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Significance of observational studies in clinical paediatric dentistry research

ABSTRACT

In clinical paediatric dentistry research, an Observational Study (OS) is one in which the investigators do not intervene in any way, for example, providing a treatment to a group of eligible participants; the investigators also are limited to the observation of one or more groups of paediatric patients whose characteristics have been registered and analysed. This design constitutes the majority of dental research, mainly with the aim of eliciting possible causes of, or estimating the prevalence of, an oral disease. However, OS may be more challenging in terms of controlling confounding factors. There are three main observational designs: cohort studies; case-control studies, and cross-sectional studies. The aim of the present article was to provide the basics of these three designs in the paediatric dentistry clinical research field. Also, some useful examples are provided of OS carried out in paediatric dentistry and published in the dental literature.

Keywords *Paediatric Dentistry Research, Observational Studies, Cohort, Case-control, Cross-sectional*

Introduction

Evidence-Based Paediatric Dentistry principles provide paediatric dentists the opportunity of applying the most valid and relevant research findings to the oral care of their patients in the clinical setting [Sutherland, 2001; Krithikadatta, 2012]. These findings are the result of studies performed on child participants and are conducted under high-quality standards of the clinical investigation and ethical considerations. In paediatric dentistry research, it is important to select the appropriate methodological design to answer a clinical question, to test the hypothesis posed, and to achieve the study objectives properly [Krithikadatta, 2012]. An Observational Study (OS) is one in which the investigators do not interfere in any way, for example, providing a treatment to a group of eligible participants and observing the results over the time, as in Randomized and Controlled Trials (RCT) [Grimes and Schulz, 2002a]. Unlike clinical trials, in OS, researchers simply observe one or more groups of paediatric patients whose data on specific health characteristics have been registered and analysed; in other words, researchers can observe what is happening, without intervening, in one or more groups of participants, from which an inference is made about a target child population [Levin, 2005; Hackshaw et al., 2006; Introductory Statistics, 2011].

Although OS are sometimes called epidemiological studies, they are not considered high-quality designs in Evidence-Based Paediatric Dentistry; they are more useful than RCT for studying the cause-effect relationship of an specific oral disease, or for describing or monitoring clinical or epidemiological issues related with oral health in children, such as prevalence, diagnosis, prognosis, or causation [Petrie et al., 2002; Krithikadatta, 2012]. When the purpose is to measure risk factors or to collect exposure data related with a disease, OS are often more adequate in terms of practicality and ethics [Sutherland, 2001]. Also, they are less costly, can be completed more quickly, and do not require patients or providers who are willing to be randomised to treatments [Grimes and Schulz, 2002a]. However, the design may yield invalid or misleading results, particularly when external factors (confounders) or bias, which can influence outcomes, are not carefully considered during the study stages [Butani et al., 2006; Althubaiti, 2016].

The aim of the present report was to provide the fundamental concepts of OS for its application when necessary in clinical paediatric dentistry research.

Classification of OS

The type of OS depends on the rarity of the disease or condition and on issues related with human or financial resources [Sutherland, 2001; Grimes and Schulz, 2002a]. OS are classified as cross-sectional, in which participants are measured on a sole occasion and do not involve their follow-up, and longitudinal, which require the participants to be evaluated over a period of time, including their being measured more than once [Petrie et al., 2002; Krithikadatta, 2012]. According to the manner in which data are collected, longitudinal studies may be either prospective or retrospective. Prospective studies

Observational clinical studies	Forwards in time	Strength of association
Cohort study	(Exposure to outcome)	+++
Case-control study	(Outcome to exposure)	+ +
Cross-sectional study	(Exposure and outcome at the same time)	+

TABLE 1 Algorithm for the classification of observational clinical studies.

collect data forward in time, in order to examine the etiology of a disease. Retrospective studies collect past exposure information on participants through interviews or recorded information [Grimes and Schulz, 2002a; 2002; Levin, 2005; Krithikadatta, 2012]. There are three main designs for responding to different clinical/epidemiological research questions as follows: cohort studies; case-control studies, and cross-sectional studies (Table 1).

Cohort studies

In clinical paediatric dentistry research, the term "cohort" applies to a group of children who share certain characteristics (e.g., gender and age), who remain as part of a study group over a period of time [Introductory Statistics, 2011]; some examples of cohorts are young children who live in the same industrialised city, infants born in a given year, or adolescents with a specific oral health condition. Cohort studies can be considered natural experiments in which outcomes are measured in real-world rather than in experimental settings [Christensen and Langberg, 2012]. In this design, it is known since the beginning of the study whether recruited child participants have been exposed or not to a risk factor (or environmental, social, or medical modalities) – e.g., a vaccine, a drug, or an environmental toxin – suspected of being causative of a specific oral disease or condition. However, the participants have not yet developed the particular condition [Sutherland et al., 2001; Grimes and Schulz, 2002b].

Cohort studies ask the question "What will happen?". Exposed and non-exposed participants are then followed over time, even for weeks/months/years, to see how many children in each group develop the disease or the outcome of interest; then, the disease rates are compared between the exposed and non-exposed [Steel and Pearce, 2009]. In this respect, cohort studies are able to quantify the incidence of a disease or other changes in oral status –the first occurrences of the event-of-interest–, a measure of disease frequency [Grimes and Schulz, 2002b; Morgenstern and Sohn, 2010]. Thus, it is a powerful way to observe the potential etiological association of these risk factors and the disease, or other outcomes-of-interest [Sutherland et al., 2001]. These studies are also undertaken to determine the natural progression of a disease, particularly chronic diseases, over a certain time period, and to identify outcomes from different dental therapies applied to paediatric patients, in order to assess the effectiveness of these therapies [Dawson-Saunders and Trapp, 1994; Levin, 2005].

A variation of this design is the retrospective cohort study ("historical cohort"), which is performed on information collected in the past from dental/medical records [Dawson-Saunders and Trapp, 1994]. Here, the researcher accesses a historical list of all exposed and non-exposed children and then determines the current case or non-case status. The study begins when the disease-of-interest is already established in the original cohort of individuals, i.e., long after the measurement of exposure; in other words, the events being evaluated actually occurred prior to the onset of the study [Dawson-Saunders and Trapp, 1994; Grimes and Schulz, 2002b].

Cohort studies are usually more powerful than other observational designs for suggesting a cause-effect relationship, are less expensive, are easier to conduct than RCT, and are ethically are acceptable. Investigators can study multiple outcomes related with the exposure. Also, cohort studies establish whether exposure precedes disease occurrence [Grimes and Schulz, 2002b]. However, cohort studies tend to be time-consuming. An additional disadvantage is that cohorts (exposed and non-exposed) often cannot be well-matched regarding confounding factors, for example in terms of social class or home/school exposures, that may influence the study results, especially when large numbers of individuals are required [Grimes and Schulz, 2002b]. Other critical problems include that the circumstances of the child participants may change during the study, and that it may be difficult to maintain a high rate of retention of the cohort members over time due to poor response from or loss of contact with child participants and their parents (denominated attrition/withdrawal bias); thus, the investigators are unaware of those who have been lost to the study, which may represent a significant amount of information. The problem is particularly more significant when subject loss does not occur randomly. Cohort designs are not adequate for rare diseases or in cases of long latency periods between cause and effect [Grimes and Schulz, 2002b; Introductory Statistics, 2011].

Statistical aspects: in general, OS measure the strength of association between exposure/treatment and outcome; the hypothesis to be tested is whether the two events are dependent or not. There are two ways to evaluate strength-of-association: through the use of frequency measures or risk ratios, and by means of hypothesis testing, to assess how dependent the two events are. This latter method implies the calculation of p values and 95% Confidence Intervals (95% CI) for effect size [Dawson-Saunders and Trapp, 1994; Machin et al., 2007].

The most common frequency measures for an outcome

studied in a cohort design comprise relative risk and attributable risk [Grimes and Schulz, 2002b; Machin et al., 2007]. After the child cohort has been monitored, the final status for every participant is recorded as a dichotomous outcome (exposed/non-exposed and with disease/without disease); therefore, child participants may be categorised as "with disease in exposed", "without disease in exposed", "with disease in non-exposed", or "without disease in non-exposed" [Jepsen et al., 2004]; thus, a 2 × 2 contingency table is built employing the frequencies in each category (see Appendix, later). Incidence rate is calculated in the two groups, exposed children and non-exposed children; then, both incidence rates are compared by arithmetic division or ratio, called Relative Risk (RR) with its correspondent 95% CI [Dawson-Saunders and Trapp, 1994; Jepsen et al., 2004]. A RR of 1 (or its 95% CI not containing 1) means that the risk of disease in those exposed to the exposure and those non-exposed to the exposure are the same; an RR greater (or less) than 1 represents the extent to which the disease in the exposed group is increased (or decreased) relative to that of the non-exposed group [Petrie et al., 2002]. Other suitable OS measure is risk difference (or attributable risk), which provides answers to questions such as "Among smoker adolescents, what percentage of the total risk of periodontal disease is due to smoking?" The risk difference is the difference among periodontal disease incidence rates in each risk factor category (smokers vs. non-smokers) [Petrie et al., 2002; Jepsen et al., 2004].

Statistical analysis for OS also employs diverse appropriate hypothesis tests according to the type of variable measured: for categorical variables, the Chi-square, Fisher exact, the McNemar, and the Mantel-Haenzel tests are used, while Student t tests and their non-parametric alternatives are utilised for continuous data [Dawson-Saunders and Trapp, 1994; Machin et al., 2007]. The logistic regression method is useful for handling multiple risk exposures and potential confounding factors [Niven et al., 2012]. When these techniques are used, 95% CI and p values should be reported [Christensen and Langberg, 2012].

Analysis of data from cohort studies becomes more complicated when other predictors are considered that change along the follow-up period, repeated outcome measures, or recurrent outcome events in the same child, together with other types of correlated data [Grimes and Schulz, 2002b; Morgenstern and Sohn, 2010].

Examples of published articles. Here, we provide two useful examples of cohort studies in the paediatric dentistry field:

1 Nirunsittirat et al. [2016] examined the possible association between dental caries in primary teeth and adverse birth outcomes (e.g., preterm or birth at <37-week gestation, low birth weight <2,500 g, and small for gestational age (SGA), such as birth weight in the <10th percentile expected weight for gestational age). Eight hundred sixty Thai children born between January 2001 and January 2002 were followed for 3–4 years using the Decayed-Missing-Filled Surface (DMFS) index and World Health Organization (WHO) criteria. The researchers reported an estimated RR (with its 95% CI) adjusted for confounding factors. Dental caries was observed in 88.2% of children, with a mean DMFS of 0.61. The adjusted RR for caries in primary teeth was 0.61 (95% CI = 0.43–0.85) for preterm, 0.89 (95% CI = 0.67–1.21) for low birth weight, and 0.96 (95% CI = 0.74–1.26) for SGA. The authors concluded that there was an inverse association between preterm and childhood caries –in other words, the DMFS was lower in preterm than in full-term children–, while the remaining two birth variables were not associated with dental caries in primary teeth (note that both 95% CI include the value 1, corresponding to a lack of an exposure/outcome association).
2 Mexican researchers [Ruiz-Rodríguez et al., 2014] investigated the potential association between maternal Streptococcus mutans levels with those of their infants, during the period from birth to 5 months of age –before primary tooth eruption–, in order to identify risk factors for S. mutans "vertical" transmission. The cohort study comprised 60 mother-infant pairs, who provided several saliva samples on days 0, 15, 30, 90, and 150 postpartum, to detect in vitro the cariogenic microorganism. Diverse related factors were considered, such as type of feeding and number of siblings, among others. At day 150, only 3% of the studied infants exhibited S. mutans in saliva, while 90% of their mothers showed a positive detection. In conclusion, it was not possible to find a real "vertical" (mother-to-infant) transmission of S. mutans with the lack of a significant association between maternal and infant microorganism levels.

Case-control studies

Case-control studies are retrospective designs aiming to find the differences in risks for a particular oral disease between sick and health individuals; this design is particularly useful for studying uncommon pathological conditions or when a long time elapses between exposure and disease initiation [Sutherland, 2001; Machin et al., 2007; Introductory Statistics, 2011; Niven et al., 2012]. Additionally, instead of measuring risk factors, case-control studies can be utilised to determine the potential relationship between "protective" factors (e.g., a beneficial procedure such as oral restorative treatment) and a therapeutic outcome-of-interest [Steel and Pearce, 2009].

These studies start with the identification of children with the disease-in-question (denominated cases) and those who do not (termed controls), selected from a comparable population, who are often matched or paired with respect to crucial confounding or modifying factors (e.g., age and gender) in order to neutralise their influence [Steel and Pearce, 2009; Introductory Statistics,

2011; Niven et al., 2012]. Then, the researchers look back in time to determine the proportion of child cases or controls who were exposed to the suspected casual factor or exposure (e.g., by means of reviewing medical or lifestyle histories or through interviews/questionnaires) [Sutherland, 2001; Introductory Statistics, 2011; Niven et al., 2012]. To obtain reliable results, it is essential for any measurement to be carried out in exactly the same manner on cases and on controls [Shahar and Shahar, 2012].

Examples of published articles. The following are two useful examples of case-control studies in the paediatric dentistry field.

1 Italian paediatric dentistry researchers [Luzzi et al., 2013] investigated the possible association between prolonged oral respiration due to allergic rhinitis (the causal factor) and the development of malocclusions (e.g., mono- or bilateral crossbite, anterior open-bite, and increased overjet), and the outcome, in the primary and early-mixed dentition of 275 children (125 cases and 150 controls) aged 5–9 years. The authors assessed the presence/absence of present or past allergic rhinitis by clinical examination and through the use of questionnaires directed to the parents. Children with a history of allergic rhinitis exhibited a three-fold increased risk of developing posterior cross-bite or overjet (Odds Ratio [OR] = 3.16; 95% CI =1.79–5.58; $p < 0.001$), but a significant association with anterior open-bite was not found ($p > 0.05$). Investigators concluded that oral breathing caused by allergic rhinitis is a real associated risk factor with the presence of malocclusions in children and adolescents.
2 American researcher paediatric dentists [Lenahan et al., 2015] evaluated the overall safety and effectiveness of a Meperidine/Hydroxyzine sedative drug combination and identified potential factors that may influence sedation effectiveness in paediatric patients. The study was performed on 248 electronic files from 131 female and 117 male children who were attended under sedation with that drug regimen at a university dental clinic. Over 80% of sedations were considered effective or very effective, according to the Frankl behavioural scale, and fewer than 5% were aborted due to lack of clinical results. Adverse effects were present only in one case. Therefore, the drug combination Meperidine/Hydroxyzine was considered safe and effective for children with poor cooperation levels.

Case-control studies are relatively quick and inexpensive with regard to other designs. Other advantages include that they allow the search for multiple risk factors and do not require a large number of cases. However, they are prone to diverse biases, which may put at risk or misinterpret the study results [Introductory Statistics, 2011]. The most important biases affecting case-control studies are, first, the recall bias, because of the retrospective nature of these studies [Niven et al., 2012]; thus, a great deal of necessary information relies on the children's and parents' memory concerning past events or facts. It has been demonstrated that children's parents who have experienced harmful exposures to a risk factor may be more motivated to recall this risk factor [Morgenstern and Sohn, 2010]. The recall bias also depends on data gathered from medical/dental records, which may be inaccurate or incomplete. Second, there is the selection/allocation bias, caused by poor selection of control participants, who should be appropriately matched with cases, with the purpose of limiting the effect of extraneous variables [Hackshaw et al., 2006; Steel and Pearce, 2009; Shahar and Shahar, 2012]. Finally, we find the measurement bias, when measurement errors are systematic, not random; for example, when an examiner persistently assigns higher scores for carious lesions in primary molars where others do not; then, it is possible that the examiner assigning higher scores for carious lesions may be biasing the caries scores estimates. In these cases, training and calibration of evaluators comprise a fundamental part of the measurement process in OS [Steel and Pearce, 2009].

A subtype of the case-control design is the nested case-control study, in which cases and controls are selected from an existing cohort, so that cases and controls maintain the same risk level during the study time [Jepsen et al., 2004]. This case-control design subtype is employed when the measurements-of-interest are too expensive or time-consuming to be employed on the whole cohort [Steel and Pearce, 2009].

Statistical aspects. It is impossible to estimate the RR from case-control studies –as in cohort studies– because this risk measure requires knowledge of disease rates rather than exposure rates; thus, the OR is calculated [Dawson-Saunders and Trapp, 1994; Petrie et al., 2002], utilizing a 2 × 2 contingency table (see Appendix). The odds of the disease are the chance of having the disease divided by the chance of not having the disease, both estimated in children who are exposed and non-exposed [Jepsen et al., 2004; Niven et al., 2012]. Thus, the OR is the division between the exposed odds and the non-exposed odds. Like the RR, a large OR must be clearly greater than 1; in other words, the OR of the 95% CI must exclude the value of 1 in order to consider the risk factor as significant (see Example 2 corresponding to this design) [Petrie et al., 2002; Jepsen et al., 2004; Niven et al., 2012].

For case-control studies with matched or paired data, there are diverse appropriate statistical techniques, such as the following hypothesis tests: paired Student t test; Wilcoxon Signed Ranks; McNemar, and Mantel-Haenzel; the logistic regression is useful for handling multiple risk exposures and potential confounding factors [Niven et al., 2012].

Cross-sectional studies

Cross-sectional studies provide a current image of

the outcome-of-interest, for example, the frequency or prevalence of an uncommon or long-term disease in the population or the possible exposure–condition association, at any given time point [Introductory Statistics, 2011]. In this design, only one measurement is taken of each participating child; thus, it is unable to provide reliable evidence of a temporal relationship between risk factors and diseases, in other words, the progression of the disease development, because the data of both variables are collected simultaneously [Petrie et al., 2002]. However, recruitment may take place across a long time period [Sedgwick et al., 2014]. Basically, researchers take a sample of children from a well-defined population and record specific information about them at one time point with respect to the variables-of-interest (e.g., exposure and outcome); then the data are analyzed [Krithikadatta, 2012].

Cross-sectional studies are divided into three types: for staging a disease; for testing new diagnostic approaches for an oral condition, and for surveys. In the first case, the purpose of the investigation is to assess the potential association (not a clear causality) between a risk factor and a condition, or for estimating the prevalence of an oral disease (another measure of disease frequency) in a particular paediatric population; prevalence represents the proportion or rate of individuals in the total population who have the disease at a point in time (point prevalence) [Jepsen et al., 2004]. Prevalence can be also assessed over a defined period of time (period prevalence), for example 1 year, when it takes time to accumulate sufficient information on the disease in a population (e.g., What proportion of young children attending a public health service have early childhood caries?). These cross-sectional studies are also classified as descriptive or analytical. Descriptive studies simply characterise the prevalence of a disease in a specified population, without carrying out comparisons. In analytical studies, data on the prevalence of both the exposure and the disease are obtained for the purpose of comparing findings or differences between exposed and non-exposed study participants [Olsen and St. George, 2004]. Such studies are important when allocating resources and planning oral healthcare services.

In the second type, or diagnostic tests, clinical investigators are interested in the usefulness of a new diagnostic procedure [Pretty and Maupomé, 2004]. The objective of diagnostic tests is to define a child as either having or not having a specific disease. Cross-sectional studies are useful to determine how good a new diagnostic test is in detecting a condition-of-interest in children who actually have this condition (sensitivity) or how good the test is for excluding patients who actually do not have the same condition (specificity). In addition, other measures should be calculated, such as positive and negative predictive values, which are the probabilities that a child who has a positive (or negative test) actually has (or does not have) the disease. Diagnostic tests are also useful for screening patients or for determining whether children in an apparently healthy paediatric population are likely to have the disease/condition-under-investigation [Petrie et al., 2002; Pretty and Maupomé, 2004]. On the other hand, surveys intend to measure knowledge, attitudes, or behaviours on a perplexing topic or to learn how people think or feel about an issue. For these purposes, researchers employ interviews or written questionnaires to collect data from the research participants only once; thus, there are losses to follow-up of survey participants [Introductory Statistics, 2011]. However, surveys may be prone to non-response bias if children who have parental consent to take part in the study differ from those who do not, which may result in a sample that is not representative of the population [Sedgwick et al., 2014]. The design of the question instrument to be applied to the individuals is critical in obtaining reliable information [Dawson-Saunders and Trapp, 1994].

Cross-sectional studies may generate valuable hypotheses; further, they are relatively easy and less costly to conduct than experiments or other types of observational studies, and allow for the comparison of many different variables at the same time [Grimes and Schulz, 2002a]. However, their results may depend on the patients' accurate recall of past events [Introductory Statistics, 2011].

Statistical aspects: prevalence is the most common measure calculated in cross-sectional studies. For comparative studies, 2 × 2 contingency tables may be also created with the aim of displaying the counts and proportions and then to perform a Chi-square test or a Fisher test to assess the significance of the difference in proportions [Olsen and St. George, 2004].

Examples of published articles. Let us now see two recent examples of dental cross-sectional studies carried out in children.

1 Brazilian researchers [Vieira-Pinto et al., 2016] investigated the possible association between orofacial motor/functional impairments with nutritional status in children and adolescents suffering from Cerebral Palsy (CP). For each variable, an evaluation instrument was employed: the "Oral Motor Assessment Scale (OMAS)" and the "Nordic Orofacial Test Screening (NOT-S)" for orofacial disability, and nutritional status following WHO and Brazilian Ministry of Health criteria. One hundred ten children aged 6–16 years with CP were included and were examined, and their motor impairment was classified. Both variables were measured once. The results of this study indicated that weight gain was favored by better orofacial functional performance, independent of gross motor function ($p = 0.034$); dystonic CP forms presented mild impairment of oral motor function compared with spastic CP forms.
2 Funieru et al. [2016] assessed epidemiological parameters (e.g., prevalence) and sociodemographic factors of gingivitis in a Romanian population of

schoolchildren (aged 10–17 years). A total of 1,595 participants were selected, and an intraoral exam was performed to calculate Silness and Löe scores, and the prevalence and extent of gingivitis. Social condition was assigned through a questionnaire. Gingivitis prevalence was 91%, with higher gingival and plaque scores in boys. Those living in non-overcrowded houses, with parents who had superior education levels, and with direct access to school dental services exhibited better gingival conditions ($p < 0.05$). The authors concluded that the gingival condition was mostly associated with social gradients.

Confounding factors and bias in OS

Confounders and bias may influence the results derived from an OS, tending to overestimate or underestimate the true population value [Hakshaw et al., 2006]. The term "confounding", according to Christensen and Langberg [2012], includes external factors that "confound the assessment of the effect of an intervention". Confounding in OS occurs when two conditions are met: subjects who receive one treatment (or who have a risk factor) have different baseline characteristics than subjects who receive another treatment (or risk factor), and these characteristics exert an influence on the risk of failure [Butani et al., 2006].

A confounding factor is related with both the causal variable and the outcome variable. If these factors are not equivalent between the study groups, the difference in the results may be affected by their influence [Introductory Statistics, 2011]. The control of known or unsuspected confounding factors in OS cannot be achieved by random allocation of patients to the study groups, due to ethical and legal reasons [Hakshaw et al., 2006]. For example, in a study that attempts to evaluate the potential effect of cigarette smoking on periodontal health in adolescents, it is impossible to allocate participants to the smoking group or to the non-smoking group in random fashion and then observe the outcomes over time [Petrie et al., 2002]. Thus, the influence of confounding factors in the results of OS may be controlled by means of stratification, matching, and regression analysis [Petrie et al., 2002; Jepsen et al., 2004; Hackshaw et al., 2006]. Further, when conducting a comparative OS, it is important to balance the child participant's risk factors or disease status at the beginning of the study in order to diminish the potential influence on the final results [Butani et al., 2006; Christensen and Langberg, 2012].

Bias is a systematic, intentional or unintentional error that distorts a true value, producing ever unidirectional, higher or lower, estimates. Bias means that a measure of the exposure/outcome association is systematically wrong, and bias may occur during the design, conduct, or analysis of a clinical study [Althubaiti, 2016]. Bias arises from two main sources: selected subjects (the participating children), and errors during variable measurement [Jepsen et al., 2004]. Unlike confounding, which can be controlled in the statistical analysis, there is usually little that can be done about bias. Once biases are present in the data, it can be nearly impossible to rid the data from them [Steel and Pearce, 2009]. No statistical tests can adjust for bias effects [Hackshaw et al., 2006], and bias can really only be managed by taking steps in advance [Steel and Pearce, 2009].

Reporting OS

Diverse recommendations on the reporting of OS have been proposed. The STROBE statement is a checklist of items that should be addressed in articles reporting on the observational designs mentioned above: cohort, case-control, and cross sectional studies. The STROBE statement is a network integrated by methodologists, researchers, and journal editors who developed recommendations to assist authors when reporting observational research and help readers in the process of appraising published articles. However, these recommendations should not be taken as prescriptions for designing and/or conducting OS in paediatric dentistry research [von Elm et al., 2007].

Ethical issues in OS

Marshman et al. [2015] have mentioned that paediatric dentistry research should be mostly conducted with children rather than on children ("what adults think children think", assuming the superiority of adult knowledge), a modern concept known as child-centered research. In clinical paediatric dentistry research, ethical concerns arise in response to both the type of data collected during an OS and the methods employed to obtain that data. Ethical approval of the study protocol by an authorised ethics board will be required prior to the study onset. The protocol should include the corresponding informed consent form, in which parental permission for the child to participate in the study is implicated [Levin et al., 2005].

According to international regulations, paediatric research is not valid if it does not offer direct benefit with minimum risk of adverse effects to children while generating scientific knowledge [Wilfond, 2007]. Some observational research in paediatric involves, first, a direct relationship between participants and researchers that generates some obligation to provide health care and information to children and parents (e.g., on the progress of the study) and, in second place, it is sometimes necessary to apply questionnaires, interviews, psychological assessments, blood samples, imaging studies, and some interventions requiring sedation and other invasive procedures, which may represent inconveniences, discomforts, or psychological or physical risks. The key ethical goals in OS carried out in children are to minimise the risk of harm to the participants and to warrant the protection and confidentiality of all retrieved data of sufficient clinical value, personal information, and outcomes from the study.

Final comments and conclusion

In OS conducted with paediatric populations, the two basic components include exposure (a risk factor, a prognostic factor, a diagnostic test, or a treatment) and outcome (the result of the exposure action, i.e., a disease, a therapeutic effect, or even death). OS possess considerable significance in paediatric dentistry research because they measure or visualize the exposure/outcome association [Krithikadatta, 2012]. Estimating and comparing relative risks, odd ratios, incidence, prevalence, and other common measures will yield valuable clinical information about oral and systemic diseases. Furthermore, OS can often be carried out more quickly and inexpensively than clinical trials and require less cooperation from children. OS may be conducted in larger populations-of-interest, thus enhancing precision, study power, and generalisability; further, they can assess multiple study hypotheses simultaneously that involve different interventions or outcomes [Introductory Statistics, 2011; Krithikadatta, 2012). These designs are useful for generating new hypotheses and to justify the performance of randomised clinical trials. Historically, OS in the health sciences have demonstrated great usefulness when the main purpose was to obtain and understand convincing cause-and-effect evidence [Krithikadatta, 2012].

Other OS that, in our opinion, deserve careful review by the interested reader, due to their research quality and clinical implications for the paediatric dentistry field, are those of Skotowski et al. [1995] and Brown et al. [1999]. The former study conducted was case-control in design. The purpose was to determine the prevalence and severity of dental fluorosis in a sample of children (8–17 years-of-age) and to evaluate potential sources of fluoride as risk factors for fluorosis in permanent teeth. Questionnaires were employed to assess previous exposures to fluoride during the first 8 years of the lives of the study participants. Fluorosis was detected in 72% of children, although this was generally quite mild. The great risk for developing fluorosis was associated with the consumption of fluoridated water and the increased use of fluoride toothpaste. The authors recommended to parents and dental practitioners the prudential employment of a fluoride dentifrice, utilizing a small amount of this type of toothpaste on the toothbrush [Skotowski et al., 1995]. The second study analysed data from the first and third National Health and Nutrition Examination Surveys (NHANES I and NHANES III), including 28,000 and 39,295 children, respectively; the purpose was to report changes in untreated carious permanent teeth among children aged 6–18 years. Dental caries levels were recorded employing the Decayed-Missing-Filled Tooth (DMFT)/S index. Then, retrieved data were related with the age, gender, race, and poverty level of participants. Results indicated that, overall, the number of carious teeth decreased dramatically, from 1.43 from the 1970-74 period (NHANES I) to 0.33 in the 1988-94 period across the four demographic variables studied. According to the authors, these findings reinforce the already apparent shift from restorative to preventive dental services in the U.S. [Brown et al., 1999].

For conducting valid and reliable OS, investigators must report all information regarding the following four issues: study design employed according to the study objectives; child-participant selection criteria and process; the way that exposures and outcomes were measured, and how confounders and biases were identified and managed [Introductory Statistics, 2011; Christensen and Langberg, 2012]. Other specific factors that may influence the outcomes of an OS include initial sample size, patient attrition and length of follow-up, unit of analysis, and statistical analysis. Also, and particularly for prognostic studies, these characteristics should be noted: those related with the operators (expertise/training level, private or hospital practice, country, and number of participating operators), paediatric patients (age, cooperation level, oral health status, characteristics such as caries activity, diet, fluoride, and medical history, restored-tooth baseline conditions, occlusion, control teeth, and expected service length of the tooth and restoration), and materials and procedures (material components, type of isolation, preoperative diagnosis, pharmacological management technique) [Grimes and Schulz, 2002b; Butani et al., 2006].

All of this information serves to determine whether the findings of an OS are likely to be generalised or applied to a particular paediatric dentistry practice [Butani et al., 2006]. In this respect, Christensen and Langberg [2012] reported that many developed and subsequently published OS in the medical or dental literature are often poorly and insufficiently reported, hampering the assessment of the study's strengths and weakness and, in consequence, impeding arriving at an informed interpretation of the results by the clinician. Thus, Butani et al. [2006] have proposed a standard model with recommendations for the conduct and reporting of OS in paediatric dentistry research.

Observational research is particularly important for evaluating the outcomes and effectiveness of oral health care in real settings. The studies mentioned previously exemplify how, historically, observational designs have provided accurate and useful information in many human-health areas, in situations in which it is impractical or unethical to conduct randomised clinical trials [Christensen and Langberg, 2012]. The majority of clinical paediatric dentistry research relies on this type of design, especially when the purpose is to obtain convincing evidence to answer specific questions on disease causality.

In conclusion, observational studies play a preponderant role in clinical paediatric dentistry research because they often render the best source of information when there is the lack of experimental substantiation, providing that careful methodological attention be paid during their development.

		Disease present		Total
		Yes (+)	No (-)	
Exposition present	Yes (+)	a (exposed and diseased)	b (exposed and non-diseased)	a+b
	No (-)	c (exposed and non-diseased)	d (exposed and non-diseased)	c+d
Total		a+c	b+d	n

TABLE 2 Notation of a basic 2 × 2 table.

Appendix

How to Build and interpret a 2 × 2 contingency table to estimate an exposure/outcome association

A 2 × 2 contingency table is a type of matrix with four cells containing frequencies or counts (e.g., a cross tabulation) in which both groups of participants are classified by the combination of two categorical variables (exposure/disease). Each individual falls exactly into one of the two categories. Both exposure and disease are measured as dichotomous or binary variables. The outcome variable determines the column categories (disease present/disease absent), while the remaining variable or exposure defines the row categories (risk factor present/risk factor absent) [Petrie et al., 2002; Sauerbrei and Blettner, 2009] (Table2).

Definitions:

- Risk: The probability of failing ill
- Risk of disease for exposed children = a / a + b
- Risk of disease for non-exposed children = c / c + d
- Relative Risk (RR) = (a / a + b) / (c / c + d)
- Risk difference = (a / a + b) – (c / c + d)
- Odds for exposed children = (a / a + b) / [1 – (a / a + b)]
- Odds for non-exposed children = (c / c + d) [1 – (c / c + d)]
- Odds Ratio (OR) = a x d / b x c
- Prevalence of disease = a + c / n

This table aims to compare to groups of child subjects for assessing the cause/effect relationship in cohort, case-control and cross-sectional studies. For diagnostic tests the table notation is substantially different [Petrie et al., 2002; Pretty and Maupomé, 2004].

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