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THE USE OF AEROBIOLOGICAL DATA ON AGRONOMICAL STUDIES

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Abstract: Pollination is only one of the many events comprising the plant development cycle; however, it is extremely important for yield where seed is required. Although successful fertilization depends on a number of environmental and endogenous factors, including climate and plant nutritional status, a sufficient quantity of pollen must reach the receptive stigma in order to enhance fertilization potential. Aerobiological research focuses on the airborne dispersal of biological particles, including pollen grains from anemophilous plants. Airborne pollen data are currently used for various purposes in agricultural research. One major use is as a source of advance information concerning variations in the final fruit harvest of wind-pollinated species. This application, first introduced in the field of plant pathology in the 1940s, was further developed in the 1970s in French studies of vineyard yield; more recently, it has been successfully tested both in crops and in non-crop forest species such as oak or birch. Nowadays, aerobiological research into the influence of pollen emission on final fruit production takes into account a number of other variables, including weather-related factors and phytopathological data; it also uses new, computerized statistical tools to obtain more precise information on agricultural yield and phytopathological risks.

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INTRODUCTION

A plant constitutes a complex biological system in which some functional units (buds) undergo an annual, genetically-determined development cycle. The cycle comprises both vegetative and reproductive development; both forms involve a number of phenological phases characterized by specific morphological and/or physiological changes. Traditionally, the study of plant phenology has relied almost solely on recording the timing of morphological changes; however, more recent research has shown that a deeper analysis of certain key phases (e.g. flowering or fruit ripening) provides a reliable biological evaluation that can usefully be applied to various crops. The phenological phases involved in flowering provide a macroscopic index of a key endogenous process influenced by external factors including soil, climate, and crop husbandry.

Aerobiology is a multidisciplinary science studying the release, dispersal and deposition of airborne living organisms; it deals with many different types of particles generated by natural or human activities, capable of producing biological effects [20]. Aerobiological analysis enables the detection of airborne pollen and spores, thus providing information on plant phenology, potential crop production, plant distribution and the health of some species, allowing certain phytopathological risks to be identified.

Airborne spore detection enables fungal diseases to be predicted and prevented; it provides valuable data which can be used to model the emission and deposition of phytopathogenic spores within crops, and to predict their transport from one crop to another [28]. The objective recording of pathogen spore levels provides the basis for Integrated Pest Management (IPM), a crop-husbandry strategy designed to overcome ecological problems (Fig. 1). IPM

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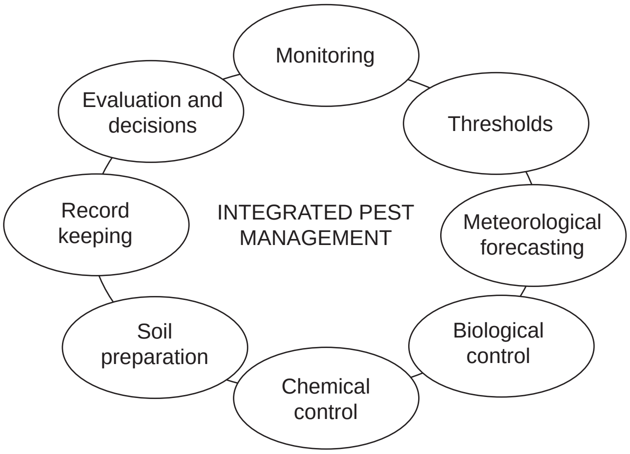

Figure 1. Integrated Pest Management Scheme.

is a sustainable method of managing pests by combining biological, cultural, physical and chemical tools in a way that minimizes economic, health and environmental risks. These new methods must be implemented in three stages: prevention, observation, and intervention. The main goal is to eliminate or significantly reduce pesticide use while at the same time maintaining pest populations at acceptable levels. Recent studies using Aerobiology Modeling System (AMS) simulations in conjunction with meteorological information have provided the basis for communications and alerts from plant pathologists to farmers.

Another major application of aerobiological data in agricultural research is the forecasting of crop production on the basis of airborne pollen data. Pollination is only one of the many events taking place in the plant development cycle; however, it is extremely important for yield where seed is required. Although successful fertilization depends on a number of environmental and endogenous factors, including climate and plant nutritional status, a sufficient quantity of pollen must reach the receptive stigma in order to enhance fertilization potential [28]. Moreover, in anemophilous plants a larger number of pollen grains are required to ensure pollination. Even at a distance of hundreds of kilometers, pollen incidence may be sufficient to effect at least some fertilization [22]. Long-distance pollen dispersal is of great importance for pollination and seed-setting in isolated specimens, and also for the long-distance transport of genes [22]. Wind pollination involves an indiscriminate, inefficient dispersal mechanism, and requires very large amounts of pollen in order to ensure proper pollination in many crops [28]. If a stigmatic surface measures 1 mm^2, then 1 million pollen grains distributed evenly over an area of 1 m^2 are required for reasonable success in fertilizing a single ovule. The efficiency of wind pollination may be expressed by the equation: $n = N \times a/A$, where n = effective pollen, N = total output of pollen produced, a = stigmatic surface, A = total area of the surroundings [25].

Pollen production, which is genetically and physiologically controlled, largely determines the pollination process [5, 37, 40, 42, 59, 69]. Therefore, since wind pollination is a less controlled process than insect pollination, anemophilous plants have a very high ovule/pollen grain ratio averaging 1/500,000 [69]. The resulting elevated airborne pollen counts provide the basis for aerobiological crop-forecasting methods. Cour & Van Campo in 1980 were the first to demonstrate the link between pollination levels in anemophilous species and subsequent yields [18]; since then, a number of authors have used airborne pollen data as a tool for forecasting grape, olive and cereal crops [7, 31, 53]. Optimized production and reliable crop forecasting are essential for efficient product marketing: armed with an advance estimate of potential yields, producers can adopt the necessary strategies to offset year-on-year variations, and can also make informed decisions on harvest planning, pricing, insurance, and stock management [39]. This is especially necessary in the context of common international agricultural policies such as that operated in the European Union, whose farmers must meet production quotas in order to be eligible for subsidies.

Over recent years, this application has been tested in non-crop forest species in order to account for variations in fruit production. Although this research is hindered by the absence of fruit production data of the sort available for agricultural crops, tentative results suggest that the considerable year-on-year annual variation in fruit production by anemophilous forest species (especially trees) is due largely to differences in pollen production and dispersal [5, 13, 35, 50].

PHENOLOGY

Phenology, a term derived from the Greek *phaino* meaning “to show” or “to appear”, is the study of periodical biological events in the animal and plant kingdoms as influenced by the environment [67]. As soon as the first farmers began to settle, plant seeds, observe crop growth and obtain annual harvests, they became aware of the link between plant development and changes in the environment. The earliest phenological research naturally focussed on agricultural crops, in view of the economic importance of weather-induced effects [6, 57, 58, 60, 68]. Airborne pollen monitoring provides an objective record of the various flowering phenophases in wind-pollinated plants. Phenological analysis enables the complex correlation between climate and floral productivity to be accurately charted in these species; plants are excellent indicators of climate change, since the onset of phenological events is closely governed by weather-related factors. As a result, plant phenology models are increasingly used for a wide range of purposes: predicting the impact of global warming on crops [31], improving primary productivity models [47, 49], forecasting airborne pollen counts [14, 33], and supporting foresters and farmers in management decisions such as the selection of reforestation sources in order to prevent frost damage [11, 36].

In general, phenological models are better termed 'pheno-meteorological' models, in that they use weather-related parameters to predict phenological events. In floral phenology, air temperature is the variable most influencing the flowering process [15]. Most of the variability in pollination onset is accounted for by heat accumulation over the preceding weeks, expressed as 'Growing-Degree-Days' (GDD^o), especially in tree species. GDD^o models must be defined by the start date for heat accumulation and by the threshold temperature above which the plant responds. These parameters may vary depending on the species and the study area. Other major variables in phenological studies include photoperiod and water availability, especially in herbaceous species [33, 34].

Plant-phenology forecasting is becoming increasingly important in agriculture, since many crop practices – including the application of chemical, biological and hormonal treatments – must be carried out during specific phenological phases. Moreover, the combined monitoring of plant phenology and airborne pathogenic spore counts has been found to enhance the success of IPM strategies. Fungal spore germination occurs only under certain conditions and during specific phenological phases [3]. Planning of chemical and biological treatments can thus be improved by taking into account not only spore thresholds but also favourable phenological phases.

Aerobiological monitoring also has ecological applications. Analysis of airborne pollen data can provide an indication of species distribution, and can be used to monitor weed invasion. Aerobiological data thus serve as bioindicators of environmental change: in some areas of Central Europe, for example, the invasion of *Ambrosia artemisiifolia* L. has been observed as a weed in summer crops. *A. artemisiifolia*, *Artemisia* spp. and other ruderal species are highly resistant to pollutants, and are seen as a sign of environmental decline; increased airborne pollen counts for these species, coupled with a decrease in tree-pollen counts, are thus indicative of the bio-deterioration of vegetation.

AGRICULTURAL PRODUCTIVITY

Pollination is a key factor for crop yield. Although, theoretically, one pollen grain per ovule would be sufficient for fertilization, in several wind-pollinated plants the average number of pollen grains reaching the stigma ranges from 5 to 20 [66].

Seasonal pollen yields vary considerably and, though pollen output per plant, also varies widely between species, most wind-pollinated species release relatively large amounts of pollen [69]. Pollen emission is the result of a long period of development, usually starting in late summer the previous year. The amount of pollen available for the following year is predetermined, since the cells designated to become pollen grains are already present. In anemophilous tree species flowering in early spring, such as *Corylus* and *Betula*, meiosis is observed in August or early September [26, 27]. Therefore, for winter-dormant trees the pollen yield depends on temperature and rainfall during the previous months. The stored resources of any plant are strained when both pollen and seeds are produced in large quantities. In many trees, variations in fruit production are due to the alternation of high-pollen-emission and low-pollen-emission years.

Aerobiological data provide information not only regarding the timing and the trend of the phenophase, but also regarding its magnitude. Airborne pollen counts are an indicator of the amount of pollen actually produced by the plant. Numerous studies have reported a close link between the quantity and quality of emitted pollen and fruit production in wind-pollinated plants [10, 28]. Pollen data can provide information regarding the final fruit harvest several months in advance. This application, first developed in the 1970s in France by Cour [17], has been successfully tested in both anemophilous crops and non-crop forest species [29, 30, 35]. Knowledge of the major biological and climate factors influencing the final harvest is becoming increasingly necessary in order to obtain reliable crop estimates and thus ensure optimized, effective crop management. This knowledge is also of great value to public agricultural institutions, for the planning of government subsidies [64]. Early and effective crop forecasting is proving essential in optimizing human and economic resources for harvesting, marketing strategies, and global commercial distribution. This is of particular importance for crops such as olive or grapes in Europe, which are major targets of European Union (EU) agricultural policy [1]. EU regulations establish production quotas, assign economic aid in cases of harvest loss due to weather-related disasters, encourage the planting or abandoning of certain crops, and establish channels of communication among producing countries to prevent market shortages and uncontrolled price rises in low-production years. Until now, the most widely-used forecasting methods have been based on plot censuses, in which the observation of a limited number of plots provided an agronomic inventory from which the total production of a region could be extrapolated [51, 62]. However, this forecasting method has certain drawbacks [7, 55]:

a) Plot yield estimates are often affected by observer subjectivity.

b) The method is costly because it requires numerous observation points.

c) The earliest estimates often show an excessive margin of error, which can be corrected only in the period close to harvesting.

As a result of these drawbacks, since the 1960s a number of authors have advocated forecasting methods based on the correlation between airborne pollen counts and fruit production in both cultivated and forest species [44, 63].

The widely-used method developed by Cour and Van Campo [18] has subsequently been applied to a range of crops, including olives, vines, cereals, citrus fruits and hazelnuts [2, 19, 39, 52, 56].

The olive tree originated thousands of years ago in the eastern Mediterranean, and later spread westwards. The adult plant is estimated to have a million flowers that are either unisexual or hermaphrodite and are arranged in bunches [2]. It is an amphiphilous species: primarily insect-pollinated, but with secondary wind-pollination. The fruit is a drupe from which olive oil is obtained. A large amount of farmland is devoted to olive production in the Mediterranean area [8]. Because of these floral, palynological and cultural characteristics, high airborne olive-pollen counts are recorded in many European Mediterranean regions. In southern Spain, Gónzalez-Minero *et al.* [39] monitored olive pollen counts using a Cour trap; analyzing their data in conjunction with agricultural yields and meteorological observations, they developed a forecasting method based on simple and multiple regression. They devised three sets of forecasting equations: for early July (the end of flowering, and six months before fruit picking); for late November (immediately before picking); and for late January (once fruit picking was over).

Airborne pollen data have been used to determine optimum harvest dates in vineyards in France, Spain and Portugal [16, 38, 46, 61]: these studies generally noted a trend towards earlier harvest dates. A correlation has also been detected between pollen counts and grape production, although the monitoring of fungal spores is essential in order to evaluate the impact of phytopathological diseases. Regression equations therefore take into account the effect of post-flowering growing conditions, and a minimum of 3–4 years are required to build reliable models. Analysis of results obtained in France indicates a strong correlation between estimated and real vine crops, with a mean R coefficient of 0.90 [7].

This method has proved effective in other anemophilous woody crops such as the hazelnut (*Corylus avellana* L.), for which Riera-Mora [62] developed a forecasting equation capable of predicting fruit production up to 7-8 months prior to harvest.

Over recent years, Hirst volumetric pollen traps [43] have proved to be an accurate tool for crop forecasting, especially for olives – to which most research has been devoted [29, 30, 32, 54]. Most equations combine olive-tree phenology, airborne pollen counts, weather data and fruit production data to yield accurate results.

Using a Hirst trap, Muñoz *et al.* [53] evaluated the correlation between Poaceae pollen counts and cereal yields in Central Spain. The chief findings were a strong correlation between June pollen counts and dry-land cereal yields (wheat, barley and triticale), and a lack of correlation between pollen counts and irrigated-crop yields (maize, rice and sorghum). A significant correlation was recorded between mean overall pollen counts in May and June and mean cereal yields, although this is likely to reflect the similar effect of environmental conditions on the wild flora producing most of the airborne pollen, and on cereal crops.

Finally, attempts have been made to forecast fruit production in non-crop tree species, and especially in woody species such as *Quercus* [13, 35], *Taxus* [5], and *Betula* [50], all of which are characterized by highly-variable fruit production. Various hypotheses have been put forward to account for the alternation between high and low production, although the variables involved remain unknown. In evergreen species such as Quercus, the resource-matching and seed-dispersal hypotheses have been scientifically ruled out by Koening *et al.* [48]. Other studies generally support the 'predator satiation' and 'wind pollination' hypotheses [21, 48]; the results obtained applying the pollen-count method support the 'wind pollination' hypothesis. Combined use of aerobiological, field phenological and meteorological data could represent a major step forward in forest fruit production research.

The pollen-count method, apart from its ability to provide advance estimates, has other advantages: deviations are lower than in the test-plot forecasting system; fewer collecting data points are needed; and it is more objective than other methods. However, the pollen-based forecasting method has certain limitations, due mainly to the lack of research programmes and the difficulty in calculating pollen-transport distances. Lack of knowledge of post-flowering factors is an additional major problem in Mediterranean areas. Improved definition of climate-related equations will help to overcome this difficulty and realize the full potential of this method. A further disadvantage is the need to establish the average distance over which pollen grains are transported in order to evaluate the fertilization potential in many plants.

AEROBIOLOGY AND PLANT PATHOLOGY

Aerobiological data enable the distribution, ecology and concentration of fungal spores to be determined. Spores dispersed in the air can travel long distances. Airborne spore monitoring provides information on daily and hourly spore counts in a given crop. In 1946, Stakman and Christensen [65] were the first researchers to apply aerobiological methods to plant pathology. A number of authors have since sought to correlate the extent of disease at a given time with airborne spore counts at the same time or previously [45]. Airborne spore counts are a bioindicator of the phenological cycle of pathogens. In the case of grapevine leaf attack by botrytis blight, a significant correlation was found between airborne conidia counts and lesions appearing one week later [12]. In these cases, aerobiological data are more useful than weather data for detecting infections at an initial stage (inoculums), although the combined use of weather and spore-count data provides a valuable tool for the development of accurate, modern Integrated Pest Management (IPM) strategies. When the farmer knows the spore risk thresholds, spore counts can serve as a disease alert if weather conditions are favourable [9]. The weather conditions favouring spore germination are usually humidity and dew temperature. The strategy most widely adopted by winegrowers to reduce the impact of fungal disease is the systematic application of chemical fungicides, generally following preset calendars based on the phenological growth stages of the grapevine [9]. However, integrated control methods are associated with reduced application of chemical treatments, and with lower economic and ecological costs, e.g. 50–80% saving of chemical sprays in the fight against *Phytophora infestans* [9]. Reduction of chemical residues also leads to an improvement in wine quality; the value of wines produced under IPM conditions is thus greater [3, 4].

Recently, several authors have combined aerobiological, phenological and meteorological data to produce equations for forecasting spore concentrations; in some cases, these equations account for up to 40% of spore-count variability when the variables with the highest correlation coefficients are included as estimators [23].

Over the last few years, certain dry areas of the Mediterranean area traditionally devoted to rain-fed farming have been switched to irrigation. This may prompt an increase in the incidence of pathogenic fungi, which are more easily dispersed by irrigation than by rain-splash; since humid environments increase the active discharge of spores, heavy rain and irrigation favour the presence of certain airborne spore types [24, 41].

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