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THEMATIC SERIES

How to write a scientific paper—Writing the methods section

Como escrever um artigo científico — Estruturação e redacção da secção de métodos

L.F. Azevedo[a,*], F. Canário-Almeida[a], J. Almeida Fonseca[a], A. Costa-Pereira[a], J.C. Winck[b,c], V. Hespanhol[b,c]

[a] *Department of Health Information and Decision Sciences — HINDS (CIDES), Faculdade de Medicina, Universidade do Por Centro de Investigação em Tecnologias e Sistemas de Informação em Saúde, CINTESIS, Porto, Portugal*
[b] *Department of Medicine, Faculdade de Medicina, Universidade do Porto, Hospital de São João, Porto, Portugal*
[c] *Pulmonology Service, Hospital de São João, Porto, Portugal*

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Introduction

The methods section is the most important part of a scientific paper because it provides the essential information that allows the reader to judge the validity of the results and conclusions of the study reported. Therefore, in this section the authors should provide a clear and precise description of how the study was performed and the rationale for the methodological choices and characteristics of the study design. This section should be written in a clear and concise manner, but should always present enough information so that: (1) the study could be replicated by other researchers, in order to evaluate the reproducibility of results (it should not be a step-by-step tutorial but should be a systematic and complete description of what was done), and (2) the readers are able to judge the validity of results and conclusions presented. This will typically be the first section to be written in a paper (although many times the last to be finalized after corrections and reviews of authors and reviewers), mainly because it should be already thought of and written as a part of the research protocol/proposal, prepared at the initial phase of the research work, and because it sets the stage for the results and conclusions presented in a paper. From a journalistic point of view this section should answ tions like ''who'', ''what'', ''where'', ''when'', ''w ''how''; and should do it having into account the balance between completeness (sufficient details replication and validity verification) and brevity (th sibility of describing every technical detail and to strictly follow the guidelines/instructions for provided by journals and recommendations regard count limits). In this article, we describe and disc general recommendations that should help prepa methods section of our manuscripts; and we propo eral structure and recommended content for this Because this section is so intimately related to the tions of science, the scientific method and the stud we begin by reviewing some general concepts and p and then follow with the presentation of a propos structure and content.

Basics of the scientific method and stud design

Although many authors and schools of thought hav ent definitions and understandings regarding this r is fair and generally consensual to say that science tematic endeavor aiming at the acquisition, deve and updating of knowledge; and knowledge could b as a set of models that aim to describe, understand,

* Corresponding author.
E-mail address: lazevedo@med.up.pt (L.F. Azevedo).

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control and eventually modify the real world.[1–4] The practice of science, generically named scientific research, aims to define questions and find answers that may contribute to the building of knowledge using the so called scientific method. The scientific method, particularly in the natural sciences, mainly relies on the empirical observation of the world, as objective and unbiased as possible, and the adequate use of processes as deduction, induction[1–3] and abduction,[4] in order to generate and test hypothesis and subsequently models that allow us to describe, explain and modify the real world. In general, scientific research questions look for the estimation of population parameters or the confirmation or discovery of relationships, eventually of a causal nature, among objects (physical matter, processes, interventions or concepts). In a quantitative research paradigm the objects are regarded as variables that researchers may measure and/or control, and a variable is simply defined as a characteristic that may vary among the subjects or units of observation under study. When estimating parameters or assessing relationships, that quantitatively translate the answer to the research question, researchers are interested in minimizing random errors and systematic errors. Random errors are associated with usual sources of variability, generally measurement and sampling variability, that may affect the estimation[5,6]; and they directly affect the precision of the parameter estimates presented. Systematic errors or bias are associated with phenomena that may affect the validity of the estimation and conclusions.[5–9] Validity simply refers to the ability that a method or a study has to measure or estimate what it really intends to measure or estimate. Thus, validity refers to the credibility of the study design and results and the degree to which these results can be applied to the general population of interest.[5,6] Internal validity refers to the credibility of the study itself and is determined by the degree to which study conclusions correctly depict the truth in the study.[5,6] External validity refers to whether the results of a study can be generalized to a larger population of interest.[5,6]

Random errors are controlled or dealt with mainly by an adequate choice of measurement methods and instruments, an adequate choice of sampling methods and sample size and an adequate use of statistical methods for data analysis and presentation.[5]

Systematic errors or bias are the main target when selecting a study design and defining the methodological characteristics of the study. The methods and strategies to control and minimize systematic errors are the main factors affecting the validity of the study results and conclusions.[5,7–11] There are many different sources of systematic errors that should be considered when designing, implementing and reporting a research study and many authors have presented different proposals for their definition and systematization.[9–11] It is classical though, particularly in the field of clinical or epidemiological research, to classify systematic errors or bias in three main categories[5,6,9]: selection bias, information bias and confounding.

1. Selection bias refers to systematic errors associated with the selection of study participants or units of observation.[5,6,12]
2. Information bias refers to systematic errors asso with the measurement or classification of study va (typically classified into three main groups: dep variables — outcome or response variables; in dent variables — predictive, exposure or interv variables and confounding variables — confounc extraneous factors) and the methods and instru used for that purpose.[5,6]
3. Confounding refers to a phenomenon where variables (confounding variables) that are asso simultaneously with the outcome and predictors study interfere with the valid estimation of th predictor's effect on the outcome.[5–8] Confounc of course, of particular importance in causal res Sadly, the real world is much more complex th would like, so simple, unambiguous, direct relatio between objects can be difficult to ascertain. T causal research, the validity of a study is judged degree to which its outcomes can be attributed to ulation of independent variables and not to the of confounding variables. It is important to emp that confounding variables are hardly ever full trolled; and in many instances the influence of variables is not fully appreciated by researchers. fore, the study design must be defined so as to con many extraneous factors as possible, so that any tial cause-and-effect relationship between two can be judged validly.

The study design is the overall plan for addressi aims or purpose of the study and answering the re questions or testing the study hypotheses.[5] It ge defines the way researchers should look at the world seeking empirical evidence regarding the research qu in order to avoid fallacies and systematic errors associated with the unstructured or unscientific em observation of the world. The selection of a research should be driven first by the research purpose (que and second by feasibility issues. Questions to conside selecting a study design include[5]:

1. How much do we known about the topic under st
2. Will there be an intervention? Will all subjects ge we control who gets the intervention? Is it feas randomly assign subjects to the intervention?
3. How often and when will data be collected from su
4. How can factors that may potentially interfere relationship between predictors and outcomes b mized or controlled?

The answer to these questions comprises the justif for the study design selected and should be always suc explained. The different study designs and methodo characteristics will affect the validity of the study r Thus, although a more thorough description of the types of study designs is beyond the focus of this art is very important that researchers know the basics reg study design and are able to adequately describe it.[5]

In conclusion, the choice of the most appropriate design and the adequate planning and implementa the research methods are the foundations of good re work; and their main purpose is exactly to minimize r

and systematic errors that may affect the answer to the research question. Thus, the methods section in a paper should essentially report in a concise but complete manner how well random and systematic errors were considered and controlled by researchers, so that the validity and precision of the estimates that quantitatively translate the answer to the research question may be judged by the readers.

Structure and content of the methods section

In most journals the ''Methods'' section is designated as ''Materials and Methods'' or ''Participants and Methods'' emphasizing the two main areas that should be addressed. First, ''Materials'' refers to what was observed (e.g.: humans, animals, tissues, cells, etc.) and the interventions (e.g.: drugs, devices, etc.) and instruments (e.g.: measurement technologies) used in the study. Second, ''Methods'' refers to how subjects or objects were selected, manipulated or observed to answer the research question, how measurements were performed and how the data were analyzed.[13–15]

The writing of the Methods section should be clear and orderly to avoid confusion and ambiguity. The methods section should ideally be structured in a set of subsections describing its main content.[13–15] A possible structure is proposed along this paper including the following subsections[13–15]:

1. Study design;
2. Selection of participants — selection criteria and selection methods;
3. Data collection — variables, methods and instruments and
4. Data analysis.

Each one of these subsections could have additional subheadings as appropriate. It should be stressed that the proposal that follows is deemed to be broad and general in scope, and should always be completed with some other specific indications in the context of the particular type of study reported. To master the writing of the methods section it is important (1) to look at many other examples of methods sections in articles with similar scopes and aims as ours and (2) to use some of the many reporting guidelines that are available for the most common study types[16,17] (e.g.: CONSORT for clinical trials[18]; STROBE for observational studies[19]; STARD for diagnostic research[20]; PRISMA for systematic reviews and meta-analysis[21]; etc.).

The writing of Methods section should be direct, precise and in the past tense. Complex sentence structures should be avoided, as well as descriptions of unimportant aspects or too much details. In general the description of procedures and measurements should be organized chronologically; and, in each subsection, content should be organized from the most to the least important.[13–15]

Study design

Typically the Methods section begins with a general paragraph describing the study design and the main methodological characteristics of the study, establishing the setting for the description of participants selection and dat tion. In the context of clinical and epidemiological r the classical classifications and characteristics r quently considered when describing and systemat study design are[5,6]:

1. The definition of the descriptive vs. analytical r the study. Descriptive studies aim to describe po parameters or associations (hypothesis generat ies) and analytical studies try to answer causal q (hypothesis testing studies).
2. Reporting the comparative vs. non-comparativ of the study (is there a group comparison?).
3. Reporting the interventional vs. non-interv nature of the study (is there an intervention to uated?).
4. Reporting the existence of control over the tions or factors under study and the exist randomization. These two criteria allow the c tion of studies into three main groups: exper quasi-experimental and observational studies. imental studies the researchers have direct over the interventions or factors under st allocate them to the subjects using a rand cess — randomization (e.g.: randomized c trials). In quasi-experimental studies research trol the interventions or factors under study do not implement randomization procedure non-randomized clinical trials). In observatio ies researchers are unable to directly con interventions or factors under study and do no ment randomization procedures (e.g.: cohort case—control studies, etc.).
5. Reporting the type of randomization procedur those are implemented (e.g.: parallel groups v over, balanced vs. unbalanced groups, comp incomplete designs, factorial designs, etc.).
6. Reporting, in observational studies, if the pa selection was based on the predictor variable studies) or the outcomes (case—control studie assessment.
7. Reporting the cross-sectional vs. longitudinal r the study (having into account the existenc assumed or factual follow-up period).
8. Reporting the prospective vs. retrospective r the study (having into account the point in tim the predictors are measured in relation to the o or the point in time where recruitment of par starts).

The different study designs and methodologica teristics will affect the validity of the study resul although a more thorough description of the vario of study designs is beyond the focus of this article, important that researchers know the basics regard design and are able to adequately describe it.[5,6]

Ethical considerations

A clear presentation of the ethical considerations i tory in all animal or human studies. Although it

be a subsection by itself, as an alternative it could be a part of the subsection ''Selection of participants'', this presentation is important and should take into account the international guidelines on good clinical and research practices.[22–25] In general, references regarding the informed consent obtained on human subjects and the approval of the research protocol by an ethical committee or an institutional review board should be presented. In Portugal, submission of the research protocol to the national data protection agency (Comissão Nacional de Protecção de Dados — CNPD) could also be necessary.[26] In the case of experimental studies on human subjects (clinical trials) the approval by a national ethical committee is legally mandatory.[27] If the study raises any additional specific ethical concern this should be adequately described (e.g.: studies on especially vulnerable subgroups).[28] Although not directly linked with the ethical considerations, it is important to stress that for experimental studies on human subjects (clinical trials) authors should give an appropriate reference to the registration of the study protocol on a clinical trials registration database.[29] Most journals today only accept for publication clinical trials previously registered.

Selection of participants — selection criteria and selection methods

The participants selected for inclusion in a study and the methods of selection will ultimately determine the limits that are placed on the generalizations that can be made regarding the study results. Judging the external validity of a study (i.e.: assessing to whom the study results may be applied) requires that a comprehensive description of the selection criteria and selection methods and descriptive data regarding the study sample be provided.[5]

This subsection could begin with a brief presentation of the study setting, in order to contextualize the study presented. This should include the setting, location(s) and relevant dates of the study, indicating, for example, periods of recruitment, exposures or interventions, follow-up and data collection.

After presentation of the study setting, there are three major topics that should be addressed in this subsection[5]:

1. Specification criteria (selection criteria),
2. Methods for selection of participants (sampling) and
3. Recruitment process.

First, a full and thorough description of the criteria for selection of participants — inclusion and exclusion criteria — should be presented and its rationale explained.[5] Authors should clearly indicate the target population and the accessible population in the study.

A general description of the characteristics of participants is also important and could also be added, but this is a matter of discussion because many authors and journals believe this should be a part of the results section. For human subjects it is important to describe general demographic and clinical characteristics. For animal subjects it is important to adequately describe the species, weight, strain, sex, age and eventually details regarding special characteristics or living conditions.

In studies involving animal models or mechanica
els, a detailed description must be provided regard
preparations made prior to beginning the experiment
tocol. In addition, all aspects of animal or tissue prep
required prior to initiation of the research protocol n
described in detail. With any animal preparation or m
ical model there must be enough detail provided so t
reader can duplicate it or evaluate its relevance.

Second, the methods for selection of participants
be carefully explained. This should include an acc
how the subjects were identified and how they wer
pled from the target population under study (sa
methods).[5] When selecting subjects from a target p
tion, probabilistic sampling methods (random sampl
preferred because they more appropriately guarante
resentativeness of the sample. When reporting proba
sampling methods authors should describe the sa
frame, the instruments used for the random selecti
cess and, if appropriate, the use of complex sa
methods with stratification or clustering and weighti
cedures. Although probabilistic methods are preferr
practical reasons, non-probabilistic sampling method
random samples) are much more common (e.g.: conse
samples, convenience samples, systematic samples,
Although non-random sampling methods do not gua
the representativeness of the sample, they do not ne
ily prevent us from validly answering the research qu
It should be stressed that non-probabilistic sampling
ods are in many instances appropriate. For example, r
randomized clinical trials do not select participants t
a random sampling process, and they still are able to
priately answer the causal question regarding effic
therapeutic interventions, relying on the random allo
of alternative interventions (randomization), even wh
participants selection was non-random. When ans
causal questions the crucial point is to be able to
ate comparable study groups and make fair comp
(equipoise) between groups and, at least in this cas
resentativeness of the sample, although also import
regarded as secondary.

The third topic to be addressed in the ''Selection
ticipants'' subsection is the recruitment process.[5] A
should describe in detail how recruitment was unde
and particularly how effective it was. They should p
a complete account of the subjects selected from th
pling frame, those that accepted and those that refu
participate, ideally with a summary of reasons for
and a brief characterization of the subjects refusing
ticipate. Methods implemented to reduce refusal rate
also be described.

In addition to the three main topics described ab
comparative studies it is also important to describe
particular methods of group allocation and/or part
selection that aim to improve their comparability. In
imental studies (randomized controlled trials) a th
account of randomization procedures should be pre
including[18]: methods used to generate the random
tion sequence, details on any restrictions to random
(stratification or blocking), methods for allocatio
cealment and implementation details of the random
process. In this type of studies this is often an in
dent subsection of the methods section. In observ

studies (e.g.: cohort studies, case—control studies, etc.) authors should describe and give details regarding the implementation of methods such as stratification and matching, whenever those are used.[19]

Finally, in longitudinal studies a full description should be presented of the follow-up procedures implemented, often as a separate subsection. This should include a description of the completeness and quality of participants follow-up (number and reasons for losses of follow-up, drop-outs, drop-ins, etc.) and, in comparative studies, methods implemented to guarantee equality of follow-up conditions, for example, blinding of researchers or healthcare professionals responsible for the follow-up and the adequate control of co-interventions.[5]

Data collection — variables, methods and instruments

The next step in the methods section is to describe the data collection process, including the variables measured and the methods and instruments used for their measurement. In a quantitative research paradigm the adequate and unbiased empirical observation and measurement of variables is the cornerstone of the scientific method; thus this subsection deserves careful and thorough consideration.

Variables are observable objects that are measured, manipulated, or controlled in a study. Variables can be concrete concepts, such as height, weight, and blood pressure, or abstract concepts, such as stress, coping or quality-of-life. Variables should be operationally defined by indicating how the variable will be observed and measured in the study. Abstract variables (constructs), such as quality-of-life or stress, should be defined both conceptually and operationally. The conceptual definition explains the theoretical meaning of the variable, while the operational definition specifies how it will be measured. For example, when measuring quality-of-life, researchers could present a brief conceptual definition of the construct, but should always add details regarding its operational definition, by indicating the model and instrument applied to measure quality-of-life, for example, by using the SF-36 health questionnaire.

In general, the variables in a study could be classified in one of four major groups[5]:

1. Predictor (independent, exposure or intervention) variables,
2. Outcome (dependent) variables,
3. Confounding (extraneous) variables or
4. Interaction (effect modifier) variables.

When describing the variables in a study the authors do not need to give a full and complete description of all variables measured, however the main predictors and all outcome variables should be described with sufficient detail as to allow replication and assessment of the quality of the measurement or classification. For these variables a full account of their conceptual definition, operational definition, classification or diagnostic criteria applied (if appropriate), methods of measurement, instruments used and a brief description of the evidence regarding their validity and reproducibility should be present detailed presentation should be extended to a variable of particular importance for the study uncommon measurement procedures or instrumen

For those variables where it is deemed neces description of the measurement methods and ins should include the manufacturer and model, calibra cedures, evidence regarding the validity and repro of instruments and how measurements were ma instruments used to measure variables must be and valid. Validity is the extent to which an in measures what it reports to measure. Reliabili to the consistency with which an instrument r a study variable. Internal consistency (e.g.: Cr alpha), test—retest reliability, and inter-rater relia examples of methods used to assess the reliabil instrument, particularly in the context of abstract (constructs) measurement. These psychometric or ric properties of instruments determine the over validity. It is important to select and describe ins that have established reliability and validity in the tion that the investigator plans to study (e.g.: older children) and use instruments that are properly tra adapted and validated for the study population. not formally prohibited, the use of instruments t not previously submitted to an adequate translatio tation and validation process impose important li to the credibility and validity of the study results ar should be always indicated.

Finally, particular methods to control bias as with the measurement or classification of stu ables should be described. For example, implemen blinding procedures for participants and for researc lecting data (especially outcomes measurement) s indicated and explained.

Data analysis

In the last part of the methods section author describe with sufficient detail the statistical meth for the study data analysis, including descriptive and methods for statistical inference.[5] This pres should have a close link to the aims of the study an precisely establish what will be presented in the re tion.

This subsection should include an initial gene tence regarding the descriptive statistics used, ha account the main types of variables analyzed (e.g or medians, standard deviations or quantile ranges, frequencies and proportions, etc.). Next, a brief tion of inferential methods used should follow, inclu indication of confidence intervals calculated, an ac the statistical hypothesis tests applied and the indi any uni- or multi-variable regression or modeling pr employed. A special note should be added regardin of confidence intervals as the best method to exp precision of parameter estimates presented in a stu presentation is increasingly deemed essential and more informative than the classical *p*-values of hy testing.

Some additional aspects regarding the statistical analysis should be addressed whenever it is deemed necessary, taking into account the main study aims:

1. Describing the specifics regarding methods used to account for confounding in observational studies (e.g.: multi-variable regression methods for effect measures adjustment, propensity scores,[30] causality modeling using directed acyclic graphs and structural models,[31–33] etc.).
2. Describing methods to examine subgroups, interactions and effect modification in experimental and observational studies.[34]
3. Describing any interim analysis, stopping rules and adjustments that may be used, particularly in experimental studies.[18]
4. Describing any particular adjustments made taking into account the sampling methods and weighting procedures used.
5. Describing methods used to account for missing data.
6. Describing methods used for sensitivity analysis.

Also important in this section is to describe the estimates and explanation of methods for the sample size and power determination.[5] The determination of the sample size before the beginning of the study is crucial to ensure the appropriate power of hypothesis testing and the precision of parameter estimates. In many instances, particularly in observational studies, a formal sample size calculation is not possible for practical reasons (for example, the study sample is assembled retrospectively or is already fixed before the beginning of the study). Even in these situations it is advisable to present results of a formal power analysis, in order to give an indication of the power of hypothesis tests and the magnitude of differences that researchers are able to detect in those settings. Some authors prefer to incorporate the paragraph regarding sample size determination as a part of the ''Selection of participants'' subsection.

Finally, an indication of the level of type I errors (alpha level) assumed in all statistical hypothesis testing (usually, a 5% alpha level is assumed) and an indication of the statistical software package used for analysis (with a reference) should be presented in this subsection.

Conclusion

The methods section is the most important part of a scientific paper because it provides the crucial information that allows the reader to judge the validity of the results and conclusions of the study reported. Therefore, in this section, the authors should provide a clear and precise description of how the study was performed and the rationale for the methodological choices and characteristics of the study design. A clear and precise account of how a study was performed, and the rationale for specific study methods are the crucial aspects of scientific writing. A proposal for the structure and content of the methods has been presented and explored giving a general guidance for the writing and assessment of the quality of this section and of the study reported. We hope that somehow this paper may comprise a useful tool for authors, reviewers and readers of scientific papers, and in particular those of the *Portuguese Jou Pulmonology (Revista Portuguesa de Pneumologia)*.

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