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Education

How to write a materials and methods section of a scientific article?

Fikret Erdemir

ABSTRACT

In contrast to past centuries, scientific researchers have been currently conducted systematically in all countries as part of an education strategy. As a consequence, scientists have published thousands of reports. Writing an effective article is generally a significant problem for researchers. All parts of an article, specifically the abstract, material and methods, results, discussion and references sections should contain certain features that should always be considered before sending a manuscript to a journal for publication. It is generally known that the material and methods section is a relatively easy section of an article to write. Therefore, it is often a good idea to begin by writing the materials and methods section, which is also a crucial part of an article. Because "reproducible results" are very important in science, a detailed account of the study should be given in this section. If the authors provide sufficient detail, other scientists can repeat their experiments to verify their findings. It is generally recommended that the materials and methods should be written in the past tense, either in active or passive voice. In this section, ethical approval, study dates, number of subjects, groups, evaluation criteria, exclusion criteria and statistical methods should be described sequentially. It should be noted that a well-written materials and methods section markedly enhances the chances of an article being published.

Key words: Article; material; methods; publication.

How to Write a Materials and Methods Section of a Scientific Article?

Up to the 18th Century scientific researches were performed on a voluntary basis by certain scientists. However from the second half of the 19th century, scientific development has gained momentum with the contributions of numerous scientists including Edison, Fleming, and Koch. In parallel with these developments, apparently each scientific field, and even their branches made, and still making magnificent progressions from the end of the 18th century. Secondary to these developments, scientific researches have been implemented systematically by universities, and various institutions in every part of the world as an integral component of national strategies. Naturally, the number of researchers who performed scientific investigations or sponsored by various institutions increased considerably. Also, as is known very well, all over the world scientists, and researchers move from one place to another to disseminate scientific knowledge. All of these scientific efforts, and activities reflect on clinical practice, and hundreds of thousands, and millions scientific articles which we can currently gain access into all of them online. As indicated by the investigator

Gerard Piel, "Without publication, science is dead" which explains the importance of publication. In other words, if you don't share your investigation and knowledge, they don't mean anything by themselves. Although sharing the knowledge is essential for writing a scientific paper, nowadays writing a scientific article is mostly learnt as a master-apprentice relationship, and therefore certain standards have not been established. This phenomenon creates serious stress especially for young investigators in their early stage of writing scientific papers. Indeed investigators receiving their residency training confront this reality finally during writing of their dissertations. Though sharing knowledge is known as a fundamental principle in writing a scientific paper, it creates difficulties in the whole world. Relevant to this issue, in the whole world investigations have been performed, and books have been written on the subject of how to write a scientific paper. Accordingly, in our country mostly local meetings, and courses have been organized. These organizations, and investigations should be performed. Indeed, nowadays, in the first assessments, the rejection rate of the journals by internationally acknowledged scientific indexes as "Science Citation Index (SCI)" and

Department of Urology, Faculty of Medicine, Gaziosmanpaşa University, Tokat, Turkey

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Correspondence:

Fikret Erdemir Yeşilırmak Mah. Bosna Cad. 3. Sok.no: 7, Mollaoğulları Apt. K: 2, D: 360100, Merkez 60100 Tokat, Turkey Phone: +90 356 213 38 44 E-mail: fikreterdemir@mynet.com

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"Science Citation İndex Expanded (SCI-extended" which have certain scientific standards, increases to 62 percent. As a matter of fact only 25% of Class A journals have been included in the lists of SCI, and SCI-extended.

As we all know very well, scientific articles consist of sections of summary, introduction, material, and methods, discussion, and references. Among them, conventionally Materials and Methods section has been reported as the most easily written or will be written section. Although it is known as the most easily written section, nearly 30% of the reasons for rejection are related to this section per se. Therefore due care, and attention should be given to the writing of this section. In the writing process of the 'Material and Methods' section, all achievements performed throughout the study period should be dealt with in consideration of certain criteria in a specific sequence. Since as a globally anticipated viewpoint, 'Materials and Methods' section can be written quite easily, it has been indicated that if difficulties are encountered in writing a manuscript, then one should start writing from this section. In writing this section, study design describing the type of the article, study subjects to be investigated, methods, and procedures of measurements should be provided under four main headings.[1,2] Accordingly, in brief, we can emphasize the importance of providing clearcut, adequate, and detailed information in the 'Materials and Methods' section to the scientists who will read this scientific article. Meeting these criteria carries great importance with respect to the evaluation of reliability of the investigation by the readers, and reviewers, and also informing them about procedural method, design, data collection, and assessment methods of the investigation, Priorly, as is the case in all scientific investigations, one should be reminded about the importance, and indispensability of compliance with certain standard writing rules. Accordingly, rules of grammar should be obeyed, and if possible passive voice of simple past tense should be used. Related to these rules, use of verbs 'investigated', 'evaluated' or 'performed' will be appropriate. Recently, expressions showing the ownership of the investigation as 'we performed', 'we evaluated', 'we implemented' have taken priority. Since the important point is communication of the message contained in the scientific study, the message should be clearly comprehensible. While ensuring clarity of the message, use of flourishing, and irrelevant sentences should be avoided.[1,3] According to another approach, since our article will be read by professionals of other disciplines, it is important to comply with certain rules of writing. To that end, standard units of measurements, and international abbreviations should be used. Abbreviations should be explained within parentheses at their first mention in the manuscript. For instance let's analyze the following sentence" The patients were evaluated with detailed medical history, physical examination, complete urinalysis, PSA, and urinary system ultrasound" The abbreviation PSA is very well

known by the urologist. However we shouldn't forget that this article will be read by the professionals in other medical disciplines. Similarly this sentence should not be written as: "The patients were evaluated with detailed medical history, physical examination, complete urinalysis PSA (prostate-specific antigen), and urinary system ultrasound." Indeed the abbreviation should follow the explanation of this abbreviation. Then the appropriate expression of the sentence should be. "The patients were evaluated with detailed medical history, physical examination, complete urinalysis, prostate-specific antigen (PSA), and urinary system ultrasound."

In addition to the abovementioned information, in the beginning paragraphs of 'Materials and Methods' section of a clinical study the answers to the following questions should be absolutely provided:

- 1. The beginning, and termination dates of the study period.
- 2. Number of subjects/patients/experimental animals etc. enrolled in the study,
- 3. Has the approval of the ethics committee been obtained?
- 4. Study design (prospective, retrospective or other). [1,2,4-7]

Still additional features of the study design (cross-sectional) should be indicated. Apart from this, other types of study designs (randomized, double-blind, placebo-controlled or double-blind, parallel control etc.) should be revealed.

The heading of the section "Materials and Methods" can be changed to "Patients and the Method" in accordance with writing rules of the journal in question. Indication of starting, and termination dates of a clinical study will facilitate scientific interpretation of the article. Accordingly, outcomes obtained during development phase of a newly implemented method might be considered differently from those acquired during conventional use of this method. Besides, incidence of the diseases, and number of affected people might vary under the impact of social fluctuations, and environmental factors. Therefore with this justification study period should be specified. Number of cases included in the study should be absolutely indicated in the 'Materials and Methods' section. It will be appropriate to determine study population after consultation to a statistician -and if required- following "power analysis" Accordingly, the need for a control group will be indicated based on the study design. Nowadays, as a requirement of patient rights, obtainment of approval from ethics committee should be indicated with its registration number. In addition, acquirement of informed consent forms from patients should be indicated. Ethics Committee approval should be obtained in prospective studies performed with study drugs. Otherwise in case of occurrence of adverse effects, it should be acknowledged that in compliance with Article #90 of the Turkish Criminal Law, a 3-year prison sentence is given to the guilty parties.[8] Since issues related to the

Ethics Committee are the subject of another manuscript, they won't be handled herein.

The following paragraph exemplifies clearly the aforementioned arguments: "After approval of the local ethics committee (BADK-22), informed consent forms from the patients were obtained, and a total of 176 cases with lower urinary tract symptoms (LUTS) were retrospectively evaluated between January 2011, and December 2012." In a prospectively designed study, methods used to communicate with the cases including face-to-face interviews, phone calls and/or e-mail should be indicated.^[1,2] Each paragraph or subheading in the 'Materials and Methods' section should be in accordance with the related ones in the 'Results' section. In other words, the sequence of paragraphs, and subheadings in the 'Results' section should be the same in the 'Materials and Methods' section.

As a next step, names of the groups, and distribution of the cases in these groups should be indicated. For instance: the statement "Cases were divided into 3 groups based on their LUTS scores as. Groups 1 (0-9; n=91), 2 (10-18; n=66), and 3 (\geq 19; n=20)" clearly delineates the scope of the study at baseline.. In the 'Materials and Methods' section the number of study subjects should be absolutely documented. Herein, after assignment of names to groups, in the rest of the manuscript, these names should be used. For example instead of saying: "Mean ages of the cases with LUTS scores between 0-9, 10-18, and ≥19 were determined to be 63.2±2.1, 62.8±4.5, and 65.7±3.9 years, respectively" it will be more comprehensible to use the expression: "Mean ages of the Groups 1, 2, and 3 were specified as 63.2±2.1,62.8±4.5, and 65.7±3.9 years." (p=0.478). Expressions indicated in the 'Materials and Methods' section should not be repeated in the "Results" section. Thus, errors of repetition will be precluded. Following the abovementioned information, the evaluation method of the cases enrolled in the study should be indicated. Hence, results of medical history, physical examination, and if performed laboratory or radiological evaluations -in that order-should be indicated. The application of survey studyif any-should be investigated, and documented. Therefore, the following sentences encompass all the information stated above: "The cases were evaluated with detailed medical history, physical examination, measurements of serum follicle stimulating hormone (FSH), luteinizing hormone (LH), testosterone (T) levels, complete urinalysis, urinary flow rate, direct urinary system roentgenograms, urinary system ultrasound, and if required cyctoscopy. Lower urinary system complaints, and erectile dysfunction were evaluated using International Prostate Symptom Score (IPSS), and International Erectile Function Scale (IIEF), respectively." Apparently, questionnaire forms were used in the above-cited study. However, methods used for the evaluation of questionnaire forms, and significance of the results obtained, and if possible, the first performer of this sur-

vey should be written with accompanying references. In relation to the abovementioned questionnaires the following statements constitute standard expressions for the 'Materials and Methods' section: "International Prostate Symptom Score (IPPS) was used in the determination of the severity of prostatic symptoms. IPSS used to determine the severity of the disease, evaluate treatment response, and ascertain the symptomatic progression, is the most optimal scoring system recommended by European Association of Urology (EAU) which classifies the severity of the disease based on IPSS scores as mild (0-7), moderate (8-19), and severe symptomatic (20-35) disease. In the evaluation of sexual function International Erectile Function Scale (IIEF) was used. IIEF is one of the most prevalently used form for the patients who consulted for the complaints of sexual dysfunction Based on IIEF scores, the severity of the disease was classified as severe (1-10), moderate (11-16), mild to moderate (17-21), mild (22-25), and no ED (26-30)."

Whether the institutions of the authors working for should be written in the 'Materials and Methods' section can be a subject of debate, generally viewpoints favour provision of this information. However, in compliance with their writing rules, some journals do not favour open-label studies where name of the study site is indicated, and this principle is communicated to the author during editorial evaluation Besides, in the 'Materials and Methods' section, the brand of the study object, and its country of origin should be indicated. (ie. if radiological methods are used, then the brand of radiological equipment, and its manufacturing country should be specified. In a study entitled 'The Impact of Computed Tomography in the Prediction of Post-Radical Nephrectomy Stage in Renal Tumours' since the main topic of the study is computed tomography, the specifications of the equipment used should be explicitely indicated. On the other hand, the details of the medical method which can effect the outcomes of the study should be also recorded. Accordingly, the methods applied for percutaneous nephrolithotomy, ureterorenoscopy, varicocelectomy, transurethral prostatectomy, radical prostatectomy (perineal, open, laparoscopic or robotic should be absolutely indicated. Then inclusion, and exclusion criteria, and if used control group, and its characteristics should be documented. Thus the following paragraph about exclusion criteria will be appropriate: Patients with a history of neurogenic bladder, prostatic or abdominal operation, and transrectal ultrasound guided prostate biopsy (within the previous 6 months), those aged <40 or >70 years, individuals with a peak urine flow rate below 10 ml/sec, and residual urine more than 150 cc were not included in the study."[1-3,9]

Some diseases mentioned in the "Materials and Methods" section require special monitorization procedures. In these cases the procedure of monitorization should be documented for the sake of the validity of the study in question. Accordingly, in

conditions such as "nephrectomy, prostatectomy, orchidectomy, pyeloplasty, varicocelectomy, drug therapies, penile prosthesis, and urethral stricture" clinical follow-up protocols should be provided.

The abovementioned rules, and recommendations are most frequently valid for a clinical study, and some points indicated in experimental studies should be also considered. Types, weights, gender, and number of the animals used in animal studies should be absolutely specified. Besides condition of evaluation of experimental animals should be noted. Then as is the case with clinical studies, approval of the ethics committee should be obtained, and documented. Accordingly, the beginning paragraphs of the 'Materials and Methods' can be expressed as follows:

"In the study, 40 Wistar-Albino 6-month-old rats each weighing 350-400 g were used. After approval of the ethics committee (HADYEK-41) the study was performed within the frame of rules specified by the National Institute for animal experiments. The rats were divided into 3 groups. Hence, Group 1 (n=7) was accepted as the control group. The rats subjected to partial ureteral obstruction with or without oral carvedilol therapy at daily doses of 2 mg/kg maintained for 7 days constituted Groups 3 (n=8), and 2 (n=8), respectively. Each group of 4 rats was housed in standard cages with an area of 40x60 cm. The animals were fed with standard 8 mm food pellets, and fresh daily tap water. The rats were kept in the cages under 12 hours of light, and 12 hours of dark. Ambient temperature, and humidity were set at 22±2°C, and 50±10%, respectively."

Herein, the method, and agent of anesthesia used (local or general anesthesia) in surgical procedures, and then the experimental method applied should be clearly indicated. For example the following sentences explain our abovementioned argu-



Figure 1. Partial ureteral obstruction model[11]

ments; "All surgical procedures were performed under xylazine -ketamine anesthesia. In all groups, ureters were approached through midline abdominal incision. In Group 1, ureters were manipulated without causing obstruction. Results of biochemical, and pathological evaluations performed in Group 1 were considered as baseline values."

"Through a midline abdominal incision partial ureteral obstruction was achieved by embedding two-thirds of the distal part of the left ureter into psoas muscle using 4/0 silk sutures as described formerly by Wen et al. [10] (Figure 1). [11] All rats were subjected to left nephrectomies at the end of the experimental study." As formulated by the above paragraph, if the method used is not widely utilized, then the first researcher who describes the method should be indicated with relevant references. One or more than one figures with a good resolution, and easily comprehensible legends should be also included in the explanation of the experimental model. For very prevalently used experimental models as torsion models cited in the "Materials and Methods" section, there is no need to include figures in the manuscript.

Appropriate signs, and marks placed on the figure will facilitate comprehension of the legends (Figure 2).

The signs used will also improve intelligibility of the target. The figures should be indicated within parentheses in their first mention in the "Materials and Methods" section. Headings and as a prevalent convention legends of the figures should be indicated at the end of the manuscript.

If a different method is used in the study, this should be explained in detail. For instance, in a study where the effect of smoking on testes was investigated, the method, and the applicator used to expose rats to cigarette smoke should be

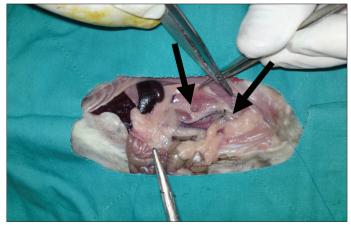


Figure 2. Ureteral segments (black arrows) seen in a rat partial ureteral obstruction model^[11]

indicated in the 'Methods' section following classical description. Relevant to the study in question, the following paragraph explaining the study method should be written: "A glass chamber with dimensions of 75 x 50 x 50 cm was prepared, and divided into 4 compartments with wire fences. The rats in the 2., and 4. cages were placed in these compartments. Each compartment contained 4 rats. Cigarette smoke was produced using one cigarette per hour, and smoke coming from the tip, and the filter of the lighted cigarette was pumped into the gas chamber with a pneumatic motor. The rats were exposed to smoke of 6 cigarettes for 6 hours. The compartments of the rats were changed every day so as to achieve balanced exposure of the rats to cigarette smoke." [12]

Meanwhile, chemical names, doses, and routes of administration of the substances used in experimental studies should be indicated. If the substance used is a solution or an antibody, then manufacturing firm, and its country should be indicated in parenthesis. This approach can be exemplified as "Animals used in experiments were randomized into 4 groups of 8 animals. Each group was housed in 2 cages each containing 4 animals. The first group did not undergo any additional procedure (Group 1). The second group was exposed to cigarette smoke (Group 2). The third (Group 3), and the fourth (Group 4) groups received daily intraperitoneal injectable doses of 10 mg/kg resveratrol (Sigma-Aldrich, St. Louis, MO, USA). The Group 4 was also exposed to cigarette smoke.[12]

After all of these procedures, method, and analytical procedure of histopathological examination used should be described -if possible- by a pathologist Similarly, biochemical method used should be referenced, and written by the department of clinical chemistry. It can be inferred that each division should describe its own method. In other words, histopathological, microbiological, and pharmacological method should be described in detail by respective divisions.

If we summarize all the information stated above, understandably sharing of the scientific knowledge is essential. Since reproducibility of a study demonstrates the robustness of a study, with the detailed approaches indicated above, reproducibility of our study is provided, and the relevant questions of "How?", and "How much?" are answered. Besides, since 'Materials, and Methods', and 'Results' sections will constitute a meaningful whole, explanations of all information related to the data mentioned in the 'Results' section should be provided. As an important point not to be forgotten, evaluation or measurement method used for each parameter indicated in the 'Results' section should be expounded in the "Materials and Methods" section. For example if you used an expression in the' Results' section like "median body mass index (BMI)

of the patients was 27.42 kg/m²", then you should beforehand indicate that comparative evaluation of BMIs will be done in the "Materials and Methods" section. In addition, the description, and significance of the values expressed in the "Results" section should be indicated in the "Materials and Methods" section. In other words, it should be stated that the patients were evaluated based on their BMIs as normal (18-24.9 kg/m²), overweight (25 kg/m²-40 kg/m²), and morbid obesity (>40 kg/m²). If you encounter difficulties in writing "Materials and Methods" section, also a valid approach for other sections, firstly simple headings can be written, then you can go into details. In brief, for every parameter, the reader should get clear-cut answers to the questions such as "How did they evaluate this parameter, and which criteria were used?".[1,3,13-15]

The last paragraph of the 'Materials, and Methods' section should naturally involve statistical evaluations. This section should be written by statisticians. Accordingly, the preferred statistical method, and the justifications for this preference should be indicated. In conventional statistical evaluations, provision of details is not required. In information indicated above, the statement "For statistical analysis, ANOVA test, chi-square test, T test, Kruskal-Wallis test have been used." is not required very much. Instead, more appropriate expression will be a statement indicating that recommendations of a knowledgeable, and an experienced statistician were taken into consideration or advanced statistical information was reflected on the statistical evaluations as follows: "Chi-square tests were used in intergroup comparisons of categorical variables, and categorical variables were expressed as numbers, and percentages. In comparisons between LUTS, and ED as for age, independent two samples t -test was used. In the evaluation of the factors effective on erectile dysfunction multivariate logistic regresssion test was used. P values lower than 0.05 were considered as statistically significant The calculations were performed using a statistical package program (PASW v18, SPSS Inc, Chicago, IL)." Herein, the type of statistical package used for statistical methods should be emphasized.

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