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How can my research paper be useful for future meta-analyses on reforestation practices?

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Abstract

Statistical meta-analysis is a powerful and useful tool to quantitatively synthesize the information conveyed in published studies on a particular topic. It allows identifying and quantifying overall patterns and exploring causes of variation. The inclusion of published works in meta-analyses requires, however, a minimum quality standard of the reported data and information on the methodology used. Our experience with conducting a meta-analysis on the relationship between seedling quality (seedling size) and field performance (seedling survival and growth) is that nearly one third of the apparently relevant publications had to be discarded because essential data, usually statistical dispersion parameters, were not properly reported. In addition, we encountered substantial difficulty to explore the effect of covariates (moderators) due to the poor description of nursery cultivation methods, plantation location and management in a significant proportion of the selected primary studies. Thus, we present guidelines for improving methodology detail and data presentation so that future reforestation-oriented research can be more readily incorporated into meta-analyses. This will help to quantitatively synthetize current state-of-knowledge and thus contribute to the advancement of the reforestation discipline.

Keywords: Data quality; Data reporting; Meta-analysis; Methodology guideline; Seedling quality; Research synthesis

INTRODUCTION

More than 2 billion hectares of our planet are in need of forest restoration (Minnemeyer et al. 2011). Outplanting seedlings will play a major role in this restoration effort (Stanturf et al. 2014). In addition, future reforestation activities will be necessarily focused on harsher sites (Oliet and Jacobs 2012). While defining the appropriate seedling stocktype to meet these needs can be achieved through a variety of methods, including the Target Plant Concept (Dumroese et al. 2016), an understanding of the interplay of nursery production techniques and factors on outplanting sites is necessary to ensure reforestation is most effective. Crucial to this understanding, and subsequent successful reforestation, is seedling quality, an often overlooked factor in many reforestation studies. A quality seedling has high potential to survive and grow adequately after outplanting under particular environmental conditions (Duryea 1984), and reflects the integration of multiple physiological and morphological attributes (Ritchie 1984) that drive the seedling's ability to become established (Grossnickle 2012).

Since early in the twentieth century, forest researchers and practitioners have been intrigued by the plant attributes that affect seedling performance after outplanting. Starting with the pioneering work of Wakeley (1954) initiated in the 1930s on the effect of seedling morphological attributes on outplanting performance, a vast number of studies assessing seedling quality attributes have been published. These studies have covered a wide range of species and forest ecosystems, and numerous morpho-physiological attributes (Duryea 1985) determined by different nursery cultivation practices. Despite several qualitative reviews on seedling quality (Ritchie and Dunlap 1980; Ritchie 1984; Duryea 1985; Wilson and Jacobs 2006; Grossnickle 2012, 2017; Grossnickle and El-Kassaby 2016), to the best of our knowledge this discipline lacks any quantitative reviews. This is unfortunate because several topics on seedling quality and forest plantations are controversial, such as the relationship between outplanting survival and seedling size (Trubat et al. 2008; Villar-Salvador et al. 2012), and are likely the result of the interactions of several factors, such as species, stocktype, and local climate that limit the capacity of qualitative reviews to describe general trends. Therefore, quantitative reviews based on statistical approaches are needed to increase our ability to synthetize and generalize the vast amount of knowledge on the interaction of seedling quality and outplanting site characteristics accumulated during the past 70 years. This is pivotal to guide new reforestation research and to provide decision-makers with evidence-based support (Stewart 2010).

Meta-analysis is a powerful, informative, and unbiased tool to quantitatively summarize evidences for a particular research question (Koricheva and Gurevitch 2014). This technique integrates several statistical methods for combining results from independent, primary studies in order to identify general patterns and to evaluate factors that may cause heterogeneity in outcomes among studies (Koricheva et al. 2013). Therefore, the application of meta-analysis techniques to the wealth of studies on seedling quality and outplanting performance may help untangle the contradictory results in this topic, such as the above-mentioned relationship between seedling morphological attributes and their post-planting survival (Grossnickle 2012), and thus contribute to the advancement of the reforestation discipline. The inclusion of primary studies on meta-analysis strongly relies, however, on the appropriate reporting of data and an exhaustive description of the methodology used, study characteristics, and location (Hillebrand and Gurevitch 2013; Gerstner et al. 2017). In this regard, the establishment of high quality standards in reporting results and methodology of published studies would increase the soundness and quality of future meta-analyses. This is especially important in seedling quality research where no quantitative reviews have been conducted.

In this article, we present a specific checklist and guidelines for reporting methodologies, data, and statistical results in reforestation research involving the use of nursery-produced seedlings. The motivation for this article arises from our experience in conducting a meta-analysis on seedling quality with an objective of elucidating if an overall effect, whether positive or negative, exists between seedling size at outplanting and their survival. Following existing protocols for searching relevant literature (Côté et al. 2013) and after establishing restrictive inclusion criteria, we identified 306 studies for further evaluation. Of these, 94 were discarded because essential statistical data required for the meta-analysis were not reported. In addition, only about half of the 306 studies provided basic information, such as field location, site preparation techniques, post-planting management, or previous land use, which hampers evaluating the influence of these factors on the survival-seedling size relationship. Some protocols for reporting data and methodologies have been published during the last few years in other disciplines, such as ecology, evolutionary biology, or medicine (Hillebrand and Gurevitch 2013; Zuur and Ieno 2016; Goodman et al. 2016). More recently, Gerstner et al. (2017) proposed updated guidelines along with a specific example of proper data reporting for ecological studies. It seems, therefore, appropriate to adapt existing protocols for high-quality publication standards to specific disciplines in order to improve the relevance of future meta-analysis on these topics. While this is our main objective here, we also aim to provide guidelines for improving the impact of seedling quality research, and how it impacts reforestation success, to be published in the future.

A BRIEF DESCRIPTION OF THE BASIS OF META-ANALYSES

Although a full description of the principles of meta-analyses is beyond the scope of this article, a basic knowledge of the meta-analysis procedure is key to understanding how data and information should be reported. Meta-analysis was originally developed for social sciences and medicine and since the 1990s it has gained prominence in other disciplines, such as ecology. This has led to excellent publications about the application of meta-analysis to ecological studies (Koricheva et al. 2013), which should be consulted by anyone interested in an up-to-date guide to conducting meta-analyses.

The first step in a meta-analysis consists in a systematic search of literature in the target topic. This involves establishing search protocols based on the combination of relevant keywords and the use of electronic search engines and databases (Côté et al. 2013). The primary databases in biological sciences are Web of Science, SCOPUS, and Google Scholar, yet relevant studies are often published outside these main traditional distribution channels constituting the so-called "grey literature". This is an especially significant source of seedling quality and reforestation research, where a substantial number of studies are published in local journals, conference proceedings, and technical reports. In this regard, specific initiatives such as the Reforestation, Nurseries, and Genetic Resources database (USDA Forest Service and Southern Regional Extension Forestry; https://rngr.net) are extremely useful in reaching grey literature. As researchers, to ensure our work is found in any systematic search, we should bear in mind the appropriate choice of keywords, an informative title, and abstract content. In this regard, journals strongly encourage authors follow their suggestions and standards well aware that this is a pivotal point to increase the likelihood of being reached through a literature search. The same applies to grey literature even if there are not the strict scientific quality and visibility rules as in formal scientific literature. In addition, from the meta-analysis perspective, the number of keywords used in the literature search has to be limited, otherwise the search output will include a large number of studies that are not relevant for the objective of the meta-analysis (Côté et al. 2013). In this regard, authors should find a balance between providing a broad vision of their work in order to be found in a broad literature search while also being sufficiently specific to be identified as relevant through a quick reading (Gerstner et al. 2017).

Once relevant studies have been identified, the second step is data extraction and its incorporation into a database. The critical information extracted is an estimate of the magnitude and direction of the outcome of the study. The outcomes of the selected studies must be then expressed on a common and comparable scale, known as effect sizes, in order to be analyzed in the meta-analysis (Rosenberg et al. 2013). Together with the effect size, it is necessary to know the precision associated to the estimation of the effect (e.g. variance, standard error, or confidence interval). This estimation of the precision from each study is used to weight its contribution to the overall effect, which is estimated together with a confidence interval. Then, we can evaluate whether the overall effect is significantly different from zero or test if any covariate might explain heterogeneity in the outcomes among studies.

"EFFECT SIZE THINKING" WHEN REPORTING RESULTS

Recently, (Parker et al. 2016) reported that about half of published articles lack key information about statistical results, which severely constrains the utility of primary research for meta-analysis. It is therefore imperative to make scientists aware of an "effect size thinking" when reporting data in research studies (Nakagawa and Cuthill 2007). In this context, a clear understanding of the different effect size metrics and their calculation would greatly help to increase the relevance of primary research for meta-analysis.

In general, research studies should report data on means, sample size, and any measure of variation (Figure 1), which must be clearly identified in the text or in figures and table captions (e.g. standard error, standard deviation, or 95% confidence interval). In addition, any hierarchical design or data aggregation should be clearly explained (Gerstner et al. 2017). This is of special relevance in seedling quality and reforestation research because field plantations are often conducted in blocks or plant attributes are usually measured in groups of plants (i.e. composite samples for nutrient analysis). Moreover, researchers often publish only a portion of the results derived from data analysis. This leads to publication bias, especially when only significant results are reported in papers (known as *p*-hacking). Ignoring weak or absent patterns when reporting data might, however, limit our capacity to estimate unbiased overall effects in meta-analysis (Parker et al. 2016). Nowadays, there is no reason to report only strong or significant relationships because journals allow the incorporation of unlimited pages as electronic supplementary material.

The most useful effect sizes for meta-analyses on seedling quality are standardized mean differences, response ratios, odds ratios, and correlation coefficients (Figure 1) (Rosenberg et al. 2013). Standardized mean differences and response ratios are used to compare mean values of two groups that often represent an experimental treatment and a control (Figure 1). This is the case, for example, when testing whether a nursery (e.g. fertilization) or field management technique (e.g. ripping) improve seedling field performance. The most common and appropriate metrics for comparing pairs of means are Hedges' *d* and the natural log of the response ratio (Rosenberg et al. 2013). If two groups are compared for binary response variables (e.g. alive *vs* dead) based on a contingency table, the most widely used effect size is the odds ratio. The Pearson's correlation coefficient is the appropriate effect size when the aim is the relationship between two continuous variables (Figure 1) (e.g. the effect of seedling morphology at outplanting on the field performance). As the distribution of the Pearson's correlation coefficient becomes skewed when it approaches ± 1, the Fisher's *z*-transformation is used to obtain an effect size with desirable statistical properties. The variance associated to a correlation coefficient is calculated from the sample size, thus it should be always provided when reporting correlation coefficients. One of the advantages of using the Pearson's correlation coefficient as effect size is that it can be calculated from a wide array of other statistics (Lajeunesse 2013), such as Student's *t*, *F*-ratio, χ^2, or Spearman's correlation coefficient among others.

REPORTING METADATA IN RESEARCH PAPERS

Meta-analysis not only serves to calculate an overall effect, but also to explore the cause of variation in the magnitude of the outcomes by examining the effect of covariates (moderators) (Koricheva and Gurevitch 2014). For instance, it might be relevant to assess how precipitation on plantation sites influences the effect of field fertilization on seedling growth. Thus, a detailed description of experimental methods, study design, and study area is crucial to evaluate causes of heterogeneity in the outcomes of primary studies. Despite this seeming obvious, our experience in conducting a seedling quality meta-analysis revealed that many research studies frequently fail to include a full description of this basic, above-mentioned information. For example, we found that about half of finally selected studies lack the exact geographical coordinates of the plantation site, which is essential for accessing climatic data. Gathering such missing data for a meta-analysis is a time-consuming task that sometimes involves contacting authors, which we found is not always successful. Here we propose a checklist of relevant information about seedling production and outplanting that we believe should be included in the material and methods of any reforestation study, especially the effects on and of seedling quality, in order to make it valuable to future meta-analyses (Table 1).

Information about seedling production in the nursery

Nursery production techniques strongly influence seedling quality (Landis 1989; Dumroese et al. 2009). Providing full information about all steps involved in the production of nursery seedlings is essential to test whether these procedures might have an effect on seedling quality (Table 1). The first thing to describe in detail is the plant material. The species name should be from a widely accepted and available taxonomic list, such as *the plant list* (http://www.theplantlist.org/), otherwise it can be difficult to match studies using the same species. The origin of seeds should be described in detail, including the provenance(s) and, if available, collection information such as location, date, and number of mother trees. Seed storage, seed selection protocols, and/or conditions and techniques used for germination are also interesting procedures to be reported.

Once plant material has been correctly described, ensure a full description of experimental and seedling growing conditions is provided (Table 1). Geographical coordinates of the nursery will be useful to determine climatic conditions under which seedlings were grown when cultivation is outdoors, as this might influence seedling quality and post-planting performance (Mollá et al. 2006). The information to be included in the description of seedling growing conditions in the nursery will depend on the planting stock raised. On one hand, bareroot and container seedlings are the two basic stocktypes in forest nurseries (Grossnickle and El-Kassaby 2016). Briefly, bareroot seedlings are grown in free soil in outdoors nurseries normally for one to four growing seasons, while container seedlings are grown in cavities with artificial media in outdoor or greenhouse nurseries normally for one to two years. This different cultivation procedure has important implications for seedling quality attributes (Grossnickle and El-Kassaby 2016). On the other hand, there are many variations for the production of these two basic stocktypes that should be reported in the methodology section. For example, bareroot seedlings can be produced under different cultivation densities in one or various seedbeds (Hahn 1984; Thompson 1984). Thus, for bareroot seedlings report stocktype age notation together with the exact dates of seeding and transplantation, as well as the cultivation density during each stage of production. Container seedlings can be grown in a wide variety of container types differing in volume and density (Dominguez-Lerena et al. 2006). Therefore, information on container density, volume, and dimensions (width, length, and depth) should be provided. This is particularly important for stocktype trials to ensure that confounding of independent factors is not an issue (Pinto et al. 2011). In addition, the spatial configuration of containers in the nursery (blocks), and the physico-chemical characteristics of growing media used for filling containers must also be detailed (type of substrate, pH, nutrient content).

Irrespective of the stocktype, studies should contain information about the environmental conditions under which seedlings were grown (Table 1). Specifically, light level (especially if shaded), watering, and fertilization regime. These cultivation factors, together with container volume and cultivation density strongly influence seedling morpho-physiological attributes and consequently outplanting performance (Driessche 1982; Villar-Salvador et al. 2004; Dumroese et al. 2005; Dominguez-Lerena et al. 2006; Puértolas et al. 2009; Andivia et al. 2014). Therefore, nursery cultivation treatments and procedures should be thoroughly described. For example, works testing different fertilization treatments should report information about the complete fertilization formulation, concentration, application frequency, and schedule (e.g. constant, exponential, late-season fertilization), and the total amount of N, P and K applied to each seedling during the cultivation. In addition, other common cultivation procedures applied during the nursery phase, such as seeding date, shoot and root pruning, use of growth regulators, mycorrhizae inoculation, or cold storage should also be reported.

Information about field plantation and management

Field trials are crucial for validating the suitability of nursery treatments and the identification of the seedling functional attributes that predict outplanting performance. Seedling quality interacts with plantation practices and site conditions to determine the success of a forest restoration program. In this context, the use of covariates related to site climate, soil preparation techniques, previous land use, or post-plantation management as moderators in meta-analyses is important for understanding if controversial issues on seedling quality are context-dependent (Table 2). This information is, however, not always available in research studies on seedling quality, in part because some plantation techniques and management strategies are so entrenched among forest practitioners that they are assumed and therefore go unreported in many research studies.

A detailed field site description is essential in any experimental and observational study. In the context of quantitative reviews, field site information can be used as covariates or to group primary studies (Table 2). Climate is a primary determinant of plantation performance (Squeo et al. 2007). The inclusion of the exact geographical coordinates is of great help to access mapped climate information, such as in the WorldClim database, but also to evaluate if a geographical bias exists in the selection of primary studies or in their outcomes. Even if the exact geographical location is included in the study, it is also helpful to provide climate data from local weather stations that might cover the specific conditions at the plantation site, and especially during the period evaluated. Beside climatic data, other information related to elevation, soil, slope (including aspect), or vegetation and presence of herbivores that might affect the plantation outcome would provide a detailed picture of the environmental context in which the plantation is conducted. Previous land-use (cropland or woodland) or degradation history in the area might also help to interpret results of individual primary studies or to use this information as moderators in the meta-analysis.

Site preparation and plantation techniques determine forest plantation success. These field techniques aim to improve soil conditions for improving water infiltration and rooting, controlling competing vegetation, and reducing animal damage, among others (Löf et al. 2012). Main soil preparation techniques include mechanical site preparation, prescribing burning, mulching, and the use of herbicides (Löf et al. 2012). A correct description of the techniques implemented before outplanting seedlings would enable the grouping of studies for meta-analysis or to facilitate further meta-analysis in this topic (Table 2). Among aforementioned soil preparation techniques, mechanical site preparation is the most widely used in forest plantations. Mechanical site preparation involves, however, a wide range of different techniques, intensities, and machinery, which makes it difficult to group studies according to this covariate if detailed descriptions are not reported. Recently, Löf et al. (2012) reviewed the state-of-knowledge concerning the use of mechanical site preparation in forest restoration projects and grouped techniques into three main categories: scarification, mounding, and sub-soiling/ripping. Other techniques, not included in this classification, like mowing, drum chopping, blading and piling can be considered as low intensity interventions, whereas deep plowing and terracing can be considered as very high intensity interventions.

Date of outplanting should be also included in the plantation description because it affects seedling outplanting performance, especially in cold and arid environments (Radoglou and Raftoyannis 2002; Palacios et al. 2009; Yang et al. 2013). In addition, by providing the exact date of plantation and first field evaluation of seedling performance it is possible to assess the effect of climatic conditions in a meta-analysis. Planting density and planting depth should be included because they have implications for seedling performance (Hainds 2004; Zhao et al. 2011; Oliet et al. 2012). Information about how the seedlings were outplanted (e.g. hand or machine) and if confounding techniques were avoided (Pinto et al. 2011) is also essential. In addition, the spatial design of the plantation field, and any obvious plot heterogeneity (e.g. different slope orientations) must be described. The date at which performance measurements were conducted is important to know exactly the period under evaluation. Finally, the use of ecotechnologies, such as tree shelters, organic amendments, mulching, and hydrogels (Piñeiro et al. 2013) should be described in detail. Specifically, tree shelters should be fully described because their size, ventilation, and light transmission have an influence on seedling survival and growth (McCreary and Tecklin 2001; de Castro et al. 2014).

Once seedlings are outplanted, several management and maintenance activities can be conducted that strongly impacts their performance. Weeding is a widespread maintenance activity in forest plantations, but can vary with site environmental conditions, planting density, and the species of weeds and outplanted seedlings (Gómez-Aparicio 2009; Kabrick et al. 2015). Thus, when weeding is performed information regarding its intensity, frequency, timing, and method should be included. Fertilization and irrigation can be done at outplanting and/or after the start of the plantation (Rey-Benayas 1998; Casselman et al. 2006). In both cases information should include when the practice was initiated, subsequent frequency, and the total amount applied per plant. For fertilization practices, the type and formulation of the fertilizer should be provided. Other maintenance and management activities, such as replanting, pruning, or thinning should also be informed.

CONCLUSIONS

Here we provide general and specific recommendations for a comprehensive reporting of methodologies, data, and statistical results in seedling quality research. Following these guidelines when writing a manuscript will not only facilitate the work of researchers involved in meta-analyses, but also will increase the options of a primary study to be included in these reviews. Thus, this should be seen by authors as an opportunity to increase the visibility, scope, relevance, and pragmatic usefulness of their studies. Independently of whether a study is included in a meta-analyses, these recommendations are good practices for research reliability and confidence. For example, these guidelines can be used as a checklist to guide during the writing of the material and method section in any reforestation studies.

As mentioned above, the identification of relevant studies and the comprehensive reporting of data and methods are critical steps in the elaboration of a meta-analysis. Increasing the detail of methodology and data (including metadata) accessibility will promote the quality and value of subsequent reviews. On one hand, open access science might be a 'silver bullet' because most scientific journals presently allow this type of publication, but if the publication cannot be made open access other options such as global repositories (e.g. arXiv.org) or online platforms (e.g. ResearchGate) can still host versions of the manuscript with more detail. On the other hand, the online availability of raw data, either in the journal website or in global repositories (e.g. Dryad) is extremely useful for meta-analysis. This will reduce the number of papers discarded because of absent, essential data as well as the requests to study authors. In addition, it will facilitate extraction of data because obtaining data from figures in published papers is time-consuming.

In conclusion, the establishment of quality standards and guidelines for data and method reporting in published studies on seedling quality and outplanting performance will ensure the greatest number of studies will be included in any meta-analysis. This will better answer fundamental questions important to any phase of the reforestation chain.

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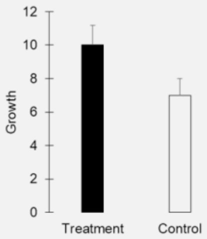
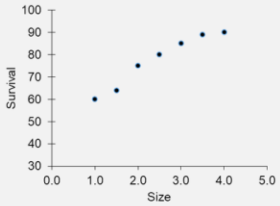
Table 1: Checklist of the information to be included in the description of the nursery phase in studies involving seedlings used in reforestation studies

Reporting information	
Seedling production in the nursery	
Plant material	Species name Seed origin (including provenance, site of collection and other relevant information about seed collection)
Seed handling	Seed storage Seed selection protocol Germination conditions
Seedling growing conditions	Nursery location (coordinates) Cultivation density Physicochemical characteristics of nursery soil or growing media Spatial configuration (blocks) Seeding date Stocktype notation (e.g. 1+0, 2+1) Transplantation date to other seedbeds (bareroot) Container type and size
Nursery treatments	Light levels Fertilization levels (including type of fertilization, fertilizer formulation, frequency of application and total amount of N, P, K) Watering levels (including frequency and total amount supplied)
Other factors	Shoot and root pruning Use of growth regulators Mycorrhizae inoculation Cold storage

Table 2: Checklist of the information to be included in the description the field plantation in reforestation studies

Reporting information	
Plantation and management	
Site description	Exact location (coordinates) Climatic conditions Soil conditions Elevation Slope and orientation Vegetation and herbivores in the area Previous land use
Site preparation and plantation	Site preparation technique (including brief description of the intensity and the machinery) Planting technique (hand or machine) Planting date Planting density Planting depth Spatial design Use and description of tube shelters
Plantation management	Weeding (including frequency, intensity, timing, and method) Fertilization and irrigation (including frequency, timing, and total amount) Other activities such as replanting, pruning, or thinning

Figure 1: Examples of questions on seedling quality research that can be addressed through meta-analysis (left column). The centre column contain information regarding the effect size used for each type of meta-analysis, and the right column provides examples of good reporting of data in primary studies.

Meta-analysis question	Effect size calculation	How to report
Has any treatment (either during the nursery phase or after plantation) an effect on field performance? 	**<u>Hedges' *d*</u>**: Calculated from the mean, associated deviation and sample size for the treatment and the control group. **<u>Natural log of the response ratio</u>**: Calculated from the same data that Hedges' *d*. **<u>Natural log of the rate ratio</u>**: It is used when results are the observed counts for two possible events (usually alive or dead plants). This data is usually reported in a 2 x 2 contingency table. It is calculated from the counts of events for the treatment and the control group.	Provide mean, associated deviation and sample size in the text, table, figure or figure caption: *Fertilization after planting significantly enhanced seedling growth (mean = 15.3 cm ± 2.4 SD, n = 300) compared to control plots (mean = 9.8 cm ± 1.9 SD, n = 278).* Even if results are not significant provide the exact values of the statistics (if means are not showed): *Soil preparation technique did not show a significant effect on seedling growth one year after plantation (F1,4 = 1.07, p = 0.32).*
Is there any relationship between any continuous variable measured in nursery seedlings and post-planting performance? 	**<u>Fisher's z-transformation of the correlation coefficient</u>**: Calculated from the correlation coefficient. The variance associated to the effect size is calculated from the sample size.	Provide the correlation coefficient and the sample size in the text or in the figure: *We found a positive correlation between the initial seedling size and survival one year after plantation (r = 0.68, p = 0.0002, n = 300).*