Chapter 7

Phytosociological Surveys in Weed Science: Old Concept, New Approach

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Abstract

Phytosociological surveys have been applied to studies on agroecosystems, especially in relation to weed populations into arable fields. These surveys can indicate trends of variation of the importance of plant populations within a crop, and whether the variations are associated to agricultural practices adopted, which can be further used to support the development of weed management programs. However, to understand the applicability of phytosociological studies for weeds, it is necessary to understand the ecological basis and determine the most appropriate methods to be used when surveying arable fields. Therefore, the aim of the present chapter is to introduce a new approach of phytosociological survey to be used as a tool for the weed science. Throughout the chapter, this new approach is presented in details covering aspects related to methods for sampling and describing weed communities. The following sequence of steps is proposed as the most suitable for a weed phytosociological and association survey: (1) overall infestation; (2) phytosociological tables/graphs; (3) intra-characterization by diversity; (4) inter-characterization and grouping by multivariate analysis; and (5) weeds association through contingency tables.

Keywords: weed species, survey, data processing, diversity, sustainability, agriculture

1. Introduction

The classification of plant species is necessary to understand the complexity of environments, being based mainly on morphology and recently aided by genetics and its functional properties [1]. Plant communities are a set of plant species within a given geographic unit, which form relatively uniform patches, distinguishable from patches of different types of vegetation adjacent to that limited area [2]. Vegetation ecology seeks to identify the species found within the same habitat, thus, describing the physiognomy of the landscape, in order to determine why the communities have a given structure as well as their mechanisms of adaptation [3]. When it comes to community studies, it is necessary to understand how environmental conditions and species interactions influence the patterns of coexistence and relative abundance of species on the local scale, but it should be taken into account the important role of spatio-temporal dynamics [4–6].

The classification of plant communities into a hierarchical system is made in an inductively synthetic way; the types of vegetation units are abstracted as basic syntaxonomic units and compiled as associations [2, 7]. The basis of the phytosociological categorization of plants, according to Blasi and Frondoni [8], is drawn into a context of botanical geography, with the primordial observation that plant species are grouped in associations in which they differ in composition and/or physiognomy, according to geographic regions and environmental conditions (e.g., climate, altitude, latitude).

In this sense, phytosociology is the science that seeks to understand, through the composition and distribution of plant species in a given phytogeographic region, the diversity of plant communities [9]. However, this is a phytogeographic and non-phytosociological approach [10], since the phytosociological approach in the original Braun-Blanquet concept is a floristic statistic based on the occurrence of plant species. In the concept proposed by Oosting [11] and Harper [12], phytosociology is the science of plant communities as well as the relationships with the environment and the processes that modify these communities. This approach is more related to functional ecology, which seeks to understand how and why ecological systems, and their components interact differently in different environments [13].

The term phytosociology, however, is directly associated with the structure of a community of plant species [14], while phytocenosis is defined as the study of plant cover [11]. In this sense, when it comes to a phytosociological survey through an inductive statistical process of inventory comparison, it is necessary to establish a conceptual class that represents a model of phytocenosis. Another concept refers to the possibility of representing patterns in floristic structure and combination, thus modeling phytocenosis as a resource for the dynamic systems theory [15, 16].

A phytosociological study considers three principles: the **analytical** (portrays the size of the inventory surface, characteristics of the sampling site, and variables such as abundance, density, dominance, and the sociability of plant species), the **synthetic** (referring to frequency of species which compose the plant community), and **syntaxonomy** (establishing the phytosociological hierarchy) [9, 11, 15, 16]. Thus, for a phytosociological study, it is necessary to take into account that vegetation varies in spatial scales, which are defined by the size of the sampled units and that several patterns can be detected only with the change in the observation scale, so the size of the sampling unit influences the analysis of spatial patterns of plant populations [17]. In this way, evaluating the quality and quantity in the composition of species of phytocenosis, as an expression of all historical, sociological, and local influences of the abiotic factors, can be a key point for understanding the floristic composition [2].

Basically, the procedures and methods for sampling and registering plant communities follow the Braun-Blanquet [9] method as it considers the analysis and description of selected plant populations as basic types [18]. This method, however, has some limitations and alternatives have been developed along the years [14].

Phytosociological surveys have been applied to studies on agroecosystems, especially in relation to weed plant populations into crops [19]. An infesting population is a result of the interactional relationship between phenotypic plasticity of each individual and long-term processes that provided adaptive flexibility to eventual changes in the natural or artificial environment [20, 21]. Thus, when conducting a phytosociological study of weed species in crops, these can indicate trends of variation of the importance of plant populations within a crop, and these variations may be associated to the agricultural practices adopted as well as subsidize the development of weed management programs.

In general, the phytosociological studies of weed plant communities in agroecosystems allow the determination of periods of control and/or coexistence between crop and weeds, and through the phytosociological indices, it is possible to determine which species are the most important in the different periods of growth of the weed community [14]. However, to understand the applicability of phytosociological studies for weed species in crops, it is necessary to understand the ecological basis and determine the most appropriate methods to be used when surveying arable fields.

1.1. Two contrasting theories

The study of vegetation played an important role in the evolution of ecological concepts through the formulation of several vegetation theories as well as methods of surveying and analyzing phytosociological data [10, 22]. In phytosociological terms, the concept of community is based on the principle of associations (different groupings of plant species, usually found in sites with similar environmental conditions) [14].

The definition of associations proposed by some authors [1, 23] considers the type of vegetation that represents the real plant communities and shares a certain combination of statistically reliable characteristics, in terms of physiognomy and stratification, ecological conditions, dynamic meaning, area of distribution, and history. This gives the association a greater value of information in ecological and geographical terms, which increases the indicator value of vegetation in a site [8]. However, there is some discussion in terms of synecological methods related to the concept of community. For instance, the concept of community proposed by Begon [24] refers to a set of species that inhabit the same area at a given time, while Gurevitch [25] refers to a group of populations that coexist in space and time, interacting directly or indirectly.

Two great ecologists, Frederick E. Clements and H.A. Gleason, presented a series of discussions on community ecology. The driving issue was whether the community was a self-organized system of co-occurring species or simply a random collection of populations with minimal functional integration. Two extreme views prevailed: one view considered a community as a “super-organism” whose species were strongly united by interactions that contributed to repetitive patterns of species abundance in space and time; in contrast, communities were a result of interactions among species, as well as between species and environment, combined with historically extreme and occasional climatic events [26].

For Clements, in his theory called "super-organisms", organisms and communities not only have their own growth and development but also evolve from predecessor communities. They supposedly have an ontogeny that one could study, just as is done with individuals and species, so one could classify the communities in a way comparable to the Linnaean taxonomy. It ultimately assumed a common evolutionary history for the integrated species [27], and the emergence and disappearance of a particular plant community was supposed to be easily and accurately estimated because it was considered as a single organism [26]. In short, the concept of plant community in this theory is defined as an autonomous, discrete, individualizable entity possessing its own structural and functional properties [27, 28]. This theory predicted that the optimum and the amplitude of the species presented distinct clusters, so expected changes in the vegetation would be abrupt [14].

On the other side, the theory of Henry A. Gleason focused on the traits of individual species that allow each to be within specific habitats or geographic areas [29]. This is a much more arbitrary unity than that imagined by Clements, since in Gleason's view, the spatial barriers of communities are not clear and assemblies of species can change considerably over time and space allowing each species to have its own tolerance to certain selection factors, thus, responding to environmental stresses in particular ways [26]. Thus, it was proposed a concept of a plant continuum in which species combinations result mostly from individual responses to environmental factors, and the occurrence of dispersion of individuals is random as a response to environmental fluctuations [28]. This theory states that the level of occurrence of a given plant species is proportional to the level of the stress the species can tolerate [25].

The theoretical opposition allowed the origin of two strands of current vegetation studies, the *discrete* versus *continuum* [26]. The same authors point out that the widely accepted view of the nature of communities is much closer to Gleason's view, since a given species may occur in species assemblies or communities, under different circumstances. When analyzing the relative importance that each of these authors gives to the different orders of factors in the vegetation theories, it is concluded that all of them consider the responses of the species to the habitat as the dominant influence in the structuring of the vegetation [28]. Begon [24] found that community ecology can be described as the study of the level of community organization rather than a spatially and/or temporally definable unit. Thus, it can be defined that the gradient of plant composition of the agglomerates is defined by the environment (or management in arable areas), and abrupt changes are observed in the composition of species within clusters when abrupt selection factors are applied [25, 26, 30, 31].

2. Aims and methods

Before reviewing the main methods and criteria involved in the sampling and evaluation of flora and vegetation, it is necessary to define some basic concepts:

1. Flora is the set of plant species present in a given place or area;
2. Vegetation refers to the quantitative aspects of plant architecture, that is, its horizontal and vertical distribution on the surface; and
3. The plant community should be understood as a set of plants of two or more plant species that coexist in a certain area, and according to the dominance of some of its species, it can be differentiated from other natural and/or altered plant communities [32, 33].

Based on Whittaker [34], "*Within phytosociological studies, the species inventory is fundamental to characterize both α diversity (species richness of a particular community considered homogeneous) and β diversity (degree of variability or replacement in the composition of species among different communities of an environment)*". Thus, in initiating a study of the composition, structure, and ordering of a plant community, the fundamentals are the choice of sampling type (random or systematic), location, size, shape, and quantity of sample units [26]. Once these are obtained, the researcher can apply the measurement of the attributes of the vegetation to be characterized (such as abundance, frequency, density, and dominance), biotypes, vertical structure, horizontal structure (coverage), and others [25, 26, 30]. The measurement of these attributes allows obtaining the importance value (IV) and biodiversity indices [26].

The purpose of phytosociological studies in the weed science does not differ much from the ecological field and rather tends to combine efforts between two disciplines (botany and ecology), in order to improve agricultural productivity and decrease the competition between crops and weeds [35]. Many of the traditional studies in weed science carried out in nonindustrialized (usually considered also under developed) countries have focused on adopting foreign technologies, with little research on biological and ecological aspects of weeds, diagnosis of population dynamics, and integrated weed management [36]. In this context, the use of phytosociological methods in the weed science can be directly associated with the nature of the treatments applied to arable fields, its intrinsic factors, and the history of the area where it will be established [14]. In arable fields, however, two implications must be considered:

1. The plots are usually much smaller in size than expected in phytosociological sampling of wild ecosystems and
2. There is a stronger set of factors influencing arable fields, such as plant density and height, history of land use, soil tillage, and application of agrochemicals, compared to natural environments.

Finally, communities of invasive plants show a behavior similar to that proposed by Gleason, in his theory of the individualistic concept of vegetable association, which states that "*Communities are the result of the interaction between individual species and their (biotic and abiotic) environment in combination with historical events*" [29].

2.1. Methods for sampling the community

The choice between the different variants of the methodologies depend on the sampling objective and the characteristics of the populations (richness and distribution) to be evaluated in each particular agroecosystem [9, 26]. Conventionally, the weed sampling methodologies do not always have prior information about these characteristics or some type of support in the decision-making process or sometimes they are not convenient. For example, on the number of points to be taken, conventional weed population sampling methodologies assume that they have homogeneous or random distribution in space [26] and this is not always the case [37] as numerous studies claim that weed distribution is in patches [38, 39]. It is necessary to emphasize that the sampling of weeds has two objectives:

1. Knowledge on the community richness and abundance, which in turn provides information for biodiversity studies (richness and structure of the communities) on the medium and long-term weed-management plans and
2. Mapping and spatial dynamics studies [9].

Some authors [26, 30] point out several sampling methods, but taking into account the limitations imposed by sampling arable fields, only two of them will be dealt with in this chapter: relevé and random quadrats.

2.1.1. Relevé

Braun-Blanquet [9] made an analogy between organisms and communities by comparing a species with a plant community for the purpose of establishing a classification of communities similar to the way organisms are classified into taxonomic groups. For him, the plant community is the basic unit of the taxonomic classification, which serves to establish a hierarchical system of classification of the communities on a world scale. The same author proposed that the selection of the area to be sampled should be carried out through the determination of the *minimum area*, which is defined as the smallest area where the floristic composition of the community is adequately represented. In the case of the relevé method, where the emphasis is given by the list of species, the minimum area is an indicator of the area needed to have a good sample of the community. The minimum area depends primarily on floristic diversity, plant size, and spacing among them in each community. The same is calculated in the field as follows:

1. A small area, say 0.25 m^2, is delineated, and the list of species present on that surface is recorded;
2. By adding the same area to the original one (2× size), the number of species should be recorded again, and the total number of species in the new quadrat should be counted again;
3. This should be done repeatedly (increase the area, count the species) until the number of species tends to stabilization; and
4. The values of the cumulative total of species (on the ordinate) corresponding to each of the successively duplicated areas (on the abscissa) are represented on a pair of perpendicular axes (**Figure 1**).

A strong slope in its initial part normally characterizes the resulting curve because the first areas incorporate a larger number of new species. Subsequently, as the sampled surface is increased, the appearance of new species in the quadrat becomes rarer and, consequently, the slope of the curve decreases tending to stabilization (**Figure 1**). The appropriate size of the sample unit should be found in the horizontal portion of the curve, and the point of inflection of the sample unit (when it is manifest) projected on the axis of the abscissa will indicate the minimum area. In general, it is convenient to use a size that exceeds a little the minimum area.

It is proposed also that each species in the list is accompanied by an estimate of its abundance-dominance by using the combined coverage-abundance scale and also by its degree of sociability (**Table 1**), both stated by Braun-Blanquet [9].

2.1.2. Random quadrats

In certain communities, the determination of frequency estimators (abundance, coverage) depends too much on the criteria of the expert in charge of the evaluation [26, 40], especially in herbaceous formations such as meadows, pastures, or high wetlands, in which it is most useful to use the method known as "random quadrat". This consists of finding subjective patterns within the community to be sampled and to conduct sampling in such a way so as not to favor

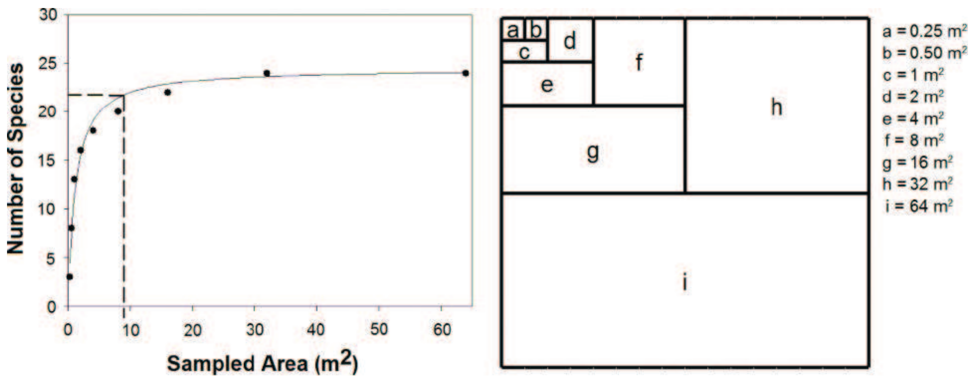

Figure 1. Calibration of the relevé method—determination of the minimal area of the single quadrat to be sampled for fidelity in terms of the number of weed species. Source: adapted from Concenço [14].

Score	Coverage-abundance		Sociability
	Number of individuals	Area coverage	
5	Any number	>75%	Large, almost pure stands
4	Any number	50–74%	Small colonies or carpets
3	Any number	25–49%	Small patches or cushions
2	Any number	5–24%	Small but dense clumps
1	Numerous	<5%	Growing singly
+	Few	Small	–
r	Scattered individuals	Small	–

Source: Adapted from Barbour [26] and Moore [40].

Table 1. Coverage abundance and sociability scales of Braun-Blanquet.

a particular pattern [26, 30]. It means that for the data to be reliable, sampling should be performed as randomly as possible. Several methods are available to help the researcher to go through and sample the area properly, but three of these methods are highlighted to be used in weed science: even spaced, by chance, and random by zones (**Figure 2**).

The geometric forms of the sample unit (called "quadrat") are basically of three types: square, rectangular, or circular [26]. These types of units allow the registration of all variables of dominance, frequency, and density of plant individuals [30]. Based on Goodall [41], surface geometry affects two aspects that significantly influence the vegetation sampling result.

First, let us consider the magnitude of the edge effect given by the area/perimeter ratio of the sample [26]. In the case of plots with rectangular shapes, the long-wide relation of the units and their directional position/orientation influence the degree of heterogeneity registered into the plot. To be considered part of the unit, the plants must be rooted within the perimeter, and the perimeter is longer in a rectangular quadrat compared to square or circular forms. Longer perimeters may increase the chance of the observer to be mistaken when deciding if a plant in the very border of the quadrat is actually in or out of the quadrat [26, 40]; this type of error is called "edge effect" [26].

If rooting occurs outside the plot area but its shoots occupy the airspace of the unit, the plant can be optionally registered as present, depending on the purpose of the survey [25, 32]. It is necessary, however, to make explicit the criterion established when defining the variable, but usually only plants rooted into the quadrat are considered.

2.2. Methods for describing the community

There is a wide variety of methods that allow the floristic characterization of a plant community, whose suitability or applicability depends on the specific objectives of each study and the

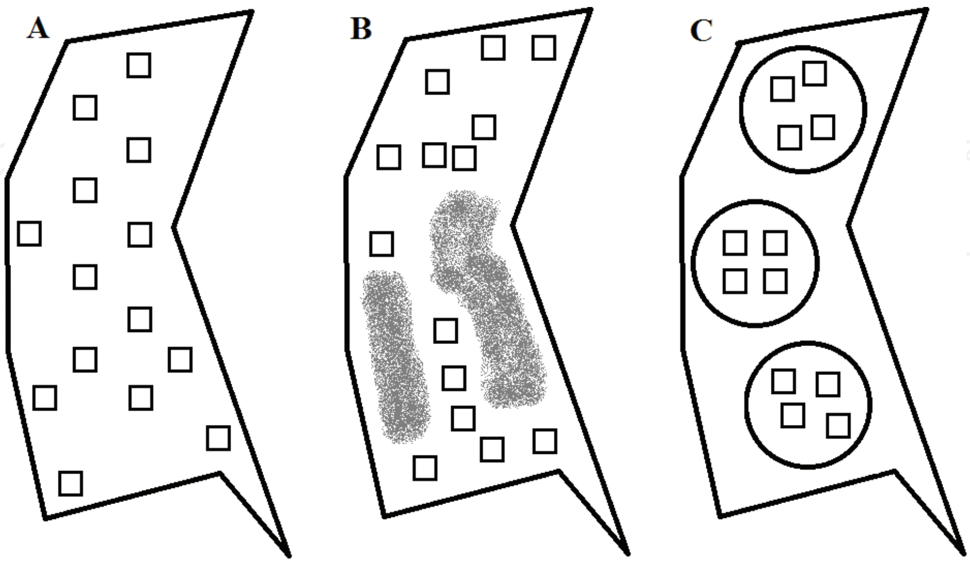

Figure 2. Distribution of samples for the random quadrats method. (A) Even spaced distribution, (B) chance distribution, and (C) random distribution of quadrats by zones. Source: adapted from Concenço [14].

structure of the community studied. However, and regardless of the method used for the floristic study, each sampling unit (quadrat) must meet the following criteria [42]:

1. It must be of sufficient size to contain the most possible proportion of species belonging to the plant community;
2. The habitat must be uniform into the sampling area, within the levels one can determine; and
3. Plant cover should be as homogeneous as possible.

2.2.1. Importance components

A fundamental aspect in the floristic characterization of a plant community is that the methodology adopted should provide an adequate representation of all the species present in the community in natural ecosystems [26]. For arable fields, one may hypothesize that it should properly represent at least most of the weed species present. Once field sampling is accomplished and all data are collected, the following parameters can be calculated [14]:

$$rDe = \frac{I}{TI} \times 100 \tag{1}$$

$$rFr = \frac{Q}{TQ} \times 100 \tag{2}$$

$$rDo = \frac{DM}{TDM} \times 100 \tag{3}$$

$$IV = \frac{rDe + rFr + rDo}{3} \tag{4}$$

where *rDe* = relative density (%); *rFr* = relative frequency (%); *rDo* = relative dominance (%); *IV* = importance value (%); *I* = number of individuals of species *x* in area *r*; *TI* = total number of individuals in area *r*; *Q* = number of samples evaluated in area *r* where species *x* is present; *TQ* = total number of samples in area *r*; *DM* = dry mass of individuals of species *x* in area *r*; and *TDM* = total dry mass of weeds in area *r*.

The *IV* locates each weed species within the community, depending on its ability to cause damage (severity of occurrence) based on the three parameters previously mentioned. In **Figure 3**, the nature of the importance components is illustrated.

2.2.2. Diversity indices

The calculation of diversity indices, α (alpha), β (beta), and γ (gamma), allows the comparative analysis of homogeneous or heterogeneous plant formations. They measure, respectively, the species richness of a community, the degree of change or replacement in species composition among different communities, and their richness in the set of communities [34, 40].

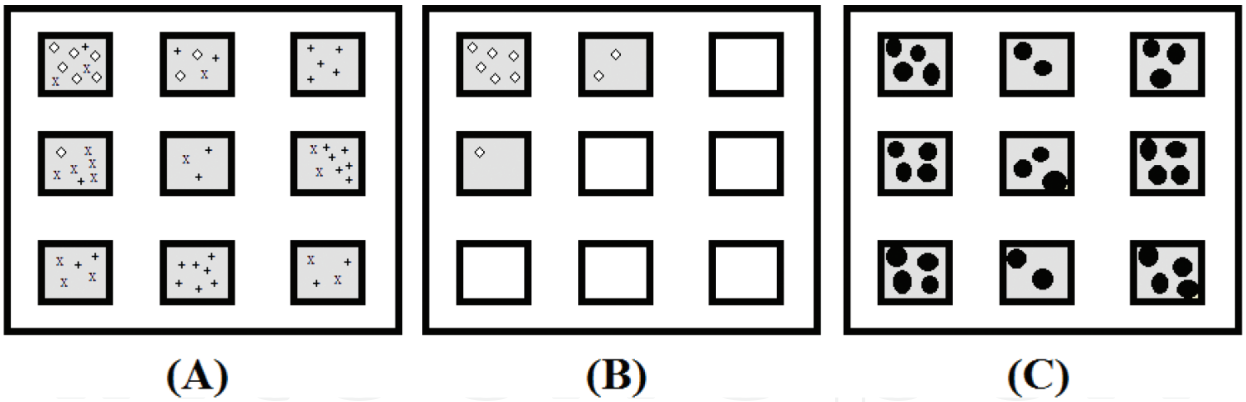

Figure 3. (A) Density, related to the number of plants of a given species found in all quadrats (each symbol is a different weed species); (B) frequency, related to the number of quadrats where a given species was found, independently of number of individuals; and (C) dominance, related to the amount of space in the canopy attributed to a given species, in arable fields, measured usually by dry mass accumulation. Source: adapted from Concenço [14].

The most widely used diversity indices are Margalef (α), Menhinick (*Dm*), Simpson (*D*), and modified Shannon–Weiner (*H*'), besides species density itself [25]. The Margalef index [43] is used to estimate the biodiversity of a community based on the numerical distribution of the individuals of the different species according to the number of individuals in the sample analyzed. It is obtained by Eq. (5):

$$\alpha = \frac{(s-1)}{\ln N} \tag{5}$$

where α = Margalef index, *S* = number of species, and *N* = total number of individuals.

This method can determine the number of taxa and the number of individuals in an ecosystem, comparing species richness among samples collected from different habitats.

The Menhinick index (*Dm*) is based on the relationship between the number of species and the total number of individuals observed, which increases together with sample size [34]. It is obtained by Eq. (6):

$$Dm = \frac{S}{\sqrt{N}} \tag{6}$$

where *Dm* = Menhinick index, *S* = species collected, and *N* = total number of individuals as sum of all species "*S*".

The Simpson index [26] is obtained by Eq. (7). Its calculation is strongly influenced by the importance of the most dominant species. Since its value is inverse to equity, diversity by Simpson is usually calculated by Eq. (8), which indicates that closer to the value of "1", the greater the equity. Simpson's *D* gives very little weight to rare species and is more sensitive to abundant species (those with greater number of individuals). The equations are:

$$\lambda = \sum Pi^2 \tag{7}$$

$$D = 1 - \sum Pi^2 \tag{8}$$

where λ = Simpson index, Pi = proportion of individuals of species "*i*" divided by the total number of individuals in the sample, and D = diversity of Simpson.

The Shannon–Weiner [26] diversity index (H) is another index commonly used to characterize species diversity in a community and is more sensitive to rare species; this is where sampling errors may be greater [26, 30, 40]. It is calculated by Eq. (9):

$$H' = -\sum [Pi \times \ln(Pi)] \tag{9}$$

where H' = Shannon–Weiner diversity index, Pi = proportion of individuals of species "*i*" divided by the total number of individuals in the sample.

It is a relationship between abundance and richness and expresses the uniformity of abundance values across all species in the sample. It ranges from "0", when there is only one species and the Neperian logarithm of "*S*" (number of species collected), when all species are represented by the same number of individuals [40].

In addition to the diversity indices, the Shannon–Weiner evenness proportion (SEP) sustainability coefficient [44], Eq. (11), is able to infer about sustainability of managements applied to production systems from static data. It considers the diversity of Shannon–Weiner calculated both from density (Eq. (9)) and from dry mass data, Eq. (10). As it is a division of one by the other, differences between H' and Hm' near zero, which correspond to SEP near "1", indicate longevity of the management practice applied and consequently of the production system, by the absence of strong species selecting factors. A visual representation of D, H', and SEP is supplied in **Figure 4**.

$$Hm' = -\sum [Mi \times \ln(Mi)] \tag{10}$$

$$SEP = \frac{Hm'}{H'} \tag{11}$$

where Hm' = Shannon–Weiner diversity index based on dry mass and Mi = dry mass of individuals of species "*i*" divided by the total dry mass of individuals in the sample.

2.2.3. Multivariate analysis

Descriptive multivariate analysis provides complementary tools to phytosociology. In this sense, classification and ordering techniques allow the identification of variation patterns in large data sets using algebraic procedures that can be translated into mathematical algorithms. Consequently, these techniques facilitate the work of comparing large sets of data from surveys with the help of computer programs.

Samples of plant communities, whether they are described by the presence or by abundance of the species that compose them, are multivariate because they present values of different variables (species) in each of the studied sites [46, 47]. Based on Matteucci and Colma [48] and Moreno [49], the degree of species turnover (beta diversity) has been evaluated mainly considering proportions

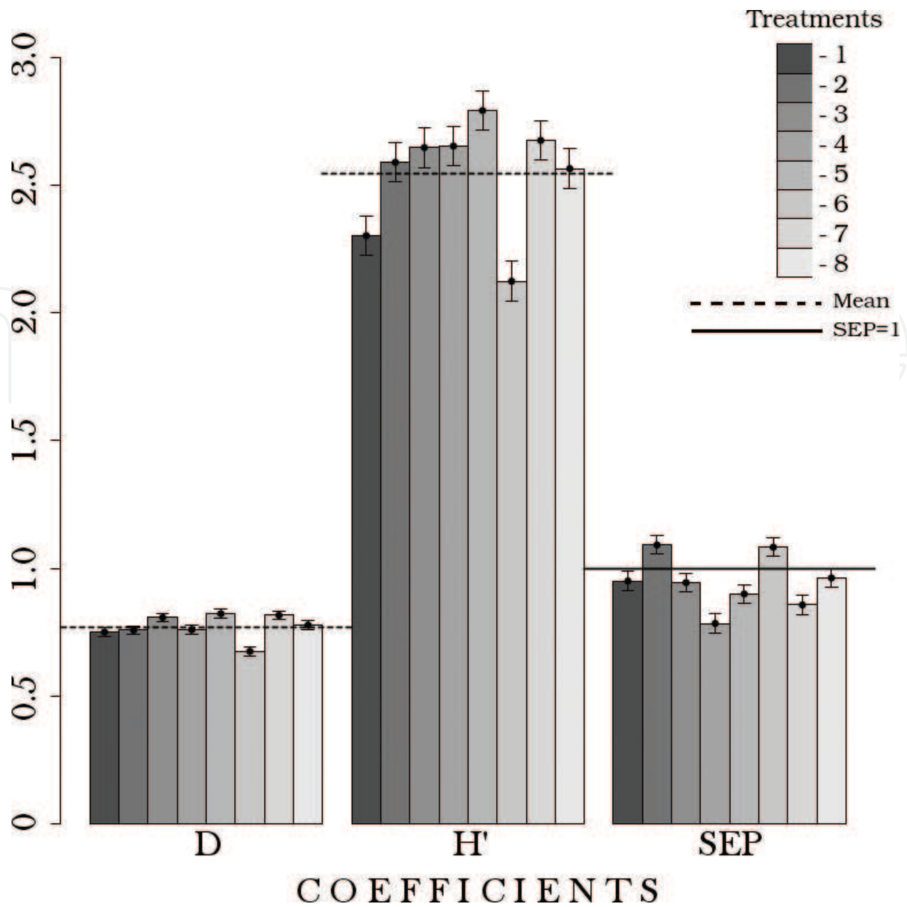

Figure 4. Application of the diversity coefficients of Simpson (*D*) and Shannon–Weiner (*H'*), as well as the sustainability coefficient *SEP*, to eight hypothetic treatments. Error bars are presented. Source: adapted from Concenço [45].

or differences. The proportions can be evaluated using indices as well as coefficients that indicate how similar/dissimilar two communities or samples are. Many of these similarities and differences can also be expressed or visualized by distances. These similarities or differences can be either qualitative (using presence-absence data) or quantitative (using proportional abundance data for each species or study group, as number of individuals, biomass, relative density, coverage, etc.).

The methods for quantifying beta diversity can be divided into two classes: similarity-dissimilarity and exchange/replacement of species. The different indices considered in the methods should be applied depending on the nature of the data (qualitative/quantitative) and what the relationship between the samples is, what it implies, how samples are organized, and how they were obtained, according to the question of interest. Thus, the similarity or dissimilarity expresses the degree of comparability in species composition and its abundances between two samples (communities).

2.2.4. Clustering by similarity

The beta diversity indices of Jaccard (*J*) and Sørensen (*So*) facilitate the comparison of areas in the composition of weed communities [50]. According to Concenço [14], these indices are considered high when they are above 0.25 (25%) and 0.5 (50%), respectively, in which a high resemblance between the areas can be interpreted. Booth [37] indicates that values should be interpreted on an absolute scale from 0 to 1, where 0 indicates total dissimilarity and 1 indicates absolute similarity.

Sørensen's index, Eq. (13), puts more weight on the co-occurrence of species, compared to Jaccard's index (Eq. (12)). Sørensen relates the number of shared species with the arithmetic mean of the species in both compared sites, while Jaccard relates the number of shared species with the total number of exclusive species [26, 40].

$$J = \frac{c}{(a + b - c)} \tag{12}$$

$$So = \frac{2c}{(a + b)} \tag{13}$$

where J = Jaccard's similarity index; So = Sørensen's similarity index; a = total number of weed species in area "a"; b = total number of weed species in area "b"; and c = number of weed species common to areas "a" and "b".

To group the areas according to their similarity, it is advised to first obtain the dissimilarity (differences) between areas (Di), by obtaining the distance from the values of J or So and "1" (Eqs. (14) and (15)):

$$Di_J = 1 - J \tag{14}$$

$$Di_{So} = 1 - So \tag{15}$$

where Di = dissimilarity (by Jaccard or Sørensen); J = Jaccard's similarity index; and So = Sørensen's similarity index.

After obtaining the dissimilarity matrix of treatment versus treatment (**Table 2**), multivariate analysis of hierarchical clustering may be performed by the unweighted pair group method with arithmetic mean (UPGMA) hierarchical clustering method [51] (**Figure 5**). The critical level for separation of groups in the cluster analysis is advised to be based on the arithmetic mean of the original matrix [26], disregarding crossing points between the same areas. Group validation is

Treat.	T1	T2	T3	T4
T1	0	0.64	0.59	0.53
T2	0.64	0	0.58	0.50
T3	0.59	0.58	0	0.46
T4	0.53	0.50	0.46	0

Table 2. Hypothetical dissimilarity matrix based on Jaccard's or Sørensen's similarity coefficients for four hypothetical treatments. Source: adapted from Concenço [14].

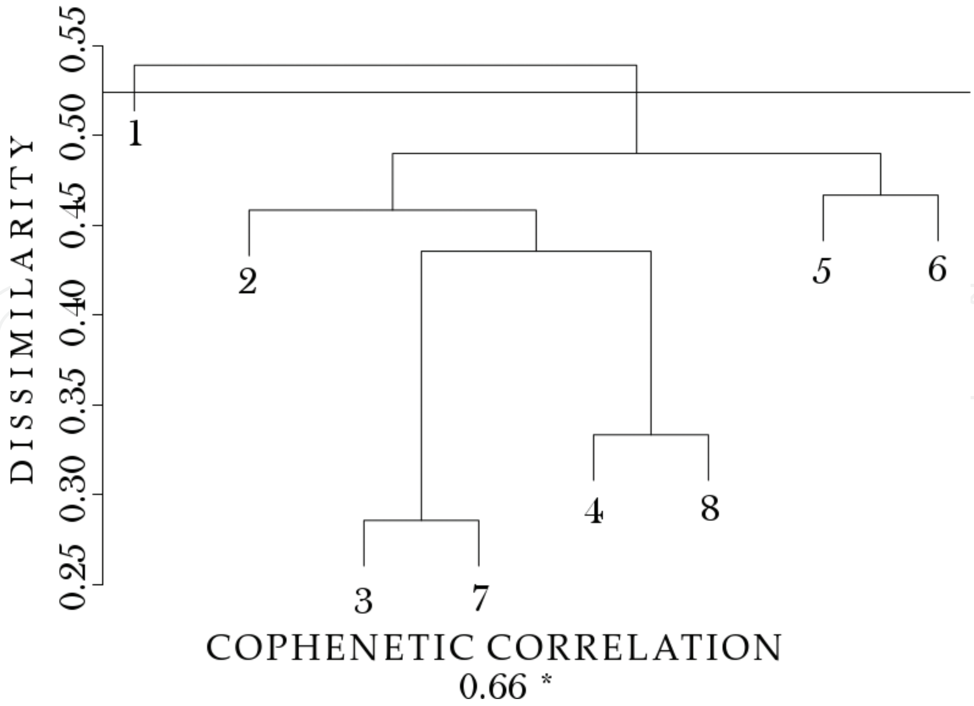

Figure 5. Cluster analysis for eight hypothetical treatments based on the dissimilarity matrix by Jaccard's coefficient and grouping accomplished by the UPGMA method. Source: adapted from Concenço [45].

usually accomplished by the cophenetic correlation coefficient [52], obtained by Pearson's linear correlation between the original matrix of dissimilarity and its respective cophenetic matrix.

3. Association between plant species

The relationships between weed species and the crop in arable fields are described by several authors in terms of competitive aspects [53, 54] and crop-yield losses [55–57]. The balancing in occurrence of weed species in these same arable fields is usually described by means of phytosociological surveys, as previously reported [14, 19, 38], being used as auxiliary to the competitive data aiming to subsidize recommendations for weed control in arable fields. But beyond a simple characterization of both crop losses by competition and the composition of weed occurrence, there is need to understand how weeds interact among them [25].

The principle of interaction among plant species is based on *associations*, which are different clusters of plant species, found generally together, in sites with similar conditions [26]. The first theory regarding plant association was proposed by Clements [27], which stated that plant communities are very organized entities. Thus, the emergence and disappearance of a given plant community could be precisely estimated [26, 27]. In contrast to Clements's ideas, Gleason [25, 29] reported that each species had its own tolerance to given selection factors; thus, they answered to environmental stresses in particular ways. Gleason, however, did not negate at all the occurrence of plant associations, defending that these associations would be more linked to environmental stresses and resource availability than to intrinsic plant traits [29, 40].

Currently, it is believed that plant association exists to a certain degree; the gradient of plant composition of weed clusters is defined by the environment (and by management in arable areas) and abrupt changes in plant composition into clusters are observed when abrupt selection factors are applied [25]. In arable fields with repeated weed management in sequential cropping seasons (same herbicide, soil tillage, crop species), associations among weed species are expected to be valid at a higher degree compared to natural environments, since unfavored species are often eliminated from the area by the weed control techniques. For instance, following repeated application of a single herbicide, those weeds that still remain into the field are most probably those who present, as a common feature, the ability to tolerate that given herbicide, be it tolerant or resistant to that herbicide [58].

In weed science, an overall comprehension regarding plant associations is usually ignored, but its importance lays on two aspects: (1) weed species with positive association among them may answer better to environmental stresses as temperature and water shortage or excess [26, 40, 54], thus, associated plants are most prone to survive, reproduce, and increase its frequency into the community as they work together and (2) the understanding of the association among weed species in arable fields would make it possible to elaborate optimized control plans, be it chemical or cultural, which are efficient over a wider range of weed species at the same time since the technician previously knows they occur together. With the characterization of weed clusters in arable fields, it would be possible to estimate the appearance of weed species into the previously characterized clusters, even before its emergence, by observing the weed species already present and comparing to the usual cluster for that given crop and management.

Thus, understanding the association among weed species in arable fields would ultimately subsidize the development of sustainable techniques for weed control, including optimized herbicide recommendations. The limitation of its application, however, is that clusters would have to be defined for every combination of crop species (soybeans, rice, maize…), cropping system (direct seeding, water seeding, conventional tillage…), and environmental conditions (mainly based on edaphoclimatic characteristics).

Several methods are available in the literature related to plant ecology to assess plant associations in natural environments [25, 26, 40], where low levels of stress and disturbance are usually present [40, 54], and the climax of the vegetation may be most wide and dynamic. Climax is roughly defined as the final and relatively permanent condition of species occurrence in a given environment, as function of climate and soil characteristics [9]. In arable fields, the vegetation climax is heavily biased by crop management; thus, the weed climax tends to be narrower compared to the observed in natural environments, with probable lower degree of uncertainties in weed cluster characterization.

In the present chapter, we aim only to use the ecological approach of plant association as a tool for the weed science, so part of the methodologies available for detailed ecological studies, for instance as presented by Braun-Blanquet [9] and Barbour et al. [26], will not be covered in the present text. The basic steps to achieve a relatively complete characterization of association among weed species, as will be discussed in the following sections, are presented in **Figure 6**.

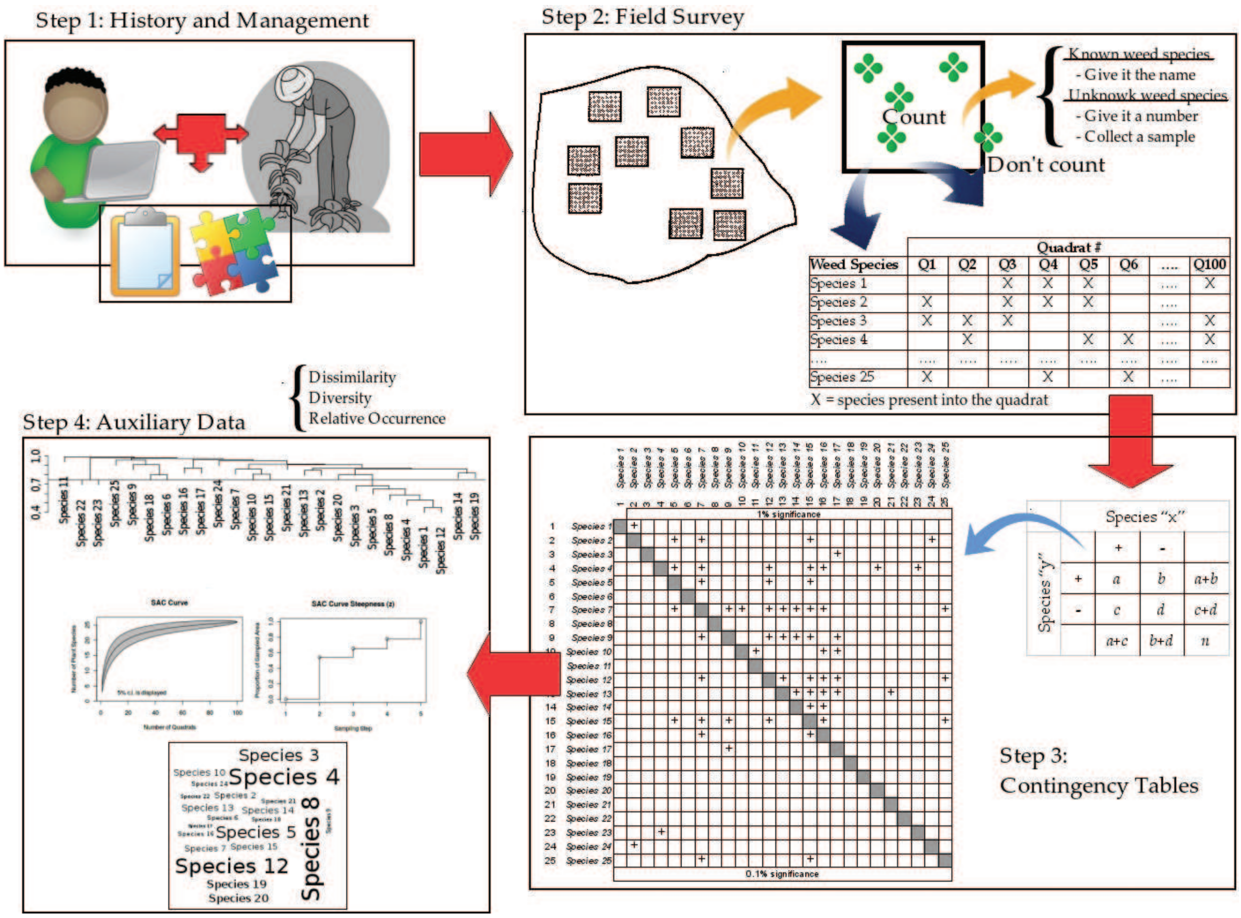

Figure 6. Basic steps to achieve a relatively complete characterization of association among weed species.

3.1. History and management of the area

The field survey for plant association, in arable fields, should be started only after an overview of the area has already been obtained from those who know the field very well. The first step in a study of plant association in arable fields includes obtaining data about the history of the area. Suitable weed management programs include extensive field scout for identifying weed populations and its seeds [59], as well as what growth stage the weeds are in Ref. [54]; talking to the farmer and his field workers will also supply valuable information about the predominance of weed species in the preceding years. Other information that should be obtained is the history about soil tillage, liming, and fertilization, as this information is needed to understand the biological nature of the predominant weed species in the area.

The main point, however, is the history of the herbicides previously applied to the field—those with long residual effect may heavily select weeds which are less susceptible to it [54]; the same may occur with frequent applications of non-residual herbicides [21]. The last 3 years of herbicide application, at least, should be known to the researcher. Perennial and long-term crops, as fruit trees or sugarcane, may mean that longer residual herbicides could have been applied, and in that case, the species associations are valid only under those or similar conditions, as in the absence of the residual effect of herbicides, other weed species would occur in that crop, at that location, and plant clusters would most probably be different.

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3.2. Contingency tables

After the survey about the history and predominant management of the area is concluded, the second step in the determination of plant associations is a field survey by launching random quadrats with fixed size into the area. Methods for optimizing quadrat distribution in the field survey are available in ecology books [25, 26, 30, 37]. In general, sampling 100 quadrats should be enough for a reliable survey [26] in average size fields, although for plant association, both the correct quadrat geometrical form and size are of great importance. The optimal geometrical form for the quadrat is round or square [40], as it will reduce the total perimeter of the quadrat to the minimum and thus help reduce the error associated with the observer deciding if an individual of a rare weed species is in or out of the quadrat [26].

Quadrat size, however, is much more important than quadrat form as the data are of frequency type; correct quadrat size is preponderant as the X^2-test will be used to decide if some specific association between two weed species occurs more often than would be expected by chance [26]. Quadrat sizes which are appropriate to the study of small plants as arable weeds are of 10 × 10 or 25 × 25 cm [40]. As bigger the weeds to be studied, as bigger should be the quadrat size. This makes sense since as bigger the plant individual, as bigger its circumference of influence over the nearby vegetation. Supposing a larger-than-advised quadrat is used for the study, there will be an unreal increase in the number of reported associations by the X^2-test.

Another point to be discussed is that the X^2-test should not be used “as is” when any of the expected values is less than “5”. In this case, Fisher’s exact test would be chosen [40] but as this test is also involved in a series of statistical controversies, the advice is to use a great number of sampling points per area (at least 100) and to apply the Yates’ correction to the X^2-test, what can be done automatically by most up-to-date statistical softwares as “R”. This should be enough for the weed science.

For each sampling point, all plant species rooted into the quadrat should be identified and recorded; there is no need to count the number of individuals per species or to assess its dry mass. Plant species which are not known at the time of the evaluation should be identified by a number and have a sample collected for posterior identification by a plant taxonomist (**Figure 6**).

The plant species should be listed by sampled quadrat and compared in pairs, the data being organized in 2 × 2 contingency tables, as follows [40]:

		Species “x”		
		+	-	
Species “y”	+	*a*	*b*	*a+b*
	-	*c*	*d*	*c+d*
		a+c	*b+d*	*n*

where: a = number of quadrats containing both species; b = number of quadrats containing only species "y"; c = number of quadrats containing only species "x"; d = number of quadrats with both species absent; and n = total number of quadrats in the contingency table

The association between plant species is estimated by the chi-square (X^2) test, usually at 5 or 1% significance, simply represented by the formula [40]:

$$X_T^2 = \frac{(\text{obs} - \text{exp})^2}{\text{exp}} \tag{16}$$

where: X_T^2= traditional chi square estimation; obs = observed values for species occurrence; and exp = expected values for species occurrence.

The expected values for each pair of occurrence in the 2 × 2 contingency tables are estimated by using the observed values from field sampling, as follows (**Table 3**).

As the association analysis uses 2 × 2 contingency tables, there is only one degree of freedom for the X^2 test. Besides, by anticipating that rare species (whose expected frequency is less than "5") may be reported, it is recommended to use Yates' correction for the X^2; thus, the following formula should be adopted as replacement for the traditional X^2 test, which automatically applies Yates' correction [40]:

$$X^2 = \frac{n[(|ad - bc|) - 0.5n]^2}{(a+b)(c+d)(a+c)(b+d)} \tag{17}$$

where: X^2 = chi-square estimation with Yates' correction; a = number of quadrats containing both species; b = number of quadrats containing only species "y"; c = number of quadrats containing only species "x"; d = number of quadrats with both species absent; and n = total number of quadrats in the contingency table.

The results of the calculated X^2 should be compared to the respective tables at one degree of freedom, and the results presented as a chi-square matrix crossing all species in pairs (**Figure 7**). The results indicate no association with a given pair of species when the probability in the chi-square tables, at the given number of degrees of freedom, is higher than the

Symbol/description		Expected quadrats[1]
a	"x" and "y" present	((a + b)/n) × (a + c) = K
b	"y" present	(a+b) − K = L
c	"x" present	(a+c) − K = M
d	none present	n − (K + L + M)

[1]The number of expected quadrats should be estimated by using the observed field values.
Source: adapted from Barbour [26].

Table 3. Contingency table estimators of expected values of occurrence for analysis of association between two species, "x" and "y".

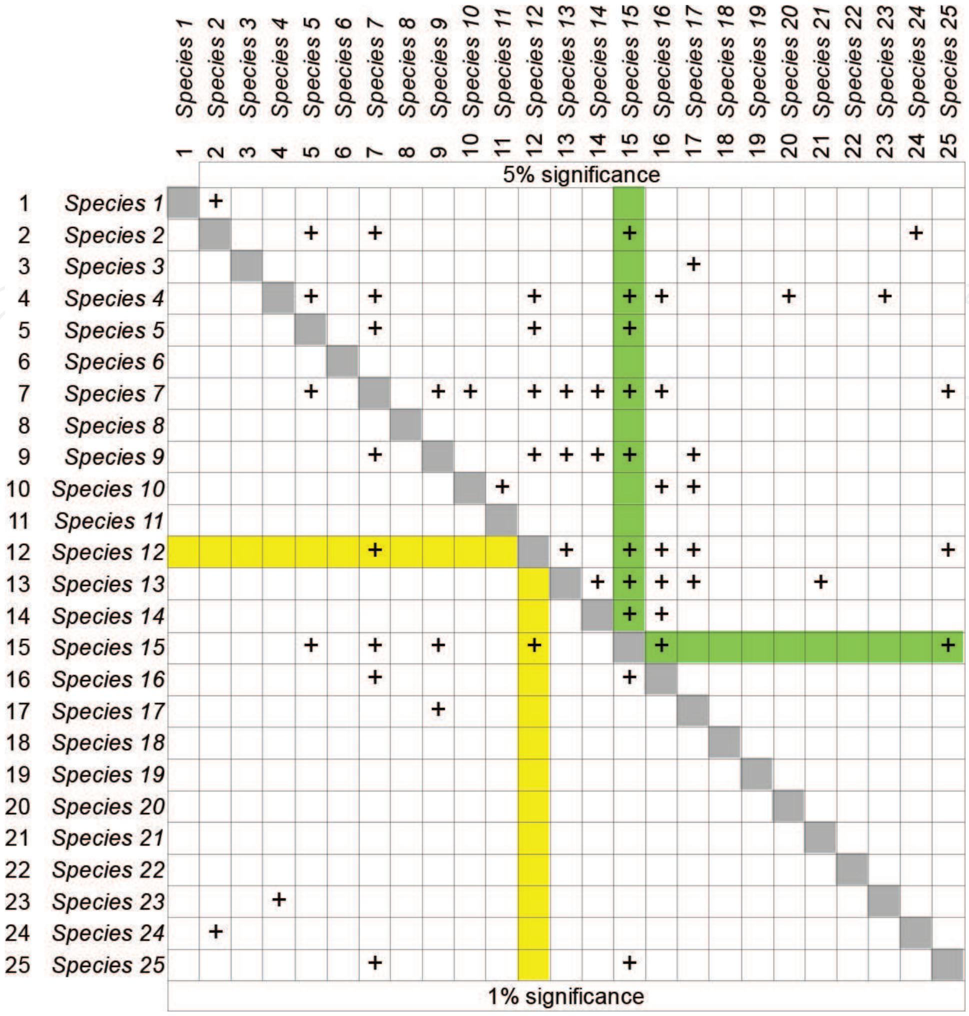

Figure 7. An example of an association matrix involving 25 weed species, at both 5 and 1% probability, according to the X^2-test. For reference, the association trees for species 15 (at 5% probability) and species 12 (at 1% probability) are highlighted in green and yellow, respectively. In this example, all associations are positive.

established significance level (usually 5 or 1%). The association is considered significant, as positive or negative association between two species, when the *observed values* of occurrence for a given pair of species are significantly higher or lower than the *expected values*, respectively [26]. An example of association matrix is supplied in **Figure 7**.

A dissimilarity between areas, Eqs. (14) and (15), is also useful for studies of plant associations. Similarly, diversity for intra-characterization of the areas may be applied to plant association studies in the same way they are applied for pure phytosociological studies (Eqs. (8) and (9)).

The relative occurrence of species and botanical families in the sampled area may be determined by its frequency of occurrence, usually considering the number of quadrats in which a given species is reported, related to the total number of quadrats, disregarding the number of individuals per quadrat:

$$Fr = 100\frac{f}{F} \tag{18}$$

where: Fr = frequency of occurrence for the given species; f = number of quadrats where the given species was present; and F = total number of sampled quadrats.

The frequency may be presented in several ways, but wordclouds make it easy to be understood. In wordclouds, the font size used to write the name of each species or family is proportional to their respective values of frequency (**Figure 6**).

4. Objections to the phytosociological method and application of the theory

Although being used for some years as a tool for the weed science, phytosociological surveys applied to arable fields have its drawbacks. As these methods were originally designed to describe natural environments, usually free from heavy anthropogenic effect, adaptations were needed for the agricultural context where the current flora present into the field is usually and mostly a result of the last cropping season's management (soil tillage system, fertilization levels, and herbicides applied, among other factors).

The main adaptations were (1) to establish the basic five steps for a reasonably complete phytosociological analysis, as described in the present text (overall infestation, phytosociological tables, diversity, similarity, and association); (2) to suggest and give preference to formulas which are less impacted by the most preponderant factors which could distort the phytosociological analysis, mainly for diversity and similarity; and (3) to use the method not only directly to the current flora into a given area but also to its seedbank through a germination study into controlled environment, as suggested by Concenço [60], and later comparing both studies (surface and seedbank samplings).

Another issue in the application of the method is its difficulty for both data collection in the field and its processing into the office, compared to what the researchers are familiar to analyze. Most weed science researchers usually adopt the visual method of evaluation for quantifying the occurrence of weeds into a given arable field, but this information is as easy as vague; it consists in taking note of the percentage of occurrence of each weed species into the field or alternatively—mainly following a herbicide application—evaluating the percentage of weed control some days after herbicide application. This method, although traditional and easy, does not supply at all information regarding the long-term behavior of weeds into the evaluated fields or its trend of occurrence for the next cropping seasons.

Another difficulty in applying the phytosociological methods for weed surveys is probably to convince the established weed science researchers to shift from the traditional evaluation methods (based on percentage of weed occurrence and control) to the phytosociological scope. The literature, however, proves that the adoption of such methods is highly positive for the sustainability of herbicide recommendations and weed management in the long term. One of the first Brazilian studies to apply the phytosociological method to the weed science, although in simple terms, was conducted by Carvalho and Pitelli [61]. Later, studies by Jakelaitis [62], Tuffi-Santos [63], Adegas [64], and several others adopted with success the phytosociological method for studies in weed science.

Although the use of phytosociological methods in the weed science is not new, the set of methods adopted is not standardized and ranges from basic to complex and from suitable to nearly unsuitable, depending on the paper. This makes almost impossible to compare studies conducted by different researchers as formulas and procedures are unlikely to be equivalent. The present chapter, however, partly intends to standardize the methods and its application.

5. Future insights

Weed science researchers will soon note that the traditional way of evaluating weed occurrence, infestation, or severity needs to move from a passive and subjective visually based assessment to most data-based decisions, and the phytosociology tends to be consolidated as the preponderant tool in this new universe of the weed science.

The difficulty in data collection for the phytosociological methods is still to be solved, but in the next few years, technologies such as GPS-driven drones with infrared imaging ability may be able to make data collection easier. Regarding data processing, the office work may still be an issue, but there are specific scripts for statistical softwares which could make the task of processing and interpreting the data easier, as the one published by Concenço [65], which makes possible to automatize phytosociological data processing into the statistical environment "R". This script, unfortunately, does not process the section of plant associations in its current version but is still a valuable tool that is freely available and adaptable.

Finally, an automatized integration from data collection into the field by GPS-driven drones, its transference to office and automatic processing by phytosociology software would provide farmers and technicians valuable tables and graphs for supporting both immediate and long-term decision-making in weed management.

6. Final considerations

This chapter discussed how some elements of phytosociology in ecology and botany can be used into the weed science as a tool for several inferences in arable fields. This is important to support recommendations for good agricultural practices while keeping up with biological conservation.

While choosing a sampling methodology for population studies in weed science, two questions should be taken into consideration: (1) *"what should be known?"* and (2) *"what will be done with the information?"* The first question addresses to the main type of information to be collected: richness or abundance. The second responds to the very purpose of sampling: biodiversity, evaluation of weed management plans, or studies in biology or ecology. By answering these two questions, one can design and choose a sampling methodology according to the information needs.

Weed relationships with edaphoclimatic traits show that the weed community is sensitive to variations in pH, water, temperature, and other resources and conditions. Each weed population is mostly competitive and dominant in those locations that meet particular conditions, and this would allow weeds to be used also as bioindicators as well as help understand the long-term dynamics of weed communities.

It is important to evaluate other sampling methods in order to know the sensitivity and accuracy of such alternatives in comparison with the ones presented here for distinct sampling objectives. One should ever think that as more species coexist in an association, and as greater sized are the plants, as bigger should be the minimum area to be sampled.

Phytosociological surveys are useful as tools to shed light on the dynamics of weed species and their interactions in arable fields. The methods, however, are the most diverse as several indices and coefficients are available, depending on the literature used as a reference by a given author. Basic care should be taken, however, when sampling and describing the plant community as well.

The following sequence of steps is proposed as suitable for phytosociological studies: (1) overall infestation; (2) phytosociological tables and/or graphs; (3) intra-characterization by diversity coefficients; (4) inter-characterization and area grouping by multivariate analysis; and (5) weeds association through contingency tables by means of the chi-square test. Other ways for presenting data should still be suitable, depending on the nature of the environment to be studied—arable fields in this case.

Literature is not clear about a set of methods for phytosociological studies, and one will hardly be able to find all the information and equations into the same source. Even classical references miss some aspects of phytosociological surveys, and some papers were published by using an unsuitable set of ecological methods to describe the weed community.

In the present chapter, a summary of methods was made in order to assist weed science researchers through their first steps into the realm of phytosociology.

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