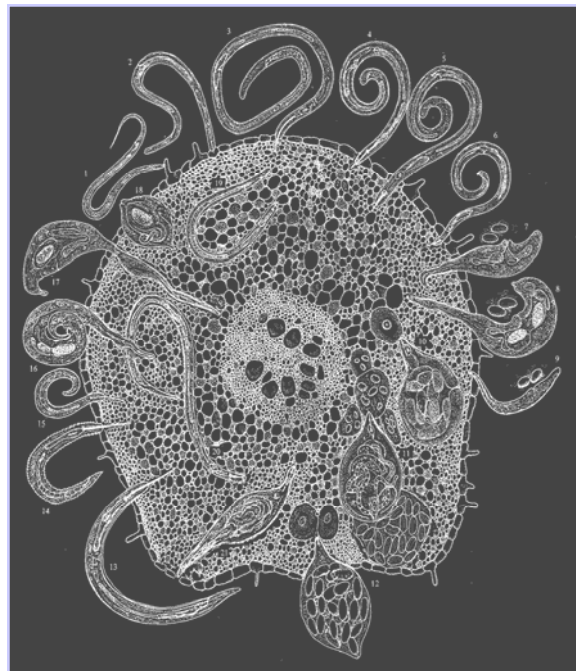


Figure 2.- Root transverse cut and characteristics of different nematodes groups

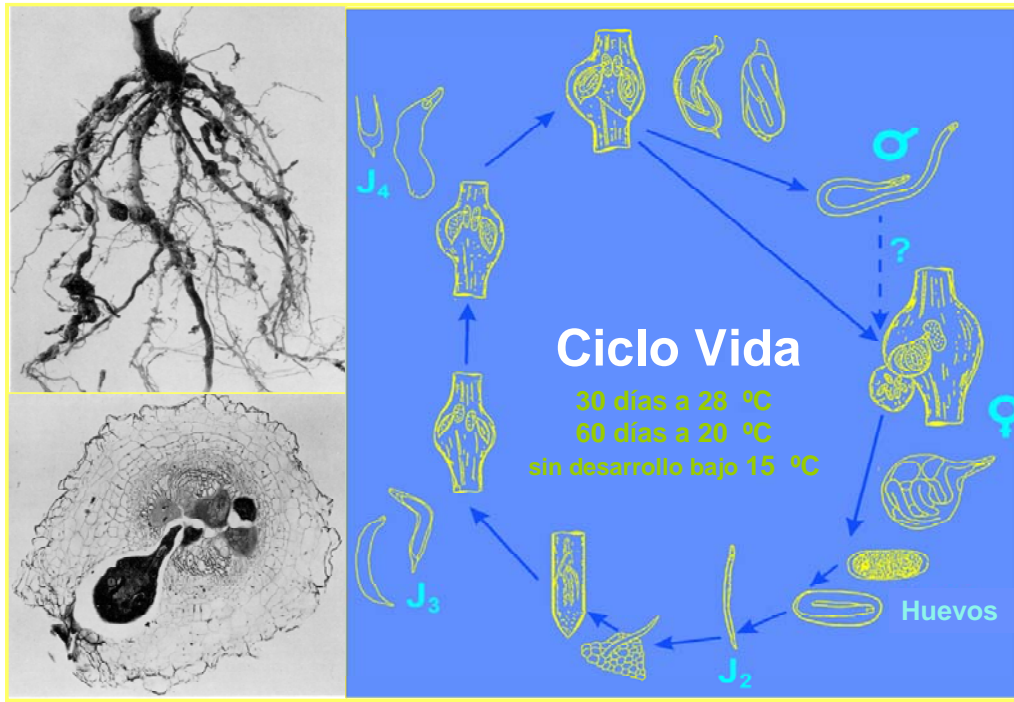


Figure 3.- Symptoms and life cycle of *M. incognita*. Temperature effect on life cycle duration

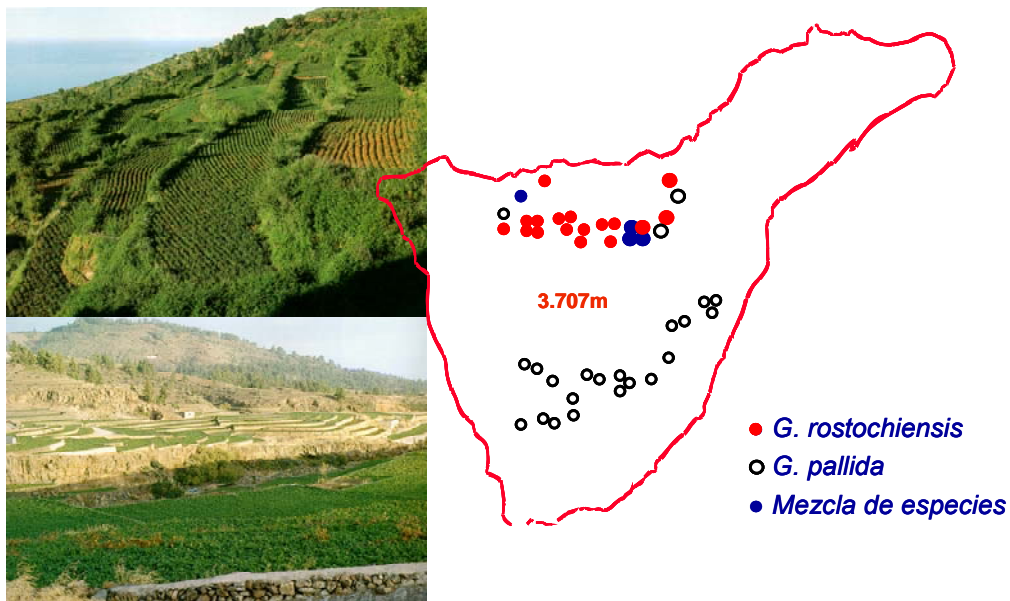


Figura 4.- Landscape diversity and virulence selection in cysts forming nematodes (*Globodera*) in Tenerife, with the use of potatoes resistant cultivars "Cara" and "Red Cara"

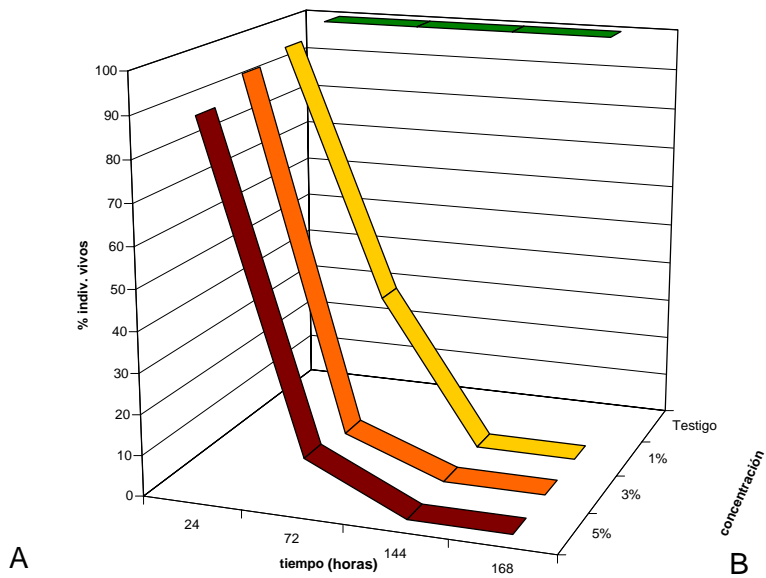


Figure 5.- Nematostatic effect of sugar beet vinasses on *M. incognita*, "in vitro" tests (Díez-Rojo 2006)

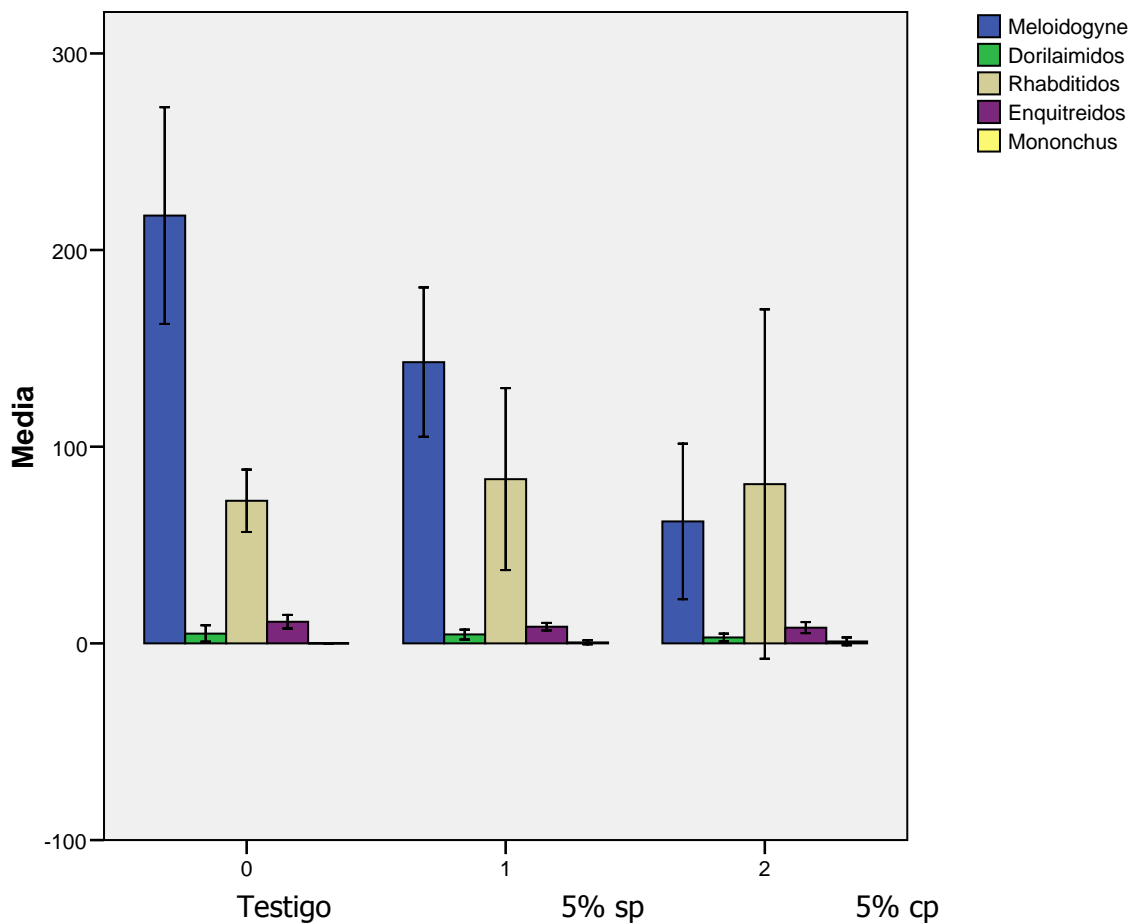


Figure 6.- Soil biodisinfection with sugar beet vinasses ($1,5 \text{ kg m}^{-2}$) in Marchamalo (Guadalajara), 3rd year; sp = without plastic cover; cp: with plastic cover

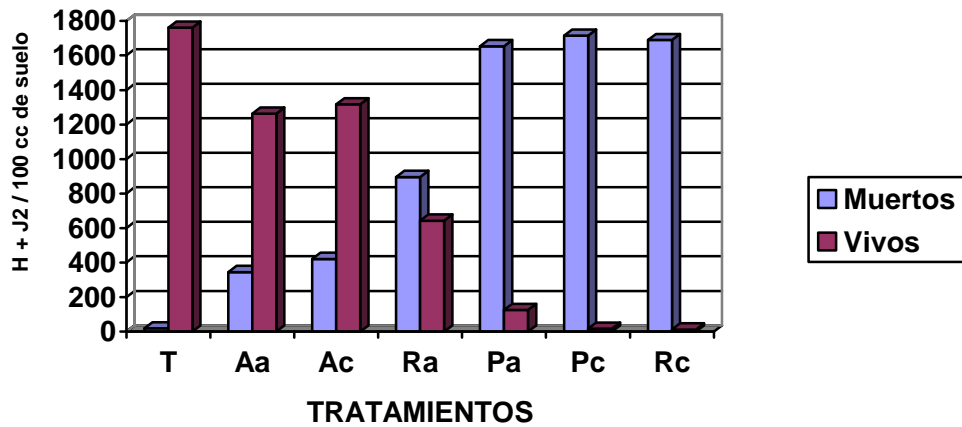
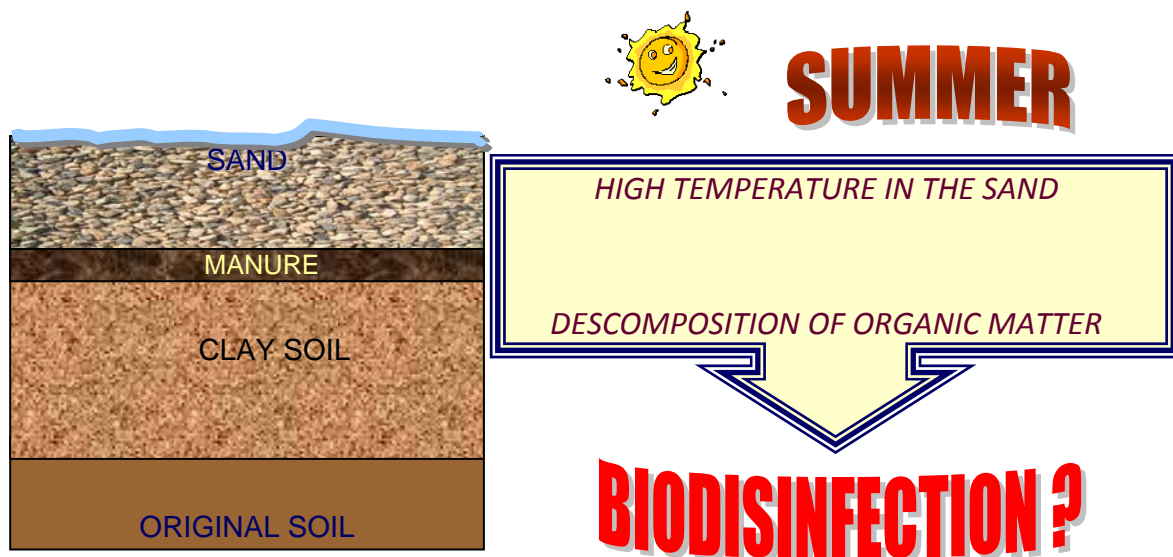


Figure 7.- Effect on *Globodera rostochiensis* on pig slurry land application (P); *Raphanus sativus* green manure (R); and blank control (A). Experiment realized in open (a) and close (c) pots, after 60 days of chamber disinfection at 30°C (lives and dead organisms 100 cc)



Figure 8.- Banana crops in "sorribas", (Barranco de las Angustias, La Palma). Typical Canarian agrarian structure of great efficacy to regulate problems caused by soil pathogens



DELAY: TO BRING NEW LAYER OF MANURE

Figura 9.- Functional aspects of "sanding" soils in Almeria related to the soil biodisinfection (Torres *et al.* 2007)

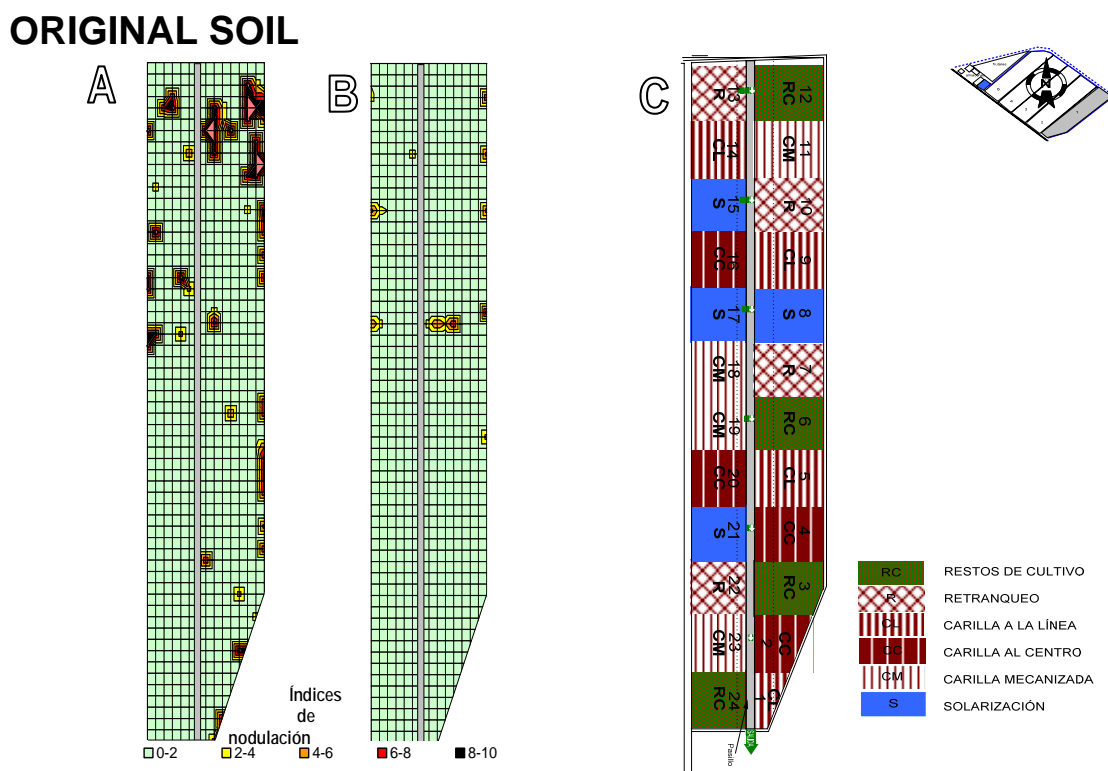


Figura 10.- Experimental greenhouse, nodulation maps: A. before treatments, B. after organic matter application combined with solarization; C. after treatments (Torres *et al.* 2007)

