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The impact of biological fungicide on growth and development of selected winter peas

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TITULO: El impacto de biofungicidas en el desarrollo de distintas variedades de guisante de invierno.

RESUMEN

En este trabajo se pretende analizar el comportamiento de distintas variedades de guisante de invierno en una zona geográfica concreta. Al mismo tiempo, se quiere poner a prueba el impacto en el desarrollo de estos dos fungicidas distintos, Polyversum y Serenade. Se ha estado llevando a cabo un experimento de campo durante el período 2018/2019 en la Estación Experimental de la Universidad de Agricultura Rolniczy en Prusy, Cracovia, Polonia. El suelo se clasifica como un chernozem degradado, generado a partir de arcilla y caracterizado por un alto contenido de nutrientes disponibles (pH = 6.3, carbono orgánico = 2.2, P205 = 15.3, K2o = 26.1, Mg = 10). Se han ido aplicando las faltas de fertilización mineral. Este ha sido un experimento de campo de dos factores aplicados en bloques al azar con tres repeticiones. La parcela es de 10m² el cultivo que precedía a este en la rotación era el trigo de invierno. El primer factor es la aplicación del fungicida (Polyversum, serenade y un sector de control), mientras que el segundo factor es el tipo variedades seleccionadas de guisantes de invierno. Las mediciones se llevaron a cabo durante el período de vegetación en las etapas características de la planta y fueron las siguientes: LAI (Índice de área foliar) es evaluado por LI-COR, también se mide el peso y la longitud de la planta. El contenido de clorofila se mide con el equipo SPAD Minolta.

Este trabajo ha sido llevado a cabo en la Universidad Rolniczy de Agricultura de Cracovia, en el departamento de agricultura sostenible, suelos y ciencias ambientales. Ha sido tutorizado por la profesora Agnieszka Klimek-Kopyra.

Palabras clave:

Fungicida; guisante; invierno; polyversum; serenade; Bioestimulante; variedades; Cracovia; biofungicida

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Location and date: Krakow, 25 January 2019

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Title (En): The impact of biological fungicide on growth and development of selected winter peas

ABSTRACT

This project aims to analyze the behavior of different varieties of winter peas in a specific geographical area. At the same time, we want to test the impact on the development of these two different fungicides, Polyversum and Serenade.

A field experiment has been conducted in 2018/2019 at Experimental Station of Rolniczy University of Agriculture in Prusy, Krakow, Poland. The soil is classified as a degraded chernozem, generated from loess and characterized by a high content of available nutrients (pH=6.3, organic carbon = 2.2, P205=15.3, K2o=26.1, Mg=10). Lack of mineral fertilization is applied. A two factorial field experiment is conducted in the randomized complete block design with three replication. The plot is 10m². The precrop was winter wheat. The first factor is fungicide application (Polyversum, serenade and control), whereas the second factor is 7 selected winter pea varieties. The measurements we have carried out are during the vegetation period and in characteristic plant stages. We measured LAI (Leaf Area Index) by LI-COR, also the weight and length of the plant are measured. Chlorofil content is measured by SPAD Minolta equipment.

This work has been carried out at the Rolniczy University of Agriculture in Krakow, in the department of sustainable agriculture, soil and environmental sciences. He has been tutored by Professor Agnieszka Klimek-Kopyra.

Key Words:

Fungicide; Pea; winter; polyversum; serenade; Bioestimulator; varieties;

Krakow; biofungicide

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1. INTRODUCTION

1.1. FUNGICIDES

1.1.1. Definition of fungicides

Fungicides are toxic substances that kill or prevent the growth of fungi and their spores. Fungicides work in different ways, but most of them damage fungal cell membranes or interfere with energy production within fungal cells.

Not all fungal diseases can be properly controlled with fungicides. For example, vascular diseases (wilting) caused by the genus *Fusarium* spp and *Verticillium* spp. They can only protect from non-infected new tissue diseases. In addition, few fungicides are effective against pathogens after they have infected the plant. Fungicides that have curative properties, which means that they are active against pathogens that have already infected the plant, tend to present an increased risk to pathogens to develop resistance to this type of fungicide.

1.1.2. History of fungicides

The properties of sulphur dust, as an effective fungicide against the so-called white diseases (one of the types of *Oidium*) were already known and used since the time of the Roman Empire. Those who had the product easily, given that Italy was and still is, one of the world's major powers, in terms of sulphur production.

Towards 1845 the annihilation of potato cultivation, caused by the fungus of mildew (*Phytophthora Infestans*) in Ireland, provoked a time of famine and emigration, of much of the Irish population and motivated the interest to find new fungicide products. This is often done empirically and the common salt was even tested and unsuccessful. Finally, it was possible to verify the effectiveness of what was called Bordelais broth, based on a suspension of copper sulphate and hydrated lime.

The Bordeaux mixture, also known as Bordelais broth, developed in 1882 and composed of dead lime and copper sulphate, was the first effective fungicide. For many decades it was used in a wide variety of plants and fruit trees.

The progress of the chemistry led, the introduction of new products, although inorganic, for the control of fungi and since the late 19th century and until the end of the Second World War the farmers used arsenical compounds and mercurials with also insecticide action to fumigate crops. All these products, although they were very effective, were extremely toxic and are currently prohibited or strictly limited to use.

The third generation of fungicides belongs to a highly developed and diversified organic chemistry. It already incorporates, from 1970, the systemic character, the penetration of the product and its distribution by the conductive vessels throughout the plant, greatly increasing the effectiveness of the treatment.

The main drawback of all these new products is that they can end up generating a resistance of the fungus in the case of repeated use of the product.

For the first time, the products were on the market with controls and an assessment of their dangers. It has been shown that in the long term, some substances are harmful to health, so many preparations used for years have been finally withdrawn. The study of damage to the environment, especially in terms of chemical compounds originating in the degradation of the commercial product, were at this stage practically unattended.

As in other agrochemicals, especially insecticides and herbicides, the main disadvantage is for the workers who apply the products, because apart from a timely intoxication there may be another cumulative character.

Currently, the research is directed towards a new line of fungicides, based on the mechanisms of antifungal control present in nature, such as the activators of natural resistance of plants or acquired systematic resistance (ASR). This implies the coordination between diverse research groups very specialized in diverse scientific fields mainly of the biochemistry.

1.1.3. Classification of fungicides

Fungicides are categorized in various forms based on different characteristics. The most commonly used features and their categories are described below (Mobility in the plant, protection function, range of activity, mode of action and chemical group)

Mobility in the plant: Contact or systemic. Contact fungicides (also called protectors) remain on the plant's surface and prevents the fungi to enter in the tissue and have no after infection activity. Repeated application is required, new growth is not protected. Many contact fungicides are potentially phytotoxic (toxic to the plant) and can damage the plant if they are absorbed.

Systemics (also called penetrating) are absorbed into the plant. Some systemics are mobilized at short distances from the site of their application, for example through foliar lamina from one surface to the other (local systemic or translaminar). Some fungicides are weak systemic and are mobilized, from the sites of application, a little more than the local systemic ones. The systemics whit phloem movement present a bidirectional mobility, some materials move in the phloem, the place where they were applied in the leaf, to other leaves or to the roots. Systemics do not mobilize again after translocation.

Most systemics are mobilized more effectively because of their translocation by the xylem tissue. When applied to the root zone, they are absorbed by the roots and mobilized upward by the transpiration current of the plant (systemic xylem movement). The systemic whit xylem movement applied to the foliage are mobilized along the leaf where they are deposited, but they cannot be redistributed outside, however, any material deposited on the stems can move towards the leaves.

Protection function: preventive or curative. Contact fungicides are products suitable for preventive use, as they work by contact on the surface of the plant where they have been applied. Repeated applications are required to protect the plant's new growth and replace the material that has been washed by rain, irrigation or degraded by environmental factors such as sunlight. Some contact fungicides are identified as "residual" products since the fungicide applied remains on the surface of the plant, occasionally as a visible residue, for several days. Due to the ability to penetrate the plant tissues, some systemic fungicides present both a

preventive and curative (eradicating) activity, in this way it can affect the pathogen after its infection.

Range of activity: one point or multiple action points. Fungicides that act in a unique way are active only at a point in a metabolic pathway in the pathogen or affect an enzyme or protein essential to the fungus. Due to the specificity of toxicity of these fungicides, these have very little effect in most organisms, can be absorbed safely by the plant, so tend to present systemic properties. As a result of this specific activity, fungi are more likely to be resistant to the fungicide, because a simple mutation in the pathogen usually allows it to overcome the effect of the fungicide, as for example preventing it from binding to the site of activity in the fungus.

Typically the ancient contact fungicides present multiple action sites, so they usually affect many fungi in different classes. Through the development of different researches and due to the increased rigor and number of regulatory tests required to register a new active ingredient, fungicide manufacturers have recently found it easier to develop single-site systemic fungicides. As a result of this approach, resistance to fungicides has become a subject of great importance in disease management.

Mode of action. Fungicides "kill" fungi by damaging their cell membrane, inactivating essential enzymes or proteins or interfering with key processes such as energy production or respiration. Others impact specific metabolic pathways such as sterol or chitin production. Some recent development products are unique because they do not directly affect the pathogen. Many of them obtain a response from the host plant known as "systemic acquired resistance" (SAR). These systemic acquired resistance (SAR) inducers basically imitate chemical signals in plants that activate plant defense mechanisms such as the production of thicker cell walls and anti-fungal proteins. The usefulness of SAR inductors, however, has been limited so far because many pathogens are able to overcome these defenses.

Chemical group: inorganic or organic. Fungicides can also be classified according to their chemical composition. Chemically, organic molecules are those that contain carbon atoms in their structures, while inorganic molecules do not present them. Many of the first fungicides developed were inorganic compounds based on sulfur or metal ions such as copper, tin, cadmium and mercury that are toxic to fungi. The copper and sulfur are still widely used. Most of the fungicides used today are organic compounds and therefore contain carbon. The term "organic" is used here based on chemical terminology and differs from the term "organic" used to describe an agricultural system that seeks to be holistic and increases the health of the agroecosystem.

1.2. THE WINTER PEA

1.2.1. Origins and dissemination

The origin of the green pea (*pisum sativum*) is related to the Middle East and Central Asia, areas where it has been cultivated for thousands of years, currently forming part of some of the traditional recipes of the eastern countries. Although these first plantations date back to the eighth millennium BC, it would be advanced in II BC when it would spread across Europe.

The ancient Greeks cultivated it and ate it, calling it "pison." Then it was cultivated by the Romans (300 years BC), among whom it was known as "Pisum", consumed as puree. The Latin writer Marcus Terencius Varro, author of the book "agronomica", details the cultivation of the pea (50 years BC), and the uses of its stems and seeds in both human consumption and livestock. The Roman writer Lucio Moderateto Columella in his book "De Culto Hortorum" (1st century BC), describes his cultivation, use and virtues.

Its use is recent in Europe, having been probably introduced from Palestine or Egypt in the Eastern European areas of the Mediterranean, an area that is considered as its main centre of diversification. However, it is very old its use in the villages of India, from where they were introduced in China.

The cultivation of the pea in England, dates from the conquest of the island by the Roman soldiers. The Roman writer Marcus Gavius Apicius, author of the book "De Re Coquinia", provides fifteen recipes for cooking and preparing peas, which were highly appreciated for their culture. It is said that the extravagant Roman Emperor Heliogabalus, famous for his sumptuous banquets where the waste was the norm, served the stewed peas mixed with gold grains or seasoned lentils with gemstones.

Until the 16th century the pea was used as dry grain and as fodder, and thereafter began to use fresh grain. The peas were one of the favorite foods of the French king Louis XIV and his wife M^a Theresa of Austria, who in his Palace of Versailles, gave sumptuous banquets in which the peas were part of the garrison of meat prepared for these events and presented in refined porcelain dishes. The size and color of the pea's own grain pleasantly surprised all the diners to such an extent that in the 17th century the Sun King (Louis XVI) had immediately prepared a recipe for his most loved ones. The name given to this legume was "petit-pois" to differentiate it from the dry and rough pea consumed by the plain people.

The first English colonizers took the pea to North America in the 17th century, where it prospered thanks to the temperature and humidity of its climate spreading through many States of the Union. It was constituted in the food preferred by the Indians, who ate it cooked with pieces of buffalo meat. As a curiosity, in the well-known tale "Cinderella", it is punished ordering to clean and to devastate peas, work considered inferior and to the service of the delight of the nobles.

1.2.2. Taxonomy and morphology

Kingdom	Plantae (plants)
Subkingdom	Tracheobionta (Vascular plants)
Superdivision	Spermatophyta (seed plants)
Division	Magnoliophyta (flowering plants)
Class	Magnoliopsida (dicotyledons)
Subclass	Rosidae
Order	Fabales
Family	Fabaceae (pea family)
Genus	Pisum L. (Pea)
Species	Pisum sativum L. (garden pea)

Table 1 Taxonomy of winter pea

The peas belong to the family Fabaceae, being its scientific name *Pisum sativum* L. The pea belongs to the genus *Pisum* (tribe Viciae).

Currently, recognize two species within the genus: the cultivated pea *Pisum sativum* and *P. fulvum*. In the cultivated, in turn, two subspecies are distinguished: *elatius* and *sativum*, the latter with two botanical varieties: *arvense* and *sativum*; the commercial varieties grown in the developed world belong to this last, although those of the *arvense* have been and are forage.

The stems are climbing and angular; Regarding the vegetative development there are some varieties of limited growth and others of indeterminate growth, giving rise to three types of varieties: dwarf, mid-sized and climbing.

The root system is not very developed, although it has a pivoting root that can be quite deep.

The leaves have pairs of leaflets and end in tendrils, which have the property of grabbing the tutors they find in their growth.

Each cluster usually carries 1 or 2 flowers, but there are also cases of three, and even 4 and 5, although the latter are rare.

The flowers consist of 5 sepals, with the two upper variables, both in form and in dimensions, which is used as a varietal character.

The pods are 5 to 10 cm long and usually have 4 to 10 seeds; they are of variable shape and color, according to varieties; With the exception of the "tirabeque", the shells of the pod have a parchment that makes them inedible.

Pea seeds have a slight latency; the average weight is 0.20 grams per unit; the germinative power is of 3 years maximum, being advisable to use for the sowing seeds that have less than 2 years since their harvesting; in wrinkled grain varieties the germinative faculty is even lower.

1.2.3. Economic importance and geographical distribution

The pea is an annual species, which occupies a large area of cultivation, since it extends almost all over the world. This crop has been acquiring greater importance in the industry, both canning and freezing.

Countries	Production of Green Pea in 2001 (tones)	Production of Green Pea in 2002 (Tones)
India	3.800.000	3.800.000
China	1.541.280	1.661.280
United States	885.000	787.715
France	474.000	418.000
United Kingdom	388.000	352.000
Hungary	283.425	280.000
Egypt	240.000	227.135
Belgium	144.000	150.000
Perú	82.559	80.909
Denmark	80.000	80.000
Morocco	79.000	68.570
Netherlands	76.800	75.000
Italia	70.902	70.318
Pakistan	70.716	72.128
Australia	65.000	65.000
Argelia	63.290	63.000

Deuthsland	62.200	61.900
Turkey	60.000	55.000
New Zeland	60.000	45.000
Spain	53.400	52.300
Canada	51.971	52.000
Mexico	48.400	49.000
Sweden	36.405	36.405
Chile	32.000	32.500
Bolivia	30.307	27.449
Filipines	29.000	29.000

Table 2 Production of winter pea

1.2.4. Soil and climate conditions

The crop develops between 6 and 30 °C, with optimum temperatures of development and reproduction included in the 16-20 ° C intervals for the day and 10-16 °C for the night. In general it does not support temperatures well above 30 ° C, negatively influencing above all in the quality of the green bean. The traditional varieties (grain) support the winter temperatures well, but evidently they are a high risk for greening.

Like most legumes, peas prefer soils of light or medium texture well drained and aerated, with pH between 6 and 7. Soils with high calcium levels produce chlorosis and harden the grain. It is a crop very sensitive to soil compaction, reducing growth and foliar area, as well as the number of flowers in the plant.

1.3. LEGUMES

1.3.1. Origins and dissemination

The legumes of grain conform an important group of alimentary crops that have played a fundamental role in the diet of almost all the civilizations of the world, for more than 20 thousand years. Archaeological findings and the iconography of ancestral cultures indicate that they were food basics in ancient Egypt, in ancient Greece and in America; very appreciated in the Inca, Mayan and Aztec cultures, for more than 5000 years.

Although some historians speak of an age of 20,000 years of this crop, what does seem proven is that in the Near East, about 10,000 years ago, there was a clear association between cereal seeds such as wheat or barley with legumes such as lentil or the peas, a crop that also reaches the beans 4,000 years before our Era.

The Egyptians, as evidenced by many vestiges found for that purpose, professed true veneration for the lentils. They were also very appreciated by the Romans, since it seems that the same ship that transported an obelisk from Egypt to Rome, during the reign of Caligula, also carried more than 800 tons of lentils.

The beans, however, were considered by the Egyptians as a despicable food. The priests did not eat them, although the people did. The Greeks or Romans also considered them despicable. Peas were common food in Rome, but not very appreciated.

The beans, cultivated by the pre-Columbian American civilizations, especially in Mexico and Peru, from 8,000 years before Christ, were not only used as food but also as currency. And, although historians do not seem to be able to agree, the truth is that they enter Europe in the years after the Discovery and soon form an inseparable part of what is now called the Mediterranean Diet.

Soy is proud of being the first legume of which written record was left: Shen Nung's books, dating from the year 2800 BC, describe the five main and sacred crops of China: rice, soybeans, wheat, barley and millet. With it, the ancients prepared high protein preparations (cottage cheese, sauces, cheeses, pasta) used to flavor and enrich their basic food in cereals. It is around the fourth century BC when they devised methods to extract their oil.

The Italian writer Humberto Eco assures that the legumes saved Europe, during the Middle Ages, from its extinction, since epidemics, wars and famines could only be fought thanks to legumes. "Without beans," Eco says, "the European population would not double in a few centuries and we would not be hundreds of millions at the moment."

The legumes in Spain, from the Middle Ages to our days, accompanied the meals of households, whether they are the most affluent or the most popular. Dishes such as "cocido"; "potajes" or "lentejas" are part of our traditional culinary culture and occupy hundreds of thousands of pages in Spanish literature.

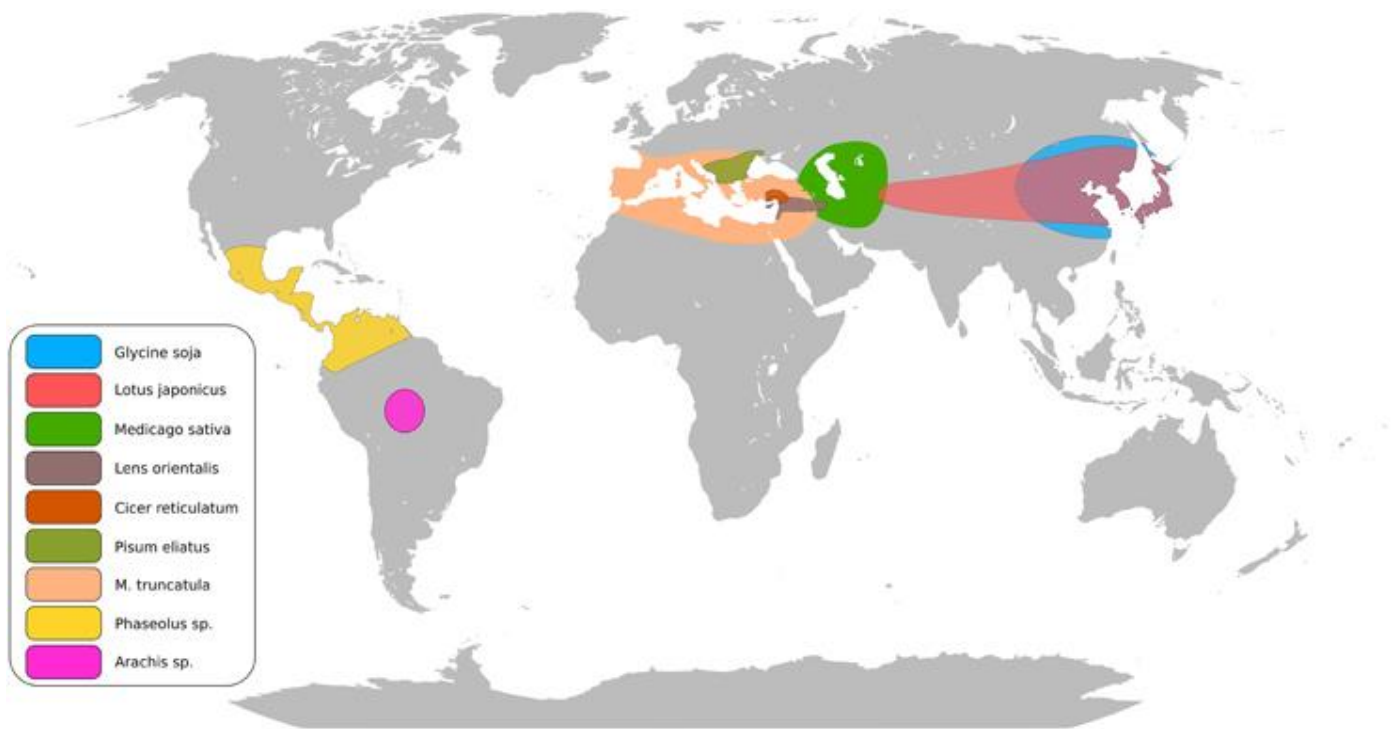


Figure 1 Geographical distribution of legumes

1.3.2. Area in Europe and in the world

Legumes are the second most important family of agricultural crop species after grasses worldwide and are used for both grain and forage, but they are underrepresented in European cropping systems which are dominated by cereals and nonlegume oilseeds. This situation has been the subject of public debate in Europe over the last decade and it is now widely acknowledged that grain legumes have the potential to contribute more to European agricultural systems by improving the agronomic performance of cropping systems and providing protein-rich food and feed and helping to reduce European dependence on imported protein.

Grain legume production contributes widely to ecosystem services by its low reliance on synthetic fertilizers; reduced greenhouse gas (GHG) emissions; increased diversification of the crop rotation with concomitant increases in above- and below-ground biodiversity and changes in weed, pest, and disease pressures; and changes in soil fertility and carbon storage.

The area of grain legumes in Europe declined almost continuously over the last 5 decades, from 5.8 million ha in 1961 (4.7% of the arable area) when recording began to 2.0 million ha in 2014 (1.6% of the arable area)(FAOstat, 2016). A major underlying driver behind the reduction in the proportion of arable land used for legume crops is the increased comparative advantage in the production of starch-rich cereals in Europe over the production of protein-rich grain legumes. Between the 1970s and the 1990s, wheat yields in north-western Europe increased annually by about 0.15 t/ha(Supit, 1997).

The decline in human consumption of Legumes in Spain and Europe has been associated with an increase in meat consumption from 20 kg / person / year in the 60s, to 50 in the 80s, after which a tendency to stabilization, being 51 kg / person / year at present. This higher

consumption of meat meant an increase in the consumption of legumes for animal feed, but not in its cultivation. This was solved by the feed industry with the importation of soybean (*Glycine max*), which has continued to increase since the 70s, which, in turn, has not helped much to the development of the cultivation of other legumes for feed. And as in everything, there is no development without investment in innovation.

This downward trend in the cultivation of legumes in Spain is similar to that found in the rest of Europe, although it contrasts with the world situation where, although the cultivation of beans and pea falls, spectacularly increases the cultivation of soybeans, and in general of all tropical legumes, but also of other cold climates such as lentils and chickpeas (Rubiales and Mikic, 2015). The decline of legume cultivation in Europe generates two disturbing dependencies. On the one hand, the consumption of nitrogen fertilizers in Spain was higher than 800,000 tons in 2012, of which we imported 65%. In the EU, consumption exceeded 10 million tons per year, of which 81% was imported. In addition, the production of these fertilizers requires great energy, basically natural gas of which we are also dependent on the outside world, being imported both in Spain and in the EU of the order of 60% of their needs. On the other hand, the EU imports 70% of the vegetable protein it consumes, of which not all are legumes.

Spain consumes more than 4.8 million tons of legume grain per year, while it does not even produce 0.3. The great majority of legume consumption in Spain corresponds to soybean (4.3 million tons), followed at a long distance by dry pea (0.2), chickpea and lentil (0.1).

In an attempt to correct this decline in the cultivation of legumes, the EU has been creating a series of aid in different formats, sometimes based on subsidies to the surface, others to production, which have had a persuasive effect while the aid has lasted, but that have not helped to consolidate the sector. Currently there is the possibility of a help of about € 100 / ha for quality legumes (beans, lentils and chickpeas) under a protected name or in organic farming, which today represents only about 5,000 hectares, with the majority (more than 80% of the total) of the production of quality legumes for human consumption (lentils, chickpeas and beans) outside these areas subject to protection that can receive this aid. In addition, the current Common Agricultural Policy (CAP) has benefited the cultivation of legumes with the aid associated with the Green Payment provided that certain environmental practices are respected, including the diversification of crops. This is awakening interest in the sector and there is a rebound in the cultivation of legumes. This unexpected demand is showing the weakness of the sector due to the lack of sufficient quantity of certified quality seed of modern varieties sufficiently adapted to our conditions, which has once again revealed the lack of planning. And, just as we can't base our production of feed on the importation of soya, we can't base our legume crop on the importation of techniques or varieties developed under other conditions. Although there are meritorious research and agronomy programs in legumes in Spain, there was a vicious circle in which the reduction of cultivation had reduced the investment in the development and registration of new varieties, as well as in the multiplication of certified seed.

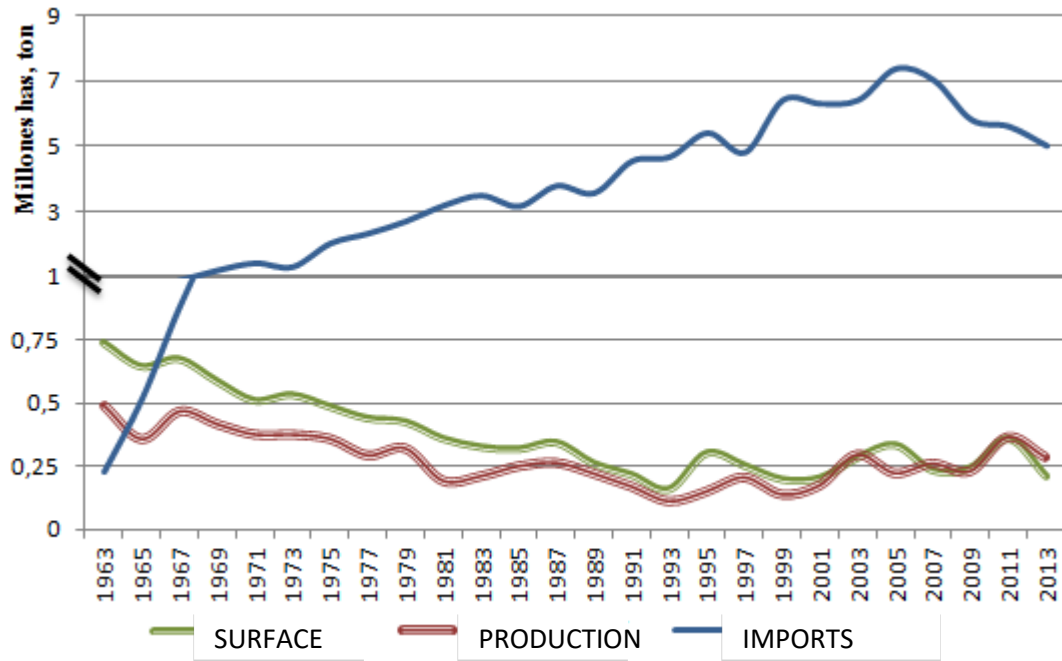


Figure 2 Evolution of the surface, production and imports in Spain in the last 50 years

The total acreage of legume grain in Spain has decreased worryingly in the 50-year series studied. At present, around 200,000 hectares of grain legumes are grown in Spain, 530,000 less than in the 1960s.

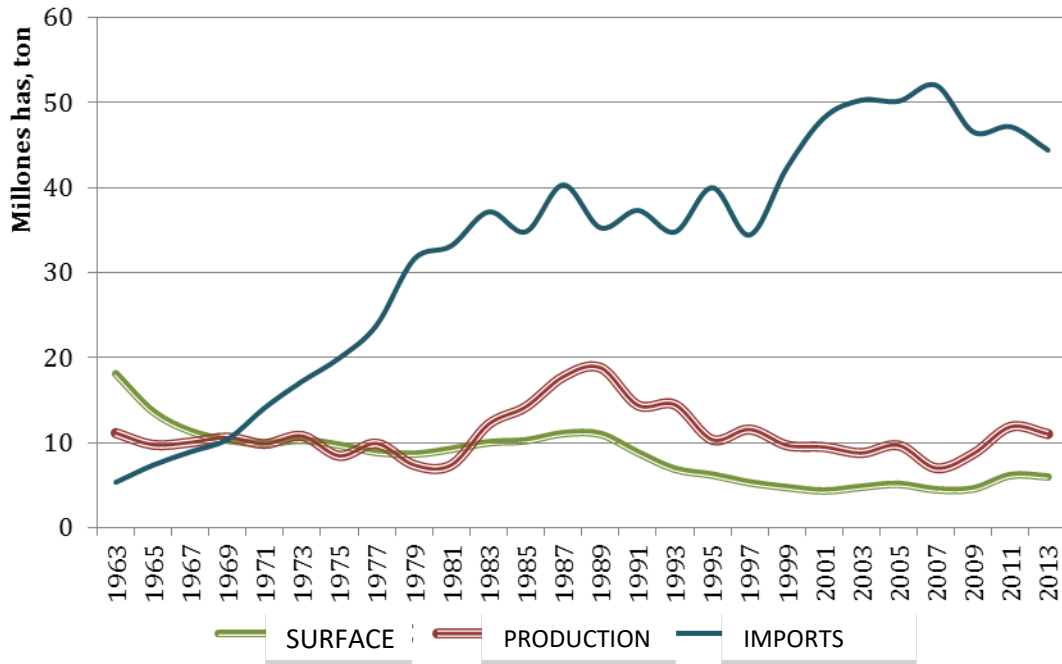


Figure 3 Evolution of the surface, production and imports in Europe in the last 50 years

In Europe, the acreage of legume grain also falls sharply (10.4 million hectares) in the 50 years studied, currently cultivating 5.7 million hectares, in contrast to what happens worldwide where the surface has progressively increased over the years, currently cultivating around 170 million hectares.

Despite the smaller area devoted to the cultivation of grain legumes in Spain, the production, although with fluctuations, we can say that it has decreased to a smaller scale than the surface, currently producing more than 280,000 tons. At European level we find that, although the cultivated surface decreases, the production is maintained and even increases (being of 10.7 million tons at present), thanks to the improvement of the yields, which is superior to that of Spain. In the world, production increases on a larger scale than the surface, producing 336 million tons of grain legumes at present.

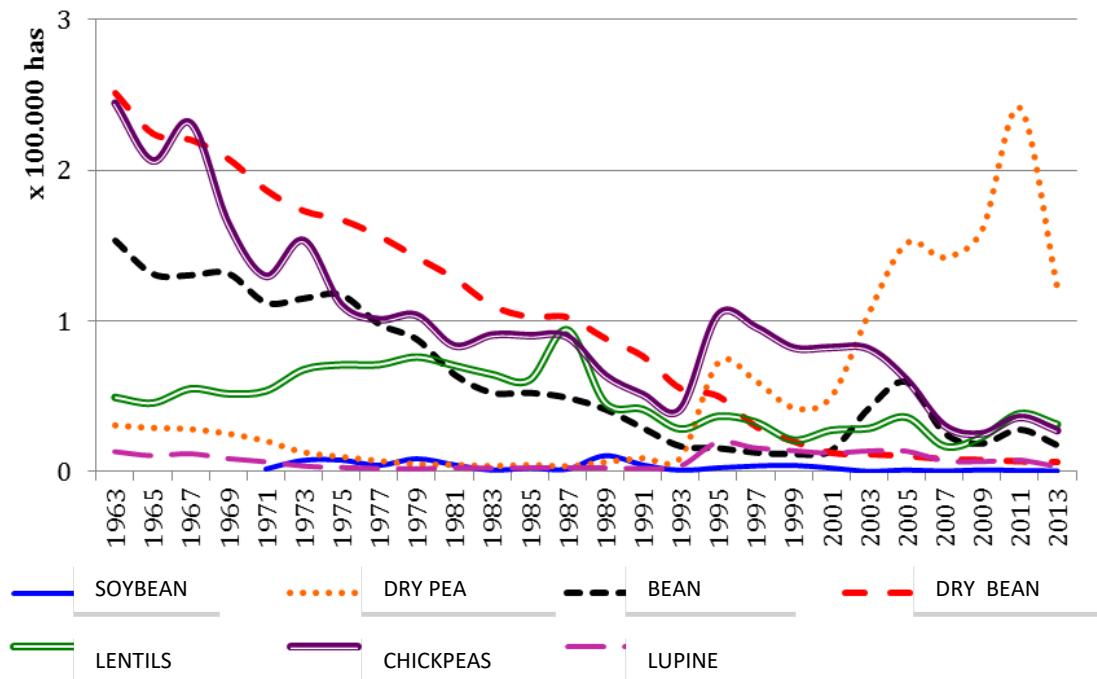


Figure 4 Evolution of the surface used for legumes in Spain during the last 50 years

Soy is the most consumed legume and at the same time least cultivated and produced in Spain (only 500 hectares and 1,400 tons in 2013), making it the most imported. We import on the order of 4.7 million tons annually, representing 96% of the total volume of legume grain imports. The possibilities of expansion of the cultivation of soy in Spain are very limited to be a summer crop with high water needs. Even so, the possibilities of expansion of the soybean at national level have a clear ceiling since our greater surface of crop is of dry land.

The only alternative in the medium-long term would be the development of varieties adapted to earlier plantings and with less water needs. In Europe, the cultivation of soybeans has experienced a spectacular increase in the last decade, being today the only legume whose surface increases, unseating even the pea. It is the most cultivated legume, with 3.2 million hectares, which accounts for more than 55% of the total hectares harvested from grain legumes, and the most produced, with 6 million tons currently. This has been linked to the efforts of producers in the countries of the Danube basin, where the weather allows the cultivation of soybean in summer with few risks. At a global level the increase in soybean cultivation began earlier, currently accounting for 65% of the total cultivated area of legume grain, greater than 111 million hectares. World production is higher than 278 million tons, with 80% of production concentrated between the United States (35%), Brazil (28%) and Argentina (17%). The current worldwide yield of soybeans is 2,500 kg / ha, with an annual gain of 25 kg / ha / year, with the legume having the greatest increase in yield in the years studied. In Spain, the yield is 2,800 kg / ha with an annual gain of 25 kg / ha / year, while in Europe it is 1,900 kg / ha with a gain of 32 kg / ha / year.

1.3.3. Benefits of legumes

Legumes are characterized by being Nitrogen fixers. This means that they produce this element for their nutrition and provide Nitrogen to the soil. To do so, they use rhizobia (*Rhizobium leguminosarum*) which are bacterias that form nodules in the roots of plants. These bacteria take nitrogen from the atmosphere to convert it to available nitrogen for the plant. While the plant provides organic components obtained by photosynthesis.

These bacteria called rhizobia can always be found in the soil and are "activated" when we sow legumes. When bacteria receive signals that there are legumes in the soil, they approach and enter the roots. It is called symbiosis, since these two organisms benefit mutually.

There are many species of rhizobia, each legume works with different ones. If we introduce a new legume to our plot, the rhizomes may take a while to generate but this improves with the years. To see if rhizomes form in the roots, we must observe the roots and the number of nodules that were formed.

Legumes are the main group of plants that are able to fix nitrogen from the air in the roots and transfer it to the soil. Being nitrogen the basic food of plants, by using legumes we are reducing the amount of fertilizer and benefiting our soil. These plants grow faster and some species are used to cover the soil. The root system is very deep, helping to support the plants and creating spaces in the soil for air and water.

In areas with eroded soils, compacted or with little organic matter, legumes are used to form the soil. In regenerative agriculture, legumes are an important basis for managing nutrients in the soil with rotations and crop associations. In addition, the flowers attract bees and beneficial insects, creating greater biodiversity in the crops.

2. AIM OF THE THESIS

We have carried out an experiment in the experimental station of the Rolniczy University in Prusy, Krakow. In this experiment we have included seven different varieties of peas to which we have applied two different biofungicides. In order to identify the effect of the biofungicides and the development of each of the varieties, three replications have been made per variety. One of the repetitions has been established as control and in the other two a biofungicide has been applied in each of them.

The main objective of this experiment is to be able to differentiate which are the most productive varieties, in order to adopt them in the future to improve the production of pea in the geographical area in which the experiment was developed. Biofungicides have also been applied to know which is a more useful tool to improve the pea crop, both healthily and productively.

The biofungicides selected for this experiment have been Serenade and Polyversum, for their antifungal qualities and their benefits for the health and nutrition of the plant, since they not only act on the fungi that attack the plant.

Due to the present problems both in Europe and in Spain, of a marked decrease in the surface and production of legumes in general, the main objective of this experiment is to provide the necessary tools and aids to increase the cultivation of legumes.

3. METHODOLOGY

As we all know, the pea is like many other legumes, it is extremely beneficial for the environment. This is due to the fact that it has wonderful qualities as atmospheric nitrogen fixing plants.

The main aim of this project is to find better adapted varieties of pea, both equally adapted to climate, soil conditions and the specific inputs, as fertilizers and pesticides. This investigation will help the farmers to improve their yields and efficiency. Therefore they will be more motivated to increase the cultivation of legumes, as it is a crucial issue the decrease of Europe legumes cultivation in the last decades.

A field experiment was carried out in the years 2018-2019 in the Experimental station of Agriculture University in Prusy, near Kraków (47°24' N lat., 7°19' E long., 300 m a.s.l.). The soil of the experiment was a Haplic Phaeozem characterized by pH 5.6, 9.4, 11.1 mg of available phosphorus, potassium and magnesium, respectively, per 100 g soil in the top soil layer. A two-factor field experiment was established in a randomized block design.

A two factorial field experiment was conducted. A first factor was the fungicide application (Serenade, Poliversum) and the second factor was the cultivars. Seven different cultivars ('Baltrap', 'Myster', 'Specter', 'Dexter', 'Aviron', 'Arka' and 'EFB33') were planted and used in the study.

The plot size was 10 m². The precrop was a winter wheat. Before sowing, non mineral fertilizers were applied. Sowing was carried out in the first decade of September. Fungicides (Serenade- at 5 L ha⁻¹, Poliversum- at 0.15 kg ha⁻¹) were applied twice (7 days intervals), in seedling stage in the first decade of October. The harvest was carried out in the third decade of June.

X	7☞	6	5❖	4■	3	2□	1○	X			POLYVERSUM
X	14	13☞	12□	11○	10❖	9■	8	X			SERENADE
X	21	20■	19❖	18	17☞	16○	15□	X			CONTROL

Table 3 Distribution of the field experiment

- BALTRAP
- MYSTER
- ☞ SPECTER
- DEXTER
- ❖ AVIRON

ARKA

⌘ EFP 33

4. RESULTS

1	BALTRAP X POLYVERSUM
2	MYSTER X POLYVERSUM
3	SPECTER X POLYVERSUM
4	DEXTER X POLYVERSUM
5	AVIRON X POLYVERSUM
6	ARKA X POLYVERSUM
7	EFP 33 X POLYVERSUM
8	SPECTER X SERENADE
9	DEXTER X SERENADE
10	AVIRON X SERENADE
11	BALTRAP X SERENADE
12	MYSTER X SERENADE
13	EFP 33 X SERENADE
14	ARKA X SERENADE
15	MYSTER X CONTROL
16	BALTRAP X CONTROL
17	EFP 33 X CONTROL
18	ARKA X CONTROL
19	AVIRON X CONTROL
20	DEXTER X CONTROL
21	SPECTER X CONTROL

4.1.1. Biomass measurements

In these graphics we can see the evolution of the average biomass of the

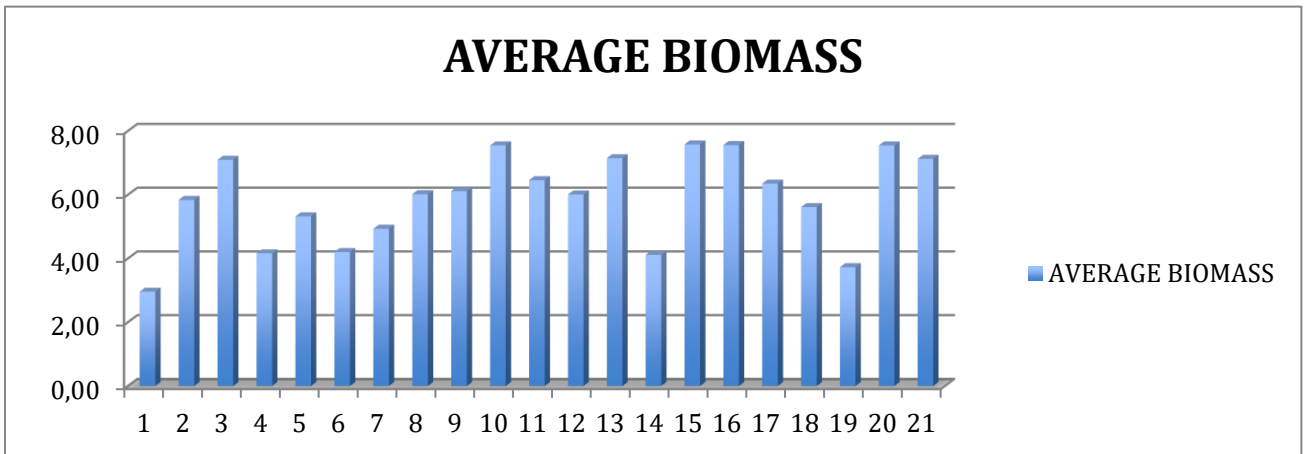


Figure 5 Average biomass measurements the 10th of april (seeding stage)

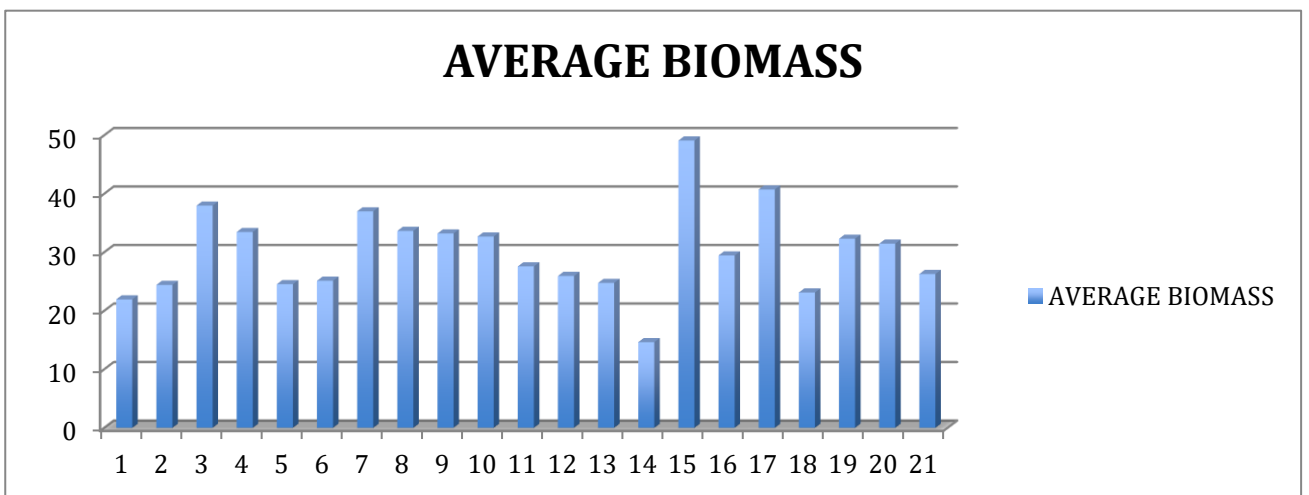


Figure 6 Average biomass measurements the 6th of may (budding stage)

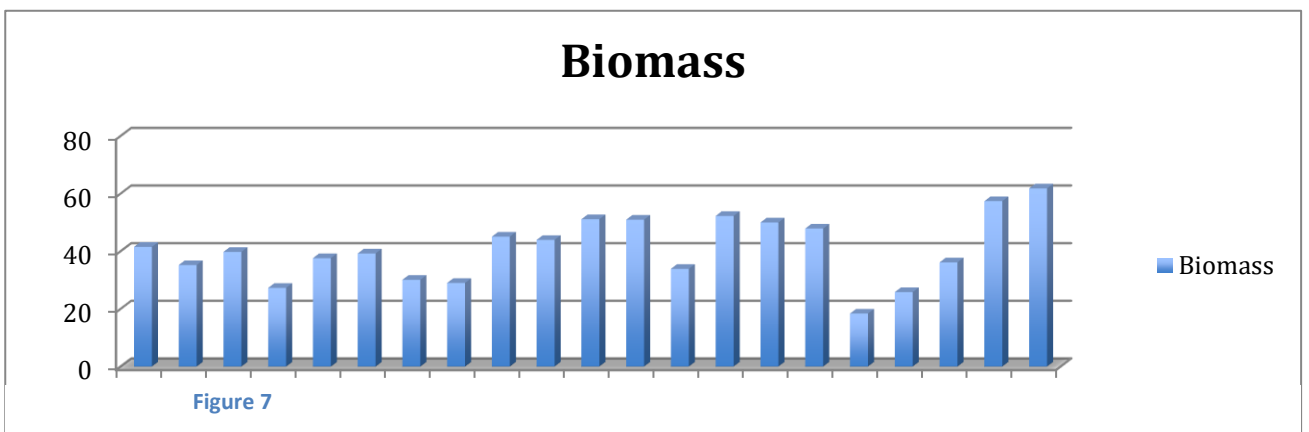


Figure 7

different varieties through the three measurements in which we measured the biomass of the fresh plants. The first measurement took place on the 10th of april (seeding stage) , followed by a second one on the 6th of may (budding stage) and finally the third the 20th of may (Flowering stage). The last measurement was made on the dry plants.

As we can see, the objects 10, 15, 16, 20 and 21 were the first ones to stand out in the first measurement. At the second one, the 15 (Myster x Control) was the object with a higher biomass by a huge difference. At the last measurement, The object 21 is finally the one to have a highest biomass in plant

4.1.2. Length measurements

First measurement

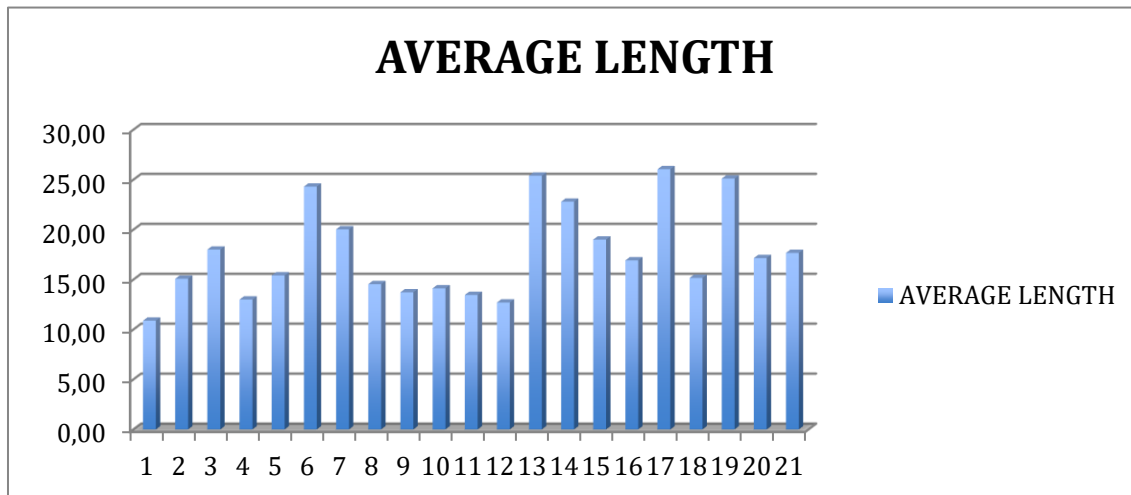


Figure 9 Average length measurement of 10th april (seeding stage)

Second measurement

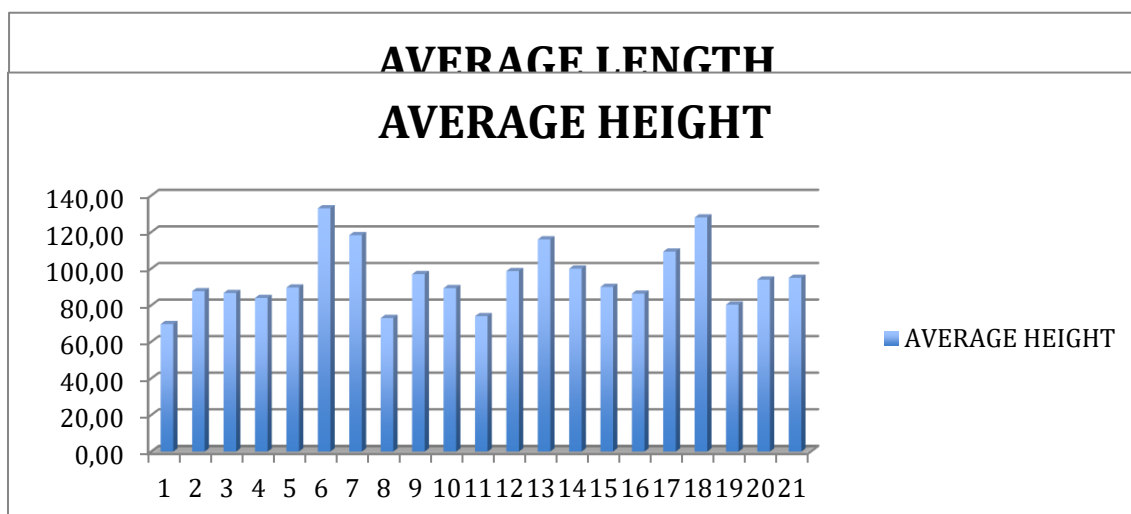


Figure 8 Average length measurements the 6th of may (budding stage)

Figure 10 Average height measurements the 20th of may (Flowering stage)

Third measurement

In the first measurements we can see that the varieties with a faster development were the 6, 13, 17 and 19. These are the Arka x polyversum, EFB 33 x serenade, EFB 33 x control and Aviron x control respectively. The EFP 33 is the variety with the faster length growth.

In the second measurement we can see that the 7, 13, 14, 17 and 19 were the largest. These are the EFP 33 x polyversum, EFP 33 x serenade, Arka x serenade and EFP 33 x control. As we can observe, the EFP 33 is the one with has a better vegetative growth.

In the third measurement we can see that the number 6 and 18 were the varieties with a higher height. These numbers are from the Arka varieties, we can see that this variety has a higher development in the last steps of growing. The numbers 7,

13 and 17, corresponding to the EFP 33 varieties with polyversum, serenade and control, are the ones with a better growth before the ones mentioned before.

4.1.3. Foliar area measurements

First measurement

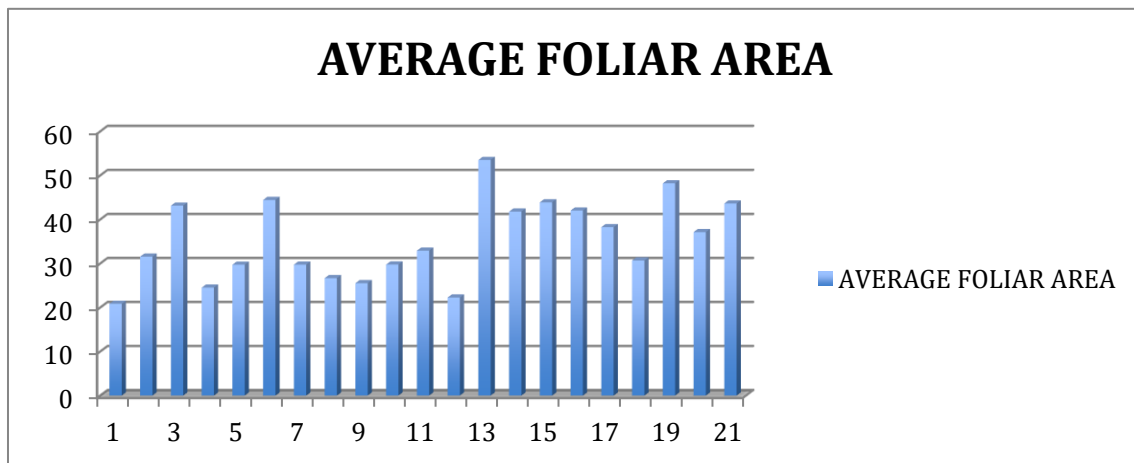


Figure 11 Average foliar area measurements of the 10th of April (seeding stage)

Second measurement

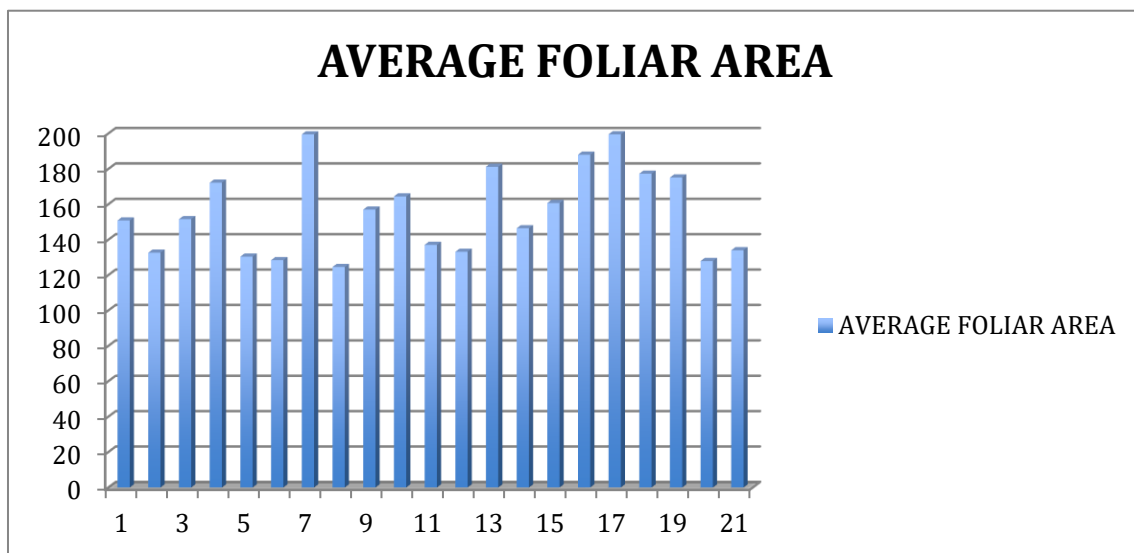


Figure 12 Average foliar area measurements the 6th of May (budding stage)

Third measurement

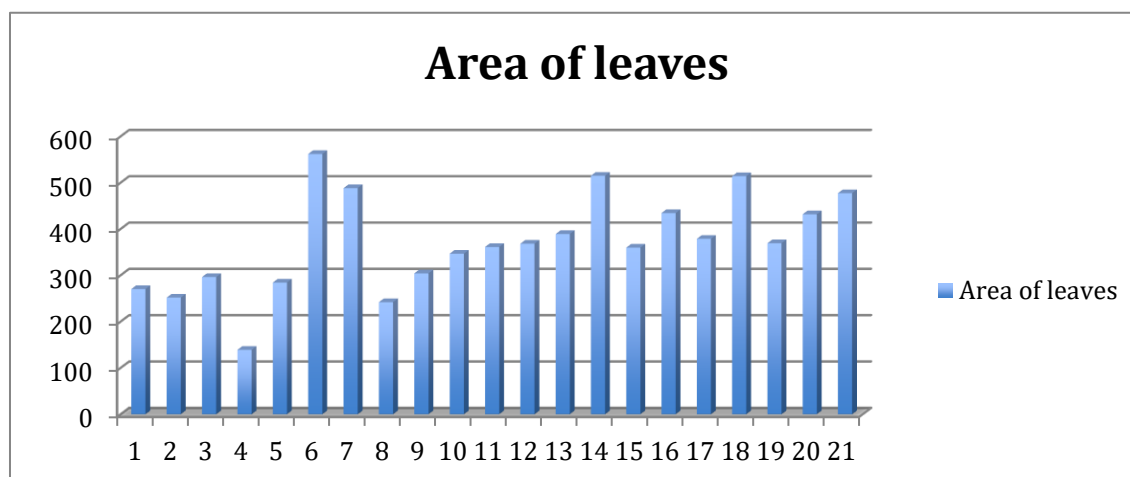


Figure 13 Average foliar area measurement the 20th of may (Flowering stage)

In the first measurement we can observe that the number 13 and 19 are the ones with higher values. These numbers are the EFP 33 x serenade and Aviron x control, respectively.

In the second measurement we can observe that all the objects have similar values but the ones with a higher value of foliar area are the 7, 13, 16 and 17. The varieties are the EFP 33 x polyversum, EFB 33 x serenade, Baltrap x control and EFB 33 x control. As previously in the length measurements, the EFB 33 is the one with a more significant development.

Finally, in the third measurement, the 6, 7, 14 and 18 are the objects with the highest foliar area. These are the ARKA x polyversum, EFB 33 x polyversum, ARKA x serenade and ARKA x control. We can confirm that the ARKA and EFB 33 are the varieties with the better vegetative development.

4.1.4. SPAD measurements

The SPAD Minolta is a compact meter designed to help users improve the quality of crops and increase crop yields by providing an indication of the amount of chlorophyll present in the leaves of the plant. The chlorophyll content of the leaves of the plant is related to the condition of the plant, and therefore can be used to determine when additional fertilizer is necessary. By optimizing the nutritional cultivated, resulting in a higher yield of higher quality crops.

The third measurement to place the 20th of may (Flowering stage), and it was the day we measured the content of chlorophyll.

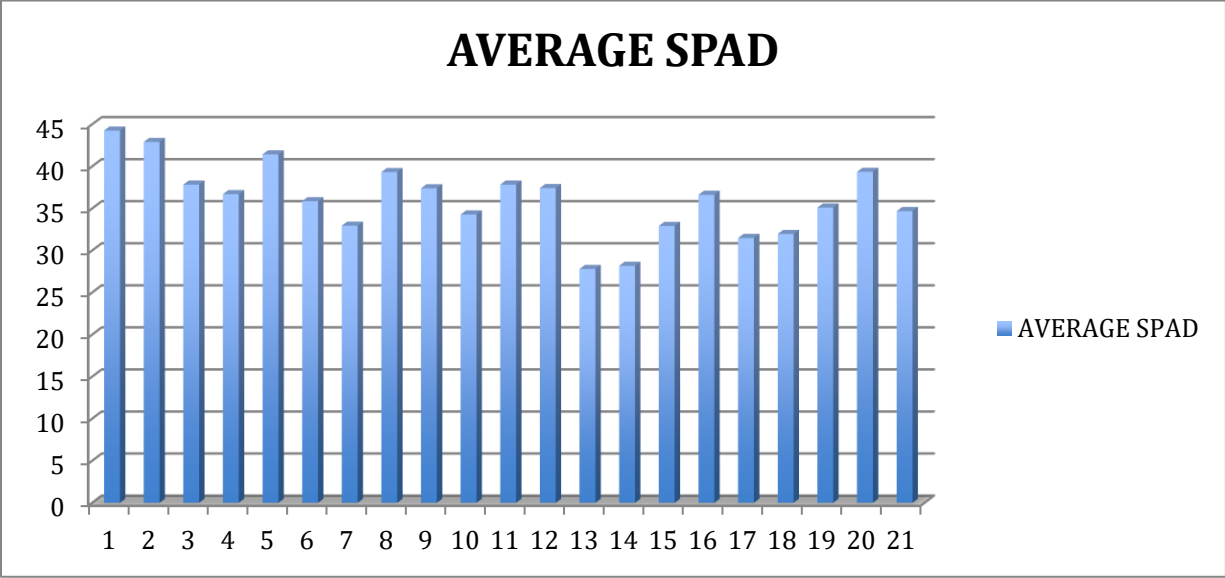


Figure 14 SPAD measurements the 20th of may (Flowering stage)

As we see in the graph, object 13 and 14 (EFP 33 x serenade and ARKA x serenade) are the ones with the lowest value of chlorophyll. The varieties Baltrap and Myster (1 and 2) had the higher values (44.25 and 42.9). These low values can be an indication of lack of nitrogen fertilization in some of the samples.

4.2 Maturity measurements

The 26th of June we made the final measurements. We measured the height, weight, number of seeds, weight of pods and weight of seeds to evaluate which is the most productive variety.

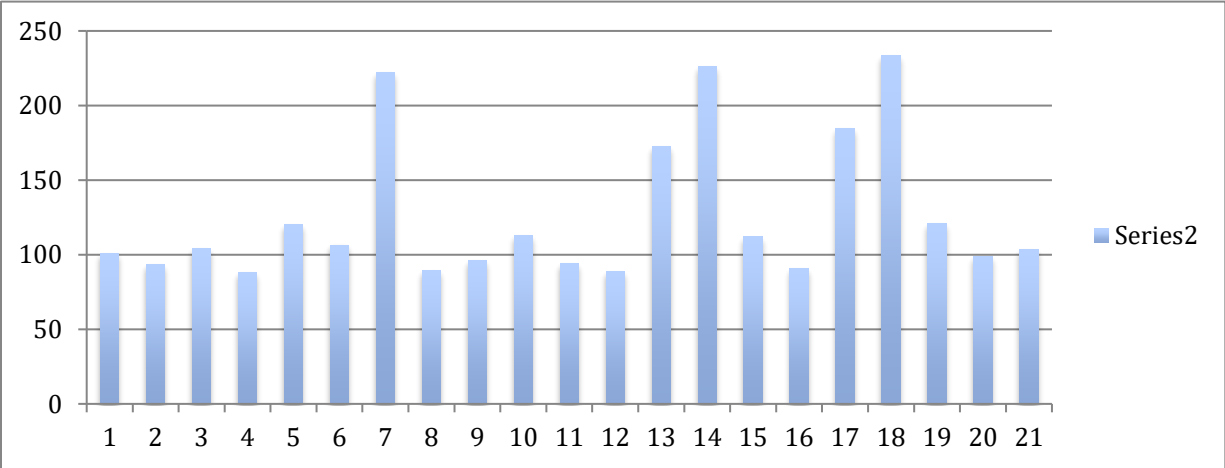


Figure 15 Average height measurements the 26th of June

As we can check in the graph of the average height, the object 7, 13, 14, 17 and 18. EFP 33 x serenade, EFP 33 x polyversum, ARKA x serenade, EFP 33 x control and ARKA x control are the varieties corresponding to these numbers respectively. The varieties ARKA and EFP 33 are the highest at the time of the maturity but this quality can't assure a good production, we had to weight the pods of each object to know which was the most productive.

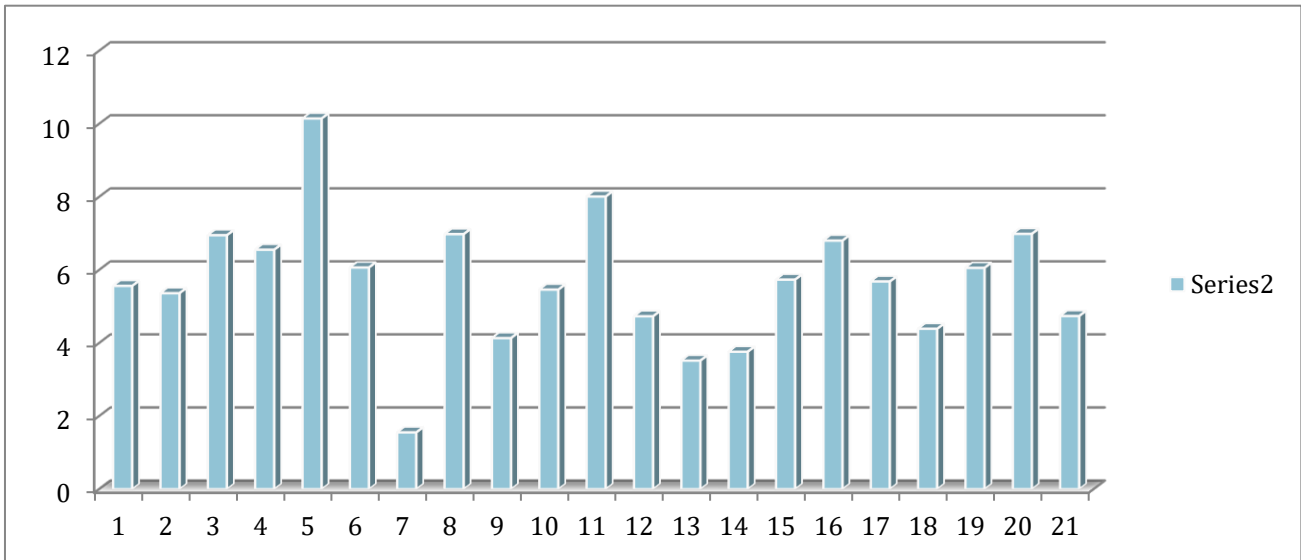


Figure 16 Average yield production measured the 26th of June

In this graph we have the yield production in tn/ha.

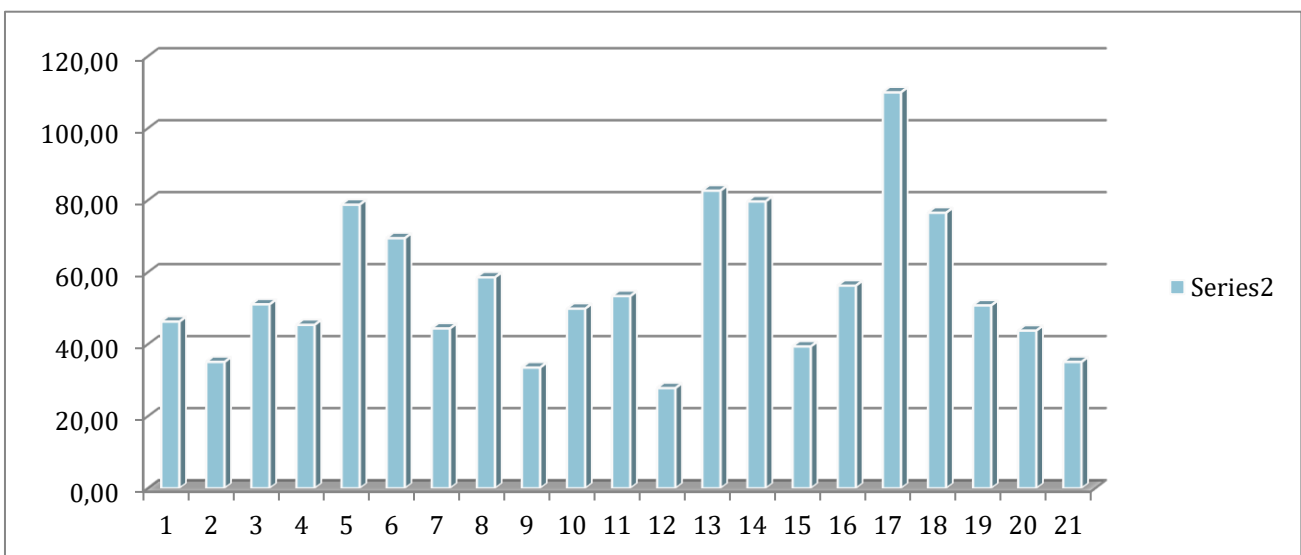


Figure 17 Average number of seeds measured the 26th of June

In this other graph we have the number of seeds per plant.

After analyzing these graphs we can say that the most productive varieties are Aviron and Baltrap, Specter has also a high production but not as the ones mentioned before. It is also important to note that the varieties with a higher number of seeds not always have higher yields.

As a conclusion, we can say that the varieties with a faster development are the EFP 33 and Arka. We can also affirm that Aviron and Baltrap were the most productive varieties. It depends of the use the farmer is going to give to his crop, he must use one or another variety, the EFP 33 must be a good variety for fodder probably, due to its fast development. Aviron and Baltrap are perfect for human fooding. This means that the varieties that are more adapted to our geographical location are Aviron and Baltrap, these ones will be the chosen ones to be selected for large-scale cultivation.

Discussion

The main objective of this study was to find those varieties best adapted to organic farming in large areas. The secondary objective was to test two biofungicides that had never been used in pea cultivation before.

After analyzing all the data collected throughout the variety test that we have evaluated, we can confirm that a number of commercial varieties have shown a much greater adaptation to the geographical area of the study. This indicates that these could be studied in the future to make a genetic improvement in some specific aspects such as the size of the grain, the height of the bushes or the amount of nitrogen supplied by the plants to the soil. Thanks to this study, it has been possible to contribute to rescue some varieties that were believed to be poorly adapted to the climate, soil and growing conditions of the study area. Some of the varieties used in the study have shown to have a good productive and quality aptitude. It should also be noted that these have a simple handling when implanting an organic crop, since among other factors the effectiveness of one of the biological products used in the study has been demonstrated. Among the varieties studied, Aviron and Baltrap varieties stand out as the most productive, although as mentioned previously, they have not been the varieties in which a more positive effect of the biofungicides tested has been seen. The Aviron variety has experienced greater growth in those plants to which the Serenade product was applied. Regarding the agricultural management of these varieties, we have found that they are easier to use since their development in length is not excessive.

The varieties that have been stood out for having a faster growth, in greater length and mass have been Arka and EFB33. Despite this, the productivity of these varieties is not significant in terms of grain production. Therefore, they could be interesting varieties for animal feed because of their high vegetative growth.

Regarding the chlorophyll measurements made in SPAD units, we can affirm that a direct correlation between these and the productivity of the plants has not been found. Therefore, these measurements have been considered ineffective in predicting the productivity of the pea crop before the time of collection. However, a correlation has been found between the leaf area of the varieties and their production. Although the most productive varieties have not been those with a more significant leaf area, we can observe a close relationship between these two variables. A larger foliar area, pea varieties are more productive.

Among all the varieties it has been possible to identify a variety that stands out for its productivity / quality / simplicity of handling relationship, the Aviron variety. Mainly due to the low pest involvement and to have a much better cycle adapted to the climatic conditions of the growing area.

Regarding the biofungicides used in the test, the Serenade product has obtained better results than Polyversum. It is important to note that the differences have not been significant and we would recommend continuing the study to find a more suitable product to improve crop productivity.

As a final conclusion, we must point out that the testing of organic varieties and products should be a permanent line of work to extend and facilitate the cultivation of peas and legumes in general in this area.

Discusión

El objetivo principal de este estudio era encontrar aquellas variedades mejor adaptadas al cultivo ecológico en grandes extensiones. El objetivo secundario era poner a prueba dos biofungicidas que nunca se habían utilizado en el cultivo de guisante anteriormente.

Tras analizar todos los datos recopilados a lo largo del ensayo de variedades que hemos evaluado podemos confirmar que una serie de variedades comerciales han mostrado una adaptación mucho mayor a la zona geográfica del estudio. Esto nos indica que estas podrían ser objeto de estudio en el futuro para realizar una mejora genética en algunos aspectos concretos

como el tamaño del grano, la altura de las matas o la cantidad de nitrógeno suministrado por las plantas al suelo. Gracias a este estudio se ha podido contribuir a rescatar algunas variedades que se creían poco adaptadas al clima, suelo y condiciones de cultivo de la zona del estudio. Algunas de las variedades utilizadas en el estudio han demostrado tener una buena aptitud productiva y de calidad. También hay que remarcar que estas tienen un manejo sencillo a la hora de implantar un cultivo ecológico, ya que entre otros factores se ha demostrado la eficacia de uno de los productos biológicos utilizados en el estudio. Entre las variedades estudiadas destacan las variedades Aviron y Baltrap como las más productivas, aunque como se ha mencionado con anterioridad, no han sido las variedades en las que se ha visto un efecto más positivo de los biofungicidas puestos a prueba. La variedad Aviron sí que ha experimentado un crecimiento mayor en aquellas plantas a las que se aplicó el producto Serenade. En cuanto al manejo agrícola de estas variedades hemos comprobado que son más sencillas de entutorar ya que su desarrollo en longitud no es excesivo.

Las variedades que han destacado por tener un crecimiento más rápido, en mayor longitud y masa han sido Arka y EFB33. A pesar de ello, la productividad de estas variedades no es significativa en cuanto a la producción de grano. Por tanto, podrían ser variedades interesantes para la alimentación animal por su elevado crecimiento vegetativo.

Respecto a las mediciones de clorofila realizadas en unidades SPAD, podemos afirmar que no se ha encontrado una correlación directa entre estas y la productividad de las plantas. Por lo tanto, estas mediciones se han considerado poco eficaces para preveer la productividad del cultivo de guisante antes del momento de la recolección. No obstante, sí se ha encontrado una relación entre el área foliar de las variedades con su producción. A pesar de que las variedades más productivas no han sido aquellas con un área foliar más significativa, podemos observar una estrecha relación entre estas dos variables. A mayor área foliar, las variedades de guisante son más productivas.

Entre todas las variedades se ha podido identificar una variedad que destaca por su relación productividad/calidad/sencillez de manejo, la variedad Aviron. Principalmente por la escasa afección por plagas y por tener un ciclo mucho mejor adaptado a las condiciones climáticas de la zona de cultivo.

En lo referente a los biofungicidas utilizados en el ensayo, el producto Serenade ha obtenido mejores resultados que el Polyversum. Es importante resaltar que las diferencias no han sido significativas y recomendaríamos continuar con el estudio para encontrar un producto más adecuado para mejorar la productividad de los cultivos.

Como conclusión final, debemos apuntar que el ensayo de variedades y productos ecológicos deben ser una línea permanente de trabajo para extender y facilitar el cultivo del guisante y de las leguminosas en general en esta zona.

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