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Geomatic Techniques to Support Phytosanitary Products Tests whithin the EPPO Standard Framework

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Chapter 1

Introduction

1.1 EPPO

1.1.1 Phytosanitary Products

Phytosanitary products, commonly used as a synonym for ”Plant Protection Products” (PPPs), are a specific category of pesticides designed primarily to maintain crop health and prevent destruction by diseases and infestations. While the term ”pesticides” is broader and also includes biocidal products used to control harmful organisms and disease carriers not related to plant protection, phytosanitary products are specifically used to control harmful organisms affecting cultivated plants (such as insects, mites, fungi, bacteria, rodents, etc.), eliminate weeds, and regulate plant physiological processes. Fertilizers, which serve for plant nutrition and soil fertility improvement, are excluded from phytosanitary products.

Phytosanitary products contain at least one active substance, which can be either chemical compounds or microorganisms, including viruses, that enable the product to perform its intended function. These active substances undergo rigorous risk assessment processes, with EFSA (European Food Safety Authority) playing a central role in conducting peer reviews at the EU level to determine if these products, when used correctly, might produce harmful effects on human or animal health, either directly or indirectly through drinking water, food, or feed.

The main categories of phytosanitary products can be distinguished based on the type of organism they target or the function they perform, including:

- Fungicides
- Insecticides
- Acaricides
- Rodenticides
- Slimicides
- Nematicides
- Herbicides
- Plant growth regulators

The parameters identified through the risk assessment are compared with the values established by directive 97/57/EC [8], which

indicates the acceptability limits for decision-making on the inclusion of active substances in the EU list (Annex I of directive 91/414/EEC [3]).

The Introduction of a product in the EU market is not only subject to audits on active substances and their safety for humans and environment but also to the evaluation of the product's efficacy and safety for the crop. World Trade Organization Sanitary and Phytosanitary Measures Agreement [11] recognizes the International Plant Protection Convention (IPPC) as the only international institution in charge of emitting standards for plant health [10]. IPPC is organized in regions. European Union (EU) countries refer to the European and Mediterranean Plant Protection Organization (EPPO). EPPO Standards are divided into Standards on Phytosanitary Measures and Standards on PPPs. PPPs standards describe the efficacy evaluation of PPPs (PP 1) and good plant protection practices. EU GEP units provide Biological Assessment Dossier (BAD) efficacy trials. GEP units are expected to follow EPPO PP 1 to assess PPPs selectivity detecting phytotoxicity effects, and efficacy in the complaint of Regulation (EC) No 1107/2009 of the European Parliament and Council [9].

1.1.2 Standard Experimental Design

Generics on efficacy assessments are reported in PP 1/181(5) [7], which describes herbicide, fungicide, bactericide, and insecticide efficacy on the target evaluation. PP 1/135(4) [5] describes the selectivity assessment procedures, in other words: the standard phyto-

toxicity assessments of PPPs. The PP 1/152 [4] standard describes the general principles for the efficacy and selectivity evaluation of PPPs, in describing the standard experimental design. Aside from the objectives of the study and the description of thesis (treatments), the PP 1/152 outlined that a comprehensive experimental design should include a description of:

- **Type of Design**
- **Sampling Method and Measures Units**
- **Statistical Analysis Plan**

For what concerns the type of design, EPPO ”envisage trials in which the experimental treatments are the ‘test product(s), reference product(s) and untreated control, arranged in a suitable statistical design’” [4]. The experimental design should be randomized, with replications and blocks, and should include a sufficient number of plots to ensure the statistical power of the analysis. The number of replications and blocks should be determined based on the expected variability of the data and the desired level of statistical significance in respect control and reference thesis. The randomization of thesis within blocks should be carried out using a suitable randomization procedure to ensure that the treatments are assigned to plots in a completely random manner. The key randomization used in phytosanitary product evaluations include:

- **Completely Randomized Design (CRD)**: Treatments randomly assigned to experimental units; statistically powerful but only

suitable for homogeneous trial areas where environmental variation is minimal.

- **Randomized Complete Block Design (RCBD)**: Groups plots into homogeneous blocks with each treatment appearing once per block; controls for environmental heterogeneity across the experimental area.
- **Split-Plot Design**: Used when one factor (e.g., cultivation equipment) cannot be fully randomized; creates hierarchy with whole plots and subplots; particularly useful when plot size or equipment constraints exist.
- **Systematic designs**: Non-randomized arrangements rarely suitable for efficacy evaluations; may only be appropriate in special cases like varietal trials on herbicide selectivity.

When designing phytosanitary product trials, the arrangement of untreated controls is critical for proper efficacy assessment. According to EPPO standards, the main purpose of untreated controls is to demonstrate adequate pest infestation, without which efficacy cannot be meaningfully evaluated. Four distinct arrangements for untreated controls exist:

- **Included controls**: The most common approach, where control plots have the same shape and size as treatment plots and are fully randomized within the experimental design. This arrangement is essential when controls will be used in statistical comparisons.

- **Imbricated controls**: Control plots are arranged systematically within the trial (between blocks or between treated plots), potentially with different dimensions than treatment plots. These observations are typically not included in statistical analyses but ensure more homogeneous distribution of untreated area effects.

- **Excluded controls**: Control plots are established outside the main trial area but in similar environmental conditions. While replication is not essential, it may be beneficial in heterogeneous environments. These observations are generally excluded from statistical analyses.

- **Adjacent controls**: Each plot is divided into two subplots, with one randomly selected to remain untreated. This approach is particularly valuable in highly heterogeneous environments but requires specialized split-plot statistical analysis.

The selection of control arrangement depends on several factors: whether the control will be included in statistical tests (requiring included controls), the degree of environmental heterogeneity (adjacent controls are preferred for high heterogeneity), and the potential for control plots to interfere with adjacent treatment plots (suggesting excluded controls when interference is likely). The trials type design is critical for the success of the study, as it ensures that the results are reliable, reproducible, and statistically valid.

After defining the experimental units through the randomization design choise, the next step is to define the sampling method and the

measures units. Target and crop-specific standards point out ”mode of assessment recording and measurements” fixing evaluation metrics in two ways: countable (discrete values) and measurable (continuous values) effects which must be expressed in absolute values, in other cases, frequency (incidence) and degree (severity) should be estimated and reported as affected percentage of the individual (ex. plant or plot) or as proportion within thesis and control expressed in percentage. As specified by PP 1/152 [4], classification by ranking (ordinal) and scoring (ordinal or nominal) is also contemplated. In the case of estimation, rather than count or measure, PP 1/152 reports ”The observer should be trained to make the estimations and his observations should be calibrated against a standard”. Calibration compliance with standards is ensured by GEP audits. Scoring and ranking scales examples are published on specific standards or the same PP 1/152. The lack of specific scales lets trial protocol authors define one inspired in range and intervals by the mentioned examples or other well-established ones. GEP units PP 1 assessments are produced by trained and experienced agronomists or biologists by visual inspection or laboratory analysis. The technician follows the trial protocol and related EPPO standards during assessment execution. The technician is critical for accuracy, precision, and repeatability. Sensitivity is determined by the trial protocol. It depends on expected differences and if a measure, a proportion, or a scale is used. For instance, in PP 1/93(3) [6] ”Efficacy evaluation of herbicides - Weeds in cereals - Observation on the crop”, phytotoxicity color modification could be measured, or estimated as proportion in respect to the untreated, or scored in EPPO scale as

PP 1/135(4) reports, or a scientifically accepted score as the European Weed Research Society phytotoxicity damage score [2] and other ones. In general, data types must undergo the classification presented in Table 1.1

Table 1.1: Different modes of observation and types of variables

Type of Variable	**Measurement**	**Visual Estimation**	**Ranking**	**Scoring**
Binary				X
Nominal				X
Ordinal			X	X
Discrete	X	X		
Continuous limited	X	X		
Continuous not limited	X	X		

The statistical analysis of trials is equally critical, providing objective assessment of treatment effects. While PP 1/152 [4] doesn't prescribe specific analyses for all situations, it emphasizes that analysis methods should align with the experimental design and data types collected. For qunatitative variables (continuous or discrete), parametric methods based on Generalized Linear Models (GLM) are recommended, including ANOVA and regression approaches. For qualitative variables (ordinal or nominal), non-parametric methods are more appropriate. Parametric analysis assumes additivity of effects, homogeneity of variance, and normally distributed errors—when these assumptions aren't met, data transformations or alternative approaches become necessary.

Statistical tests, particularly F-tests of orthogonal contrasts, should focus on biologically relevant comparisons specified during the design stage: untreated control versus treatments (establishing trial validity), reference products versus control (demonstrating coher-

ence), test products versus reference (evaluating efficacy), and comparisons among test products (identifying superior treatments). For efficacy trials, EPPO suggests one-sided tests since the aim is comparing products against references or controls, with appropriate multiple comparison procedures when needed.

Through adherence to these rigorous design and analysis standards, researchers can generate reliable evidence to support phytosanitary product registration while ensuring that products demonstrate consistent efficacy across relevant agricultural conditions.

1.1.3 Digital Approaches

While the EPPO experimental design standards provide a solid foundation for conducting phytosanitary product trials, the increasing availability of digital tools and technologies offers new opportunities to enhance the quality (in the ”Quality of a mode of observation” sense [4]) and efficiency of these assessments. Digital approaches can automate data collection and analysis, improving the reproducibility of results, ultimately accelerating the development and registration of effective phytosanitary products.

To regulate the use of this kind of technologies, the EPPO published a new standard, PP 1/333(1) [1], which filled the gap in the use of digital technologies in phytosanitary product efficacy and selectivity trials. This standard provides guidelines for incorporating digital tools into trial protocols, where digital tools are intended as a combination of hardwares and softwares delivering samples quantitative or quali-

tative measurments of the samples in a semi-automatic or automatic fashon. The delivered measurments must respect the same quality standards of the manual ones, and the digital tools must be validated before the trial execution. The standard also provides guidance on the validation of digital tools, which should be performed by comparing the results of digital and manual assessments. The validation report should demonstrate that the digital tools provide reliable and consistent results compared to manual assessments. The benchmarks for the validation depends on the type of variables measured:

- **Continuous**: digital measurment correlation with manual measurment must deliver a coefficient of determination (R) higher than 0.85.
- **Ordinal and Nominal**, the validation should include the comparison of the frequency of the different classes of the digital and manual assessments.
- **Binary**, the validation should include the comparison of the frequency of the two classes of the digital and manual assessments.

1.2 Geomatic Technics

1.2.1 Photogrammetry

1.2.2 Geostatistics

1.3 Machine Learning

1.3.1 Approaches

1.3.2 Computer Vision

Chapter 2

Thesis Aims and Framework: A New Statistical Analysis Workflow

Chapter 3

Study Cases

3.1 Continuous Variables

3.1.1 Plant Count

3.2 Ordinal and Nominal Variables

3.2.1 Phytoxicity Score

3.3 Binary Variables

3.3.1 Embedding Spaces for Control Sample Anomaly Detection

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