8

COHORT STUDIES

OBJECTIVES

After reading this chapter, you should be able to:

1. Distinguish between open and closed source populations as they relate to cohort study design.
2. Describe the major design features of risk-based and rate-based cohort studies.
3. Identify hypotheses and population types that are consistent with risk-based cohort studies.
4. Identify hypotheses and population types that are consistent with rate-based cohort studies.
5. Elaborate the principles used to select and measure the exposure in cohort studies.
6. Design and implement a valid cohort study to investigate a specific hypothesis.

8.1 INTRODUCTION

The word **cohort** denotes a group of study subjects that has a defined characteristic in common, and, in epidemiological study design, the characteristic of interest is the exposure status. In a cohort study design, we follow subjects from exposure forward to outcome (Grimes and Schulz, 2002). The study subjects can be individual animals, people, or aggregates of animals such as litters, pens or herds/flocks/kennels. Most frequently, the outcome of interest is the occurrence of a specific disease, although other outcomes such as premature removal from the herd (culling) or death might be the main focus of the study.

If the exposure status of potential study subjects is known beforehand, the selection of the study groups from the source population can be based directly on the exposure status (*eg* when we select 2 cohorts, a group of exposed and a group of non-exposed study subjects). If the exposure status is not known beforehand, another approach is to select a single group of subjects within which there will likely be a range of the exposure(s) of interest. Then, we determine the exposure status of each subject. We denote the first design as **cohort study** and the latter as a **single cohort** or **longitudinal** study; however, for purposes of study design we need not differentiate between them. In both instances, following their selection, we would ensure that the study subjects do not have the disease(s) of interest. Then, we would observe the study subjects for a defined follow-up period and compare the incidence of the disease in the groups defined by exposure status. In production-animal medicine, the outcome is often production measured on a quantitative scale (*eg* weight gain, milk production, race time, eggs per year *etc*). Nonetheless, comparing the production of exposed and unexposed subjects also fits the cohort study design paradigm. For example, we might follow 2 groups of calves, one that received adequate colostrum within 8 hours of birth and one that did not, to ascertain the impact of these exposures on future growth rates (see Example 8.1). In some studies, the initial sampling is based on the exposure status but it is not possible (practical) to measure incidence of new cases so the researchers measure prevalence of the disease at the time the study subjects are selected. We treat these as cross-sectional studies (see Chapter 7 for details).

The basis of the cohort study design is to compare the frequency of an outcome in 2 groups of subjects that are similar in all regards except for exposure; the quality of the study results will depend on how closely the real study comes to that ideal. Comprehensive (Rothman and Greenland, 2008) and classical reviews of cohort study design and analysis are available (Prentice, 1995; Samet & Munoz, 1998), as well as a more recent series of articles on the critical appraisal of cohort studies, from a clinical perspective (Mamdani *et al*, 2005; Normand *et al*, 2005; Rochon *et al*, 2005). The latter discussions focus on methods to prevent selection

Example 8.1 A risk-based retrospective cohort study investigating health and productivity outcomes

Dewell *et al* (2006), studied the association of neonatal serum immunoglobulin (IgG1) concentration with health and performance in beef calves. In this study, 1,568 crossbred beef calves from beef-breed dams, that were 4 years of age or older and had been bred to purebred bulls in each of the years 1996, 1997, and 1998 at a US Meat Animal Research Center were eligible for inclusion (note the restrictions for entry to the study). Sera were collected from calves between 24 and 72 hours after birth and analysed to determine IgG1 concentration (*ie* the exposure level measured on a quantitative scale). Outcomes included weight gain, morbidity and mortality from birth to weaning (approximately 200 days of age).

bias and confounding. We will discuss the reporting of cohort studies subsequently (see Section 8.8).

Each specific study presents its own unique challenges, but the starting point for all studies is to clearly and concisely state the objective(s). This includes defining the **target** (the population to which inferences will be made) **and source populations** (the population from which the study group will be drawn), the **unit of observation (***eg* individuals or aggregates**)**, the **exposure**(s), the **disease**(s), the **follow-up period** and the setting (*ie* context or venue) of interest. If sufficient biological facts are known, the hypothesis should indicate the amount or duration of exposure that is believed to be needed to 'cause' the disease. Clarifying the study objective(s) often helps us decide whether current or past exposure is relevant, whether lifetime exposure or exposure in a narrower window of time is important, whether repeated measures of exposures are required and if so, how to handle changes in exposure status.

Depending on the availability of suitable records, cohort studies can be performed **prospectively** or **retrospectively**. In a prospective study design, the disease has not occurred at the time the study starts; whereas, in a retrospective study design, the follow-up period has ended, and the disease event has occurred, when the study subjects are selected. Prospective studies provide the opportunity for more detailed information-gathering and attention to recording the details of interest (*eg* Jacob *et al* (2005)) than retrospective studies which require suitable existing databases (*eg* Egenvall *et al* (2005)).

8.2 STUDY GROUP

When selecting the exposure groups, it is best if the groups come from one source population. This helps ensure that the study subjects have numerous characteristics in common and it can reduce the background 'noise' and/or the risk of unmeasured confounding. As an example, the source population might be geographically defined (*eg* swine farms in a given area), or it could be a virtual population as in the 'group of dogs at a clinic' or the 'group of farms served by a veterinary practice'. Other study-subject specific eligibility criteria such as age, breed, herd size *etc* can be used to define the study groups; all eligibility criteria should be specified explicitly. Most often, the study subjects are chosen purposively, not randomly, from the source population. Although this increases the risk of selection bias, it is often the only practical way to proceed with a study. This aspect of study design will be discussed in greater detail in Chapter 12.

Usually, in developing a cohort design, we assume that an equal number of exposed and non-exposed individuals will be selected. However, there is nothing magical about this assumption and, if cost or other practicalities dictate different sample sizes across exposure categories, then this can be accounted for. Initial estimates of sample size can be performed assuming the disease is measured by risk (see Section 8.2.1), as shown in Chapter 2. This approach is often sufficient for initial estimates of sample size even if the population is open and a rate-based study (see Section 8.2.2) must be used. Computer software for sample size estimation usually allows for unequal sample sizes and repeated measures when planning the sample size. More recent software is available to estimate sample size when using multivariable regression models, or proportional hazard models for analyses (Latouche *et al*, 2004). Cai and Zeng (2007), discuss sample size estimation in cohort and case-cohort designs. Matsui (2005) discusses sample-size estimation for rate-based designs where the outcome is survival time. Mazumdar *et al* (2006), discuss sample-size estimation in strata-matched designs with survival time outcomes.

8.2.1 Risk-based (cumulative incidence) designs

This is the simplest form of cohort study, but it requires that a number of assumptions are met in order for the design to be valid. First, the exposure groups must be defined at the start of the study and remain unchanged during the study (*ie* they are **fixed cohorts**). Second, the study groups must be **closed** in that all subjects must be observed for the full risk period (*eg* a few days around calving for milk fever in dairy cows, or the first 30 days in a feedlot for respiratory disease in feedlot calves). In this design, it is best if there are few (or no) losses (losses include study subjects that develop other diseases, or die from other outcomes, as well as withdrawals from the study). If the percentage of study subjects that is lost becomes large (some use >10% as a cutpoint), this will begin to cast doubt on the validity of the study findings. For these reasons, risk-based designs work best for diseases (outcomes) with a relatively short biological risk period (*eg* milk fever after calving in different breeds of dairy cow). Since all subjects are observed for the full risk period, this allows the calculation of risk in each of the exposure categories. For chronic diseases such as foot lesions, where the risk period is lifelong and hence, usually greater than the follow-up period, a rate-based design is often preferred.

In a 2X2 table, the summary format for classifying the study subjects in a risk-based cohort study is shown below:

	Exposed	Non-exposed	Total
Diseased	a_1	a_0	m_1
Non-diseased	b_1	b_0	m_0
Total	n_1	n_0	n

In this design, we select n_1 exposed and n_0 non-exposed individuals from the N_1 exposed and N_0 non-exposed individuals in the source population. Having ensured that none of the study subjects has the disease at the start of follow-up, we observe all subjects for the defined follow-up period. During the follow-up period, we note that a_1 exposed subjects develop the disease out of the n_1 exposed subjects and a_0 non-exposed subjects out of the n_0 non-exposed subjects develop the disease. Overall, we observe a total of m_1 diseased and m_0 non-diseased subjects. The 2 risks (R) of interest are:

$$R_1 = a_1/n_1 \qquad \text{and} \qquad R_0 = a_0/n_0$$

Note The denominator of interest is the number of subjects in each exposure category. Example 8.1 describes a risk-based cohort study.

8.2.2 Rate-based (incidence density) designs

In many instances, all study subjects are not under observation for their full risk period. This is especially true if the source population is dynamic and the follow-up period is long. Thus, some subjects may be added to the study group part way through the biological risk period for the outcome of interest. In addition, a significant proportion of subjects may withdraw from the study part way through the follow-up period. Also, the exposure status of subjects might change during the study period. In these situations, we cannot just count the number of exposed and non-exposed subjects; rather, we need to accumulate the amount of 'at-risk time' contributed by each study subject in each of the exposed and non-exposed groups. Thus, the denominator becomes the amount of study-subject-time per exposure group and this requires a rate-based

approach to study design and analysis.

In this design, each of the initially selected exposed and non-exposed subjects contributes 'at-risk' time to the denominator of the rates until they develop the disease, or are lost to the study, or their observation ends because the study is terminated. If new individuals are added to the study group during the follow-up period, then the amount of time-at-risk for each study subject is added to the appropriate exposed or non-exposed category.

In a 2X2 table, the summary format for classifying the study subjects in a rate-based cohort study is shown below.

	Exposed	Non-exposed	Total
Diseased	a_1	a_0	m_1
Animal-time at risk	t_1	t_0	T

Initially, we might select n_1 exposed and n_0 non-exposed individuals from the source population. All these subjects are followed for the duration of their time-at-risk within the study period. In so doing, we observe a_1 exposed cases of the disease out of t_1 animal-time units of exposure and a_0 non-exposed cases of the disease out of t_0 non-exposed animal-time units. Here t_1 is the sum of all of the exposed time-at-risk contributed by the study subjects whether they developed the disease or not. Similarly t_0 is the summed time-at-risk study subjects contributed in the non-exposed category. The 2 rates (I) of interest that we wish to estimate would be:

$$I_1 = a_1/t_1 \qquad \text{and} \qquad I_0 = a_0/t_0$$

If the follow-up time is relatively short, the above rates can be used to measure disease frequency. If the follow-up time is sufficiently long that the assumption of a constant rate over the entire follow-up period is highly suspect, survival analysis methods can be used to overcome this difficulty (see Chapter 19). Examples 8.2, 8.3 and 8.4 describe 3 rate-based cohort studies.

8.3 THE EXPOSURE

In cohort studies, our objective is to identify the consequences of a specific exposure factor. The exposure refers to any potential cause of disease and, as examples, these might range from characteristics of the study subject, to infectious or noxious agents, to housing, management or feed-related factors. Although measuring exposure might seem simple, at first glance, careful thought should be given to the manner in which it is measured and expressed. Each study design should include the details of what constitutes exposure, and whenever possible, we should specify how long after an **exposure threshold** is reached before one might reasonably expect to see the disease arise from that exposure (*ie* the **induction period**).

Exposure status can be measured on a dichotomous scale (*eg* exposed or non-exposed), an ordinal scale (*eg* low, medium, or high dose), or a continuous scale (*eg* organisms per gm of feces, ppm of a toxin in air or water, gm of colostrum ingested *etc*). Exposure can be expressed separately in terms of dosage and duration or as a combination of the two (*ie* perhaps their product). Often it is necessary to decide whether lifetime exposure, historical exposure, or current exposure is the best measure of 'exposure'. Because both exposure status and 'time exposed' are crucial components of a valid cohort study, it is vital to reduce the measurement error for exposure. To achieve this often requires a good understanding of the 'exposure agent'

Example 8.2 A rate-based cohort study to assess associations between selected factors and incidence rates of fracture

Verheyen *et al* (2007) studied the potential impact of age and parity of the mare on the fracture rate in her offspring. The study included data from 335 Thoroughbred racehorses from 8 trainers, located across England. Horses joined the study when they entered training as yearlings. Data on the study horses' date of birth, dam and sire were collected when they were enrolled. Training speed and distance was recorded daily and each horse was monitored for up to 2 years. The outcome was a confirmed diagnosis of fracture, excluding fractures that resulted from a traumatic incident.

Since each horse potentially had a different follow-up period, fracture rates per horse-month was the outcome. Descriptively, the fracture rate in the first foal of a mare was 0.56 per 100 horse-months of risk and 1.24 per 100 horse-months in foals from parity 2 or greater mares, giving an incidence rate ratio (*IR)* of 0.45. A Poisson regression model was used to estimate the impact of dam age and parity on the fracture rate. This approach related the occurrence of a case to the number of horse-months of exposure in each age (or parity) category of mare, and any association between the potential risk factor(s) and the case status was described by an *IR.*

and of the etiologically high-risk period for disease causation.

8.3.1 Permanent exposures

The exposure might be a permanent factor (*ie* time-invariant) or a factor that can change over time. Permanent exposures include factors such as sex, breed and one-time exposures such as vaccination, or whether or not a calf received sufficient colostrum within 12 hours of birth. Permanent and 'one-time' exposures are relatively easy to measure, but even here a moment's thought would suggest that defining 'sufficient' or 'inadequate' with respect to colostrum intake in a calf might be more complex than it first appears to be because it may have a time of exposure as well as a quantity of exposure component. In any event, for factors where the exposure is based on a threshold or dosage, the amount of exposure necessary to deem an individual as being 'exposed' needs to be clearly stated. In early studies of a health problem, the objective might be to assess if there is an exposure threshold, and if so at what cutpoint? If the disease event occurs before exposure is completed, it should not be included as an event in the analysis because exposure has not been completed, so it could not have caused the disease. These issues are shown graphically in Fig. 8.1.

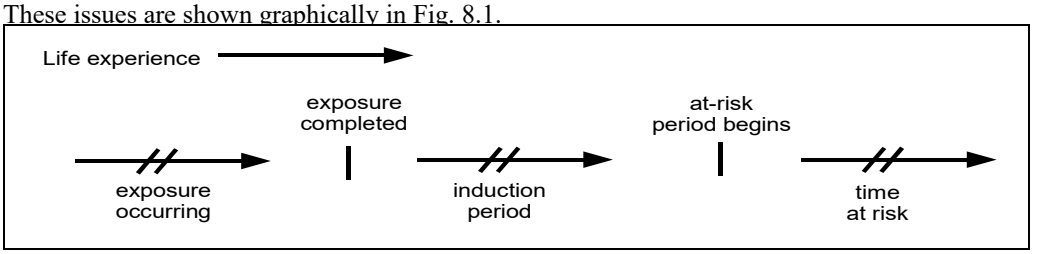

Fig. 8.1 Life experience with exposure, induction period and time at risk

When exposure is measured on a continuous scale but, for purposes of the analysis, categorisation of exposure is desirable, the criteria for categorisation should be clearly explained. An example of a cohort study with a permanent exposure factor (24-72 hour immunoglobulin level in calves) is presented in Example 8.1.

Example 8.3 A retrospective cohort study of survivorship to the occurrence of canine hip dysplasia

van Hagen *et al* (2005), studied the incidence of, and risk factors for, hind-limb lameness caused by canine hip dysplasia (CHD) in a birth cohort of boxers. In this study, 1,863 purebred boxers from litters born in The Netherlands between January 1994 and March 1995 were followed until June 2002. The diagnosis of CHD (n=97) was made by the client's veterinarian and was based on clinical signs (the diagnostic criteria were not specified). Dogs that were lost to follow-up were regarded as censored. Risk factors included individual data such as sex and whether a dog was sexually intact or neutered. If neutered, the age at neutering was recorded (neutered dogs contributed time-at-risk in both the unexposed (non-neutered) and exposed (neutered) categories. Time to the development (diagnosis) of CHD was the outcome of interest. Thus, associations between the risk factors and the time to CHD were analysed using a proportional hazards model (see Chapter 19).

8.3.2 Non-permanent exposures

Non-permanent exposures can include factors such as ration, housing, or environmental exposures that can change over time, or study subject specific factors such as undergoing specific procedures such as hoof-trimming or neutering where the timing (age, stage of lactation *etc*) of the procedure can be important. For these exposures (*eg* the type of housing experienced by a cow over 2 lactations), both the timing of, and the extent of, the exposure might be important to measure and analyse. This adds complexity to the measurement of the exposure factor (*eg* the timing of neutering in Example 8.3). Sometimes a simple summary measure of exposure will suffice (*eg* days spent on concrete versus dirt flooring), whereas in other studies more complex measures of exposure are needed (*eg* the number of days spent housed in different stall designs where the stall size and the flooring material also might need to be considered). The more information that can be collected on exposure, such as the exposure level(s), when exposure started, and when (if) exposure stopped, the better, as it adds credibility to conclusions about causal relationships, is more useful for preventive action or intervention, and enhances our biological understanding of the problem. Examples 8.3 and 8.4 are cohort studies with exposures that changed over time.

To obtain the exposure time, for each study subject, the time-at-risk in each exposure category accumulates from the moment exposure is completed until the event of interest occurs, the

Example 8.4 A retrospective cohort study to assess time to outcome in cohorts of Irish cattle herds

Olea-Polpelka *et al* (2004) retrospectively studied over 6,000 herds that had a new occurrence of bovine tuberculosis in 1995 (the 'exposed' group) and over 10,000 herds that were free of tuberculosis during 1995 (the 'unexposed' group). The outcome was the time until the next tuberculosis breakdown, if one occurred, during the next 5 years. The exposure status of each herd in this study was based on the number of tuberculosis cattle reactors during the 1995 episode of bovine tuberculosis (including 0 reactors in the clear herds) and 5 categories of increasing severity were formed. Although this was a fixed cohort (only herds with new bovine tuberculosis in 1995 were followed and all were followed for up to 5 years), it was not realistic to assume a constant risk over the entire follow-up period so survival methods (proportional hazards) were used for analysis. This essentially compared the incidence rate of new occurrences (after the 1995 episode cleared) in each of the exposure categories over a time period of up to 5 years.

subject becomes lost to follow-up or the study ends. With losses to follow-up, time-at-risk accumulates until the last date exposure status is known (if the precise time of loss is unknown use the midpoint of the last known exposure period). If there is a known induction period following completion of exposure, then, until that period is over, the time at risk of 'exposed' individuals should be added to that of the non-exposed group. Some researchers prefer to discard the disease experience during the induction period for exposed individuals because of uncertainties about the duration of the induction period. In the face of uncertainty about these effects, this is likely the best choice providing there is sufficient time-at-risk in the non-exposed group to maintain precision.

Note that when the exposure status can change, an individual study subject can accumulate time-at-risk in both exposed and non-exposed groups. Previously non-exposed subjects contribute time-at-risk to the exposed category after the exposure threshold is reached. Similarly, if previously exposed individuals become non-exposed, we would add the non-exposure time (of previously exposed individuals) to the non-exposed cohort only after the time period when any lag effects that could be present had ended. Provided lag effects are minimal, when different exposure categories exist for the same study subject, the exposure category assigned to subjects who develop the disease is that level of exposure the subject was in at the time the outcome event occurred.

So far, in this section we have classified the exposure status of study subjects as exposed or non-exposed (*ie* a dichotomous exposure) or perhaps on an ordinal level of exposure category. However, in many studies, exposure is measured on a continuous scale and the threshold to complete exposure may either be unknown, or it is deemed more appropriate to model the exposure-outcome association in a dose-response manner. As in other instances, maintaining the continuous scale has advantages because the categorisation of a continuous exposure variable usually results in loss of information. In this instance, one might relate the disease frequency (*ie* risk or rate) to exposure on a continuous exposure scale using an appropriate regression model (Waldner, 2008a; Waldner, 2008b); (see also Example 8.1).

8.4 Ensuring exposed and non-exposed groups are comparable

If the study subjects in the different exposure groups are not comparable with respect to factors related to both the outcome and exposure, a biased (*ie* confounded, see Chapter 13 for a discussion of confounding) assessment of the exposure-outcome association can result (Klein-Geltink *et al*, 2007). In general, one or more of the following 3 approaches can be used to help ensure that the exposed and non-exposed groups are comparable except for their exposure status. The first of these approaches is applied prior to subject selection and involves the use of exclusion or restricted sampling of study subjects. In this approach, we identify variables likely to be confounders and then we restrict the selection of study subjects to those that have only one level of these variables (*eg* include only one age, one breed, or one sex of animal in the study). This prevents confounding by the specified factors, serves to reduce the background variability among study subjects, and might help reduce confounding from other unknown factors. A second approach is used at the time of study-subject selection and involves matching the level of confounders in study subjects across the exposure categories. To accomplish this, we identify major confounding variables and then select the non-exposed subjects so that they have the same level of the confounder as the exposed subjects (the exact criteria for matching should be specified and reported). Matching can help achieve greater study efficacy as well as prevent confounding in cohort studies. The third method to prevent confounding is to use

analytic control. In this approach, we identify and measure the important confounders and then use statistical control (*eg* ranging from Mantel-Haenszel-type stratification to multivariable regression approaches) during the analysis to adjust for these confounders (see Chapters 13 for a more detailed discussion of confounding), in an attempt to obtain unbiased measures of association.

8.5 FOLLOW-UP PERIOD

To enhance the validity of a cohort study, the follow-up process must be as complete as possible and unbiased with respect to exposure status. Achieving unbiased follow-up may require some form of observer-blinding process as to exposure status. 'Blinding' can be implemented in both prospective and retrospective studies (although the latter has more limited options). For example, in a prospective study, one set of researchers who is unaware of the exposure status can be assigned the task of follow-up. In a retrospective study, the researchers reviewing records for the outcome should be kept unaware of the exposure status whenever possible. In either situation, the date of outcome occurrence should be as accurate as possible to reduce the possibility of measurement error. If passive surveillance for cases is used, cases occur when identified (*eg* this might be the date of first symptoms, or veterinary examination). With active surveillance and regular evaluation of study subjects it is feasible to get more accurate data on time of outcome occurrence (Jacob *et al*, 2005).

Unless the study period is short, it is helpful to enumerate and characterise the population at risk at specified times during the study as noted by Tooth *et al* (2005); these numbers should be reported. Collecting ancillary information is useful to help manage issues such as loss to follow-up because of competing risks including culls or sales of study subjects, and to assess if censorship is unrelated to exposure.

8.6 MEASURING THE OUTCOME

Each study will need explicit protocols for determining the occurrence and timing of outcome events. Clear definition(s) of **diagnostic criteria** are useful to ensure as few diagnostic errors as possible (*eg* what constitutes hip dysplasia or lameness). This can prove difficult in retrospective studies when only the summary diagnostic information is available. The specific diagnostic criteria should be included in the study plan for prospective studies. When possible, in prospective studies, also ensuring **blinding** of the diagnosticians is helpful to equalise, but not necessarily reduce, diagnostic errors.

Since the disease is measured as incidence, strictly speaking, this requires at least 2 examinations: the first at the start of the follow-up period to ensure that the study subjects did not have the disease of interest, and the second to investigate whether or not (and when) the disease developed during the observation period. Including only new disease events circumvents the reverse-causation problem from measuring prevalence as well as ensuring that the associations are not biased by duration-of-disease effects and survival bias (see Chapter 12). In retrospective studies, one often has to assume freedom from the disease at the start of the follow-up period. In prospective studies, it is desirable to formally ensure that the study subjects are free of the disease at the start of follow-up.

If clinical diagnostic data are used to indicate the occurrence and timing of the disease event, the incident date usually will be based on time of diagnosis not on time of occurrence of

disease. For diseases that might remain in the subclinical state for extended periods, ignoring this difference could lead to inferential errors. If the study group is screened for the disease event at regular intervals, then the time of occurrence of the disease should be placed at the midpoint between examinations.

As noted previously, when epidemiological methods are used in the context of animal production medicine, the outcome often is a production parameter, not disease occurrence. Here, the dates of assessment of production are of interest. Examples of a cohort study with a continuous production variable as an outcome include investigations to identify factors affecting the growth rate of swine (Johansen *et al*, 2004) or factors influencing the milk production of dairy cows (Berry *et al*, 2007). In the latter study, repeated measures of milk production (approximately monthly) as well as lactation totals were available. In this and other studies, the researchers need to decide which of these measures is most important for evaluating the hypothesis of interest and ensuring that the data for the appropriate analytic methods are obtained.

One of the advantages of a cohort study is that we can assess multiple outcomes from a given exposure factor. In terms of causal inferences, Kunzli *et al* (2001) have indicated that following a defined group of study subjects over time allows the researcher to capture all deaths in the study group regardless of whether the effects of exposure are short or long term. However, if multiple outcomes are assessed (*eg* Berry *et al* investigated both milk production and disease occurrence), some might be significantly associated with the exposure by chance alone. In this instance, it might be best to consider the study as hypothesis-generating not hypothesis-testing, unless the outcomes were specified *a priori,* or a penalty is applied to the P-value which is deemed as 'statistically significant'.

8.7 ANALYSIS

8.7.1 Risk-based cohort analysis

If the source population is closed, we can measure the average risk of disease(s) and survival times during the follow-up period. Bivariable risk-based analyses are shown in Chapter 6, and stratified analyses (to control confounding) in Chapter 13. Traditionally, multivariable models have been built using logistic-regression models (Chapter 16) which use odds ratios as the base measure of association. For example, Green and Cornell, (2005) used logistic analysis in a risk-based study of factors associated with a cattle herd 'breaking down' with tuberculosis. Recently, it has been shown that using log-binomial models (see Chapter 16) and Poisson models (see Chapter 18) allow direct unbiased estimation of risk ratios which is the natural measure of association for a risk-based cohort study.

Cheung (2007) has described the use of linear regression if risk difference, rather than risk ratio, is the association measure of interest.

Both Cox (2006) and Greenland (2004) discuss the estimation of population attributable fractions (AF_p) in cohort studies. Cox suggests using a log-linear model approach for adjusted AF_p when the prevalence of exposure is known (*ie* a single cohort or longitudinal study sampling) or estimable (available in some cohort study situations). Greenland demonstrates how to obtain a variety of association measures, including AF_p, using one or more of logistic, log-linear and Poisson models.

8.7.2 Rate-based cohort analyses

If the source population is open, rates are used to measure disease frequency (see Example 8.3). Most formal analyses of rate data in the veterinary literature have used survival models (*eg* Egenvall *et al* (2005); Jacob *et al* (2005)). Callas *et al* (1998) compared a proportional hazard, Poisson and logistic model for the analysis of cohort data, and concluded that one of the former 2 approaches would be preferable to logistic analysis. This has been confirmed more recently by others (Greenland S, 2004). For multivariable analyses of grouped data, you can use a Poisson regression model (see Chapter 18) that includes the study-subject time at risk in each exposure category as the offset; the coefficients from this model provide direct estimates of the incidence rate ratio. As noted earlier, the incidence of disease is expressed relative to the time at risk in each level of exposure, not to the number of exposed (or non-exposed) individuals. If the time of disease (outcome) occurrence is of more interest than the fact of its occurrence, survival models are the method of choice (Case *et al*, 2002). Olea-Polpelka *et al* (2004) provide an example of this approach (see Example 8.4). Example 8.5 contains an example of a rate-based cohort study of colic in horses.

If the measure of exposure is a composite (*eg* 'total exposure' determined from the exposure level multiplied by the number of days of exposure), then it might be advantageous to study the 2 components separately, in the same model, because their effects might differ (it might be the chronicity of exposure rather than the exposure level that increases the risk of disease).

8.8 REPORTING OF COHORT STUDIES

As mentioned in Chapter 7, there has been a widespread initiative to improve the reporting of observational studies (STROBE; (von Elm *et al*, 2007)). We elaborated on these in this chapter as they should be used to help plan and report the study, as well as to help you, the reader, assess the validity of published cohort studies (see Table 8.1 for design aspects specific to cohort studies and Table 7.2 for all observational study types).

Example 8.5 A retrospective cohort study of risk factors for survival of horses after surgery for colic

Proudman *et al* (2005), reported on a study of factors affecting long-term survival of horses recovering from surgery of the small intestine. The source population was 382 horses that had colic surgery at the Faculty of Veterinary Science, University of Liverpool between March 1998 and March 2004. The different exposure groups were based on data for a number of potential risk factors including age, breed, clinical pathology parameters, and the nature, extent and duration of surgery. Survival time was measured as a continuous variable starting at recovery from surgery until death, censoring or March 18, 2004. The survival time varied from one day to over 1,500 days (no descriptive statistics were given by the authors). Data were analysed using a Cox proportional hazards model.

Table 8.1 Criteria used to rate published cohort studies (Tooth et al, 2005)

Criteria
1. Are the objectives or hypotheses of the study stated?
2. Is the target population defined?
3. Is the sampling frame defined?
4. Is the study population defined?
5. Are the study setting (venue) and/or geographic location stated?
6. Are the dates between which the study was conducted stated or implicit?
7. Are eligibility criteria stated?
8. Are issues of 'selection in' to the study mentioned?
9. Are the number of participants justified?
10. Are numbers meeting and not meeting the eligibility criteria stated?
11. For those not eligible, are the reasons why stated?
12. Are the numbers of people who did/did not consent to participate stated?
13. Are the reasons that people refused to consent stated?
14. Were responders compared with non-responders?
15. Was the number of participants at the beginning of the study stated?
16. Were methods of data collection stated?
17. Was the reliability (repeatability) of measurement methods mentioned?
18. Was the validity (against a 'gold standard') of measurement methods mentioned?
19. Were any confounders mentioned?
20. Was the number of participants at each stage specified?
21. Were reasons for loss to follow-up quantified?
22. Was the 'missingness' of data items at each wave mentioned?
23. Was the type of analyses conducted stated?
24. Were 'longitudinal' analysis methods stated?
25. Were absolute effect sizes reported?
26. Were relative effect sizes reported?
27. Was loss to follow-up taken into account in the analysis?
28. Were confounders accounted for in the analyses?
29. Were missing data accounted for in the analyses?
30. Was the impact of biases assessed qualitatively?
31. Was the impact of biases estimated quantitatively?
32. Did authors relate results back to a target population?
33. Was there any other discussion of 'generalisability'?

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