EFFICIENCIES OF OBSERVATIONAL PLANS FOR AN ILLNESS-DEATH MODEL

by

Ørnulf Borgan
Institute of Mathematics, University of Oslo,
Blindern, Oslo 3, Norway

Knut Liestøl
Institute of Informatics, University of Oslo,
Blindern, Oslo 3, Norway

Peter Ebbesen
Danish Institute for Research
on Cancer, Radiumstationen,
Aarhus 8000 C, Denmark

SUMMARY

Disease development is described by the progressive Markov illness-death model with one disease. The efficiencies of four observational plans are discussed: the simple survival experiment, serial sacrifice, periodic diagnosis, and complete observation. The discussion is based on the "semi-parametric" assumption of piecewise constant intensities. The efficiency of simple survival experiments is shown to be very low, serial sacrifice experiments to be moderately efficient, and periodic diagnosis to be almost as efficient as complete observation. An application to the analysis of leukaemia development in laboratory mice is given.

KEY-WORDS: Animal experiments; Efficiency studies; Markov illness-death model; Observational plans.

1. Introduction

In recent years there has been an increasing interest in the development of suitable methods for the study of time to appearance and rate of progression of diseases (cf. Neyman, 1980). Such methods include both an efficient observational plan for the collection of the data and an adequate statistical model for their analysis.

Our interest in these problems emerged from an attempt to analyse data from experiments on the development of lymphatic leukaemia among laboratory mice. The data were obtained from simple survival experiments, i.e. the animals had only been examined after death. Such experiments are often considered to be inefficient, and this seemed to be the case in our study as well. However, in this situation observational plans including serial sacrifice (killing of certain mice at prespecified times) and diagnoses of live animals based on examination of blood samples were possible alternatives. This led us to study the efficiency of such observational plans. The purpose was both to be able to design a satisfactory experiment for the leukaemia study, and to obtain some general quantitative results on the efficiency of various observational plans.

Our study is set in the context of a Markov illness-death model with one disease (Fix and Neyman, 1951, Sverdrup, 1965, Chiang, 1968). Applications of Markov illness-death models to survial/sacrifice-experiments have also been studied by Turnbull and Mitchell (1978), Mitchell and Turnbull (1979), Berlin, Brodsky and Clifford (1979), and Kodell and Nelson (1980).

The plan of the paper is as follows. In the next section we introduce the illness-death model. Section 3 gives a precise description of the observational plans, while in Section 4 we show how the estimation of the intensities of the illness-death model may be carried out. The efficiency results are presented in Section 5.

In Section 6 we outline briefly the results of the preliminary analysis on the development of leukaemia based on the simple survival experiments, discuss the working out of the improved observational plan, and give the main conclusions from the new experiment.

A more extensive discussion of the technical subjects treated in this paper is given by Borgan, Liestøl and Ebbesen (1979), and the experiment on leukaemia development is analysed in detail in Ebbesen, Borgan and Liestøl (1982).

2. The Markov illness-death model

The Markov illness-death model with one disease is illustrated in Figure 1. Live individuals are classified as either having or not having a disease A. At death, the individuals move to the corresponding "dead" state. We limit our attention to chronic diseases, i.e. diseases where no cure is possible.

The transition probability $P_{ij}(x,y)$ is the probability that an individual aged x in state i will be in state j at age $y \geq x$. The force of morbidity is then given by $\sigma(x) = \lim_{y \downarrow x} P_{01}(x,y)/(y-x)$ and similar definitions apply to the forces of mortality μ and ν. (Note that ν is the intensity of dying <u>with</u> disease A, not <u>from</u> it.) The transition probabilities are given by

$$P_{00}(x,y) = \exp\{-\int_x^y \alpha(s)ds\}, \quad P_{11}(x,y) = \exp\{-\int_x^y \nu(s)ds\},$$
$$P_{02}(x,y) = \int_x^y P_{00}(x,s)\mu(s)ds, \text{ and} \qquad (2.1)$$
$$P_{01}(x,y) = \int_x^y P_{00}(x,s)\sigma(s)P_{11}(s,y)ds,$$

where $\alpha(x) = \mu(x)+\sigma(x)$.

3. Observational plans

Although the methods and results in this paper are of interest in other connections, we have adapted our terminology to animal experiments. For all observational plans under consideration, we suppose that the study starts with a fixed number, n, of individuals of the same age (denoted 0), all in state 0. We assume that all individuals behave independently of each other.

The four observational plans considered here are as follows:

(1) Simple survival experiment. The only data recorded for each individual case are exact age at death and disease status at death.

(2) Serial sacrifice. At the start of the investigation, the animals are divided at random into K groups with n_j animals in group j ($\Sigma n_j = n$). At a prespecified age x_j, with $0 < x_1 < \ldots < x_K$, the animals still alive in group j are killed and examined. For animals which die from natural causes before this age, one observes exact age and disease status at death.

(A slightly different sacrifice design is discussed by Berlin et al. (1979). They assume that the animals sacrificed at age x_j are a randomly drawn sample of all animals alive at that age.)

(3) Periodic diagnosis. At prespecified ages $x_1, x_2, \ldots, x_K$, with $0 < x_1 < \ldots < x_K$, individuals still alive are diagnosed. When an individual dies, one observes exact age and disease status.

(4) Complete observation. The individuals under study are observed continuously, and exact age at all transitions between the states are recorded. This observational plan will be used as a yardstick for measuring the efficiencies of the three incomplete observational plans.

4. Assumptions on the intensities and parameter estimation

4.1 Introduction

Due to identifiability problems (Clifford, 1977, Berlin et al., 1979, Borgan et al., 1979, Neyman, 1980) it is impossible to adopt a non-parametric approach for the incomplete observational plans of Section 3 (i.e. plans 1-3). The identifiability problems are resolved by restricting the class of possible intensities to a suitable class of parametric functions. The appropriate way of choosing these functions will, of course, primarily depend on the prior knowledge of the situation under study. However, for the Markov illness-death model, non-trivial computational problems must also be considered before a specific parameterization is adopted. (The maximum likelihood estimators usually have to be found by a combined use of numerical maximization and integration algorithms.)

The numerical problems are reduced considerably if one adopts the "semi-parametric" assumption of piecewise constant intensities. This assumption is common in demography, epidemiology and actuarial science (Hoem, 1976) and may also be a realistic "approximating assumption" in animal experiments (Neyman, 1980). Furthermore, by successive introduction of more parameters through a finer partitioning of the studied age interval, one moves from a strong parameterization toward a "non-parametric" description. Accordingly, this assumption also seems suitable when the purpose is to illustrate the effect of the parameterization on the relative efficiencies of the different observational plans (see also Borgan et al., 1979).

4.2 Piecewise constant intensities

Assume that the total age span $[0, x_K>$ can be partitioned into K subintervals $[x_{j-1}, x_j>$, $j=1,\ldots,K$, with $x_0=0$, and that there exist parameters μ_j, σ_j, ν_j such that $\mu(x)=\mu_j$, $\sigma(x)=\sigma_j$, and $\nu(x)=\nu_j$ for

$x \in [x_{j-1}, x_j >$. Furthermore, let $\alpha_j = \mu_j + \sigma_j$, which makes $\alpha(x) = \alpha_j$ for $x \in [x_{j-1}, x_j >$. For simplicity we use the same notation for the partitioning of $[0, x_K >$ as we do for the ages at which sacrifice and diagnosis occur in the observational plans 2 and 3 of Section 3. (In this paper sacrifice and diagnosis are assumed to be carried out at the partitioning ages.)

Now, for $t \leq l_j$, with $l_j = x_j - x_{j-1}$, (2.1) gives

$$P_{00}(x_{j-1}, x_{j-1}+t) = \exp(-\alpha_j t), \quad P_{11}(x_{j-1}, x_{j-1}+t) = \exp(-\nu_j t),$$

$$P_{02}(x_{j-1}, x_{j-1}+t) = \mu_j \{1-\exp(-\alpha_j t)\}/\alpha_j, \tag{4.1}$$

and for $\alpha_j \neq \nu_j$

$$P_{01}(x_{j-1}, x_{j-1}+t) = \sigma_j \{\exp(-\nu_j t) - \exp(-\alpha_j t)\}/(\alpha_j - \nu_j).$$

Furthermore,

$$P_{00}(0, x_{j-1}+t) = P_{00}(0, x_{j-1}) P_{00}(x_{j-1}, x_{j-1}+t) ,$$

and (4.2)

$$P_{01}(0, x_{j-1}+t) = P_{01}(0, x_{j-1}) P_{11}(x_{j-1}, x_{j-1}+t) + P_{00}(0, x_{j-1}) P_{01}(x_{j-1}, x_{j-1}+t) .$$

Hence, $P_{00}(0,x)$ and $P_{01}(0,x)$ may be found for any value of x by successive application of (4.1) and (4.2).

4.3 Maximum likelihood estimation

In this subsection we will outline how the maximum likelihood estimators may be found, and how one may derive their asymptotical distributional properties.

(1) Simple survival experiment. Contributions to the likelihood by individuals who die with and without the disease are easily obtained from equations (4.1) and (4.2). An individual who dies at

age $V \in [x_{j-1}, x_j >$ contributes an amount $\nu_j P_{01}(0,V)$ to the likelihood if it dies with the disease, otherwise its contribution is $\mu_j P_{00}(0,V)$. The maximum likelihood estimators $\mu_1^*,\ldots,\mu_K^*,\sigma_1^*,\ldots,\sigma_K^*$, $\nu_1^*,\ldots,\nu_K^*$ can be found by numerical methods. Moreover, it follows by standard maximum likelihood theory, that, as the number of individuals increases to infinity, the estimators are asymptotically multinormally distributed with the proper expectations, and with an asymptotic covariance matrix which may be found as shown in the Appendix.

(2) Serial sacrifice. The animals who die from natural causes give contributions to the likelihood similar to those given above. An animal in the jth group which is sacrificed contributes $P_{01}(0,x_j)$ if it suffers from the disease, $P_{00}(0,x_j)$ if it is disease-free. Again the maximum likelihood estimators $\tilde{\mu}_1,\ldots,\tilde{\nu}_K$ may be found by numerical methods, and their asymptotic properties may be found by calculations parallel to those outlined in the Appendix.

(3) Periodic diagnosis. Here the total likelihood may be written as a product of K factors, where each factor only contains the three parameters belonging to the age interval between two examinations. The contributions to the factor corresponding to the jth age interval are the following: An individual who is disease-free at age x_{j-1} and alive at age x_j contributes $P_{00}(x_{j-1},x_j)$ if it is disease-free at age x_j, $P_{01}(x_{j-1},x_j)$ otherwise. If the individual is disease-free at x_{j-1} and dies at age $V \in [x_{j-1},x_j >$ it gives a contribution $\mu_j P_{00}(x_{j-1},V)$ if it is disease-free, $\nu_j P_{01}(x_{j-1},V)$ otherwise. An individual who has the disease at age x_{j-1} contributes $P_{11}(x_{j-1},x_j)$ if it is still alive at age x_j and $\nu_j P_{11}(x_{j-1},V)$ if it dies at age $V \in [x_{j-1},x_j>$. Also in this situation the maximum likelihood estimators $\hat{\mu}_1,\ldots,\hat{\nu}_K$ may be found by numerical methods. Their asymptotic properties may be found by standard maximum likelihood theory.

(4) Complete observation. For the complete observational plan the maximum likelihood estimators $\hat{\mu}_1,\ldots,\hat{\nu}_K$ are the usual occurrence/exposure rates whose asymptotic properties are well-known (see e.g. Hoem, 1976).

5. Efficiencies of the observational plans

In the following comparison of the observational plans, our discussion focuses on the relative efficiencies. As an efficiency measure we use the usual ratio between the asymptotic variances of the estimators for the complete plan and the estimators for the three incomplete plans, i.e. the asymptotic relative efficiency. (Note that all efficiencies are computed relative to the number of individuals, such that the amount of work needed for each individual is not taken explicitly into account.) The efficiencies will depend on the partitioning of the total age span and on the values of the intensities through the products $\mu_j l_j$, $\sigma_j l_j$ and $\nu_j l_j$, where l_j is the length of the jth age interval. Nevertheless, our study indicates that the general tendencies are adequately illustrated by means of the following examples.

We examine a situation where one is interested in estimating the intensities for a one year period. Thus, we assume that data are collected during this year. For the simple survival experiment, we also assume that deaths which occur after this year are observed (cf. Section 3), and in the computations for this observational plan, the intensities are always assumed to be constant after the first year. Efficiencies of the three incomplete observational plans and asymptotic variances for the complete sampling scheme are given in Table 1. The period from 0 to 12 months is divided into one, two, three, or four equally long intervals, and all intensities are taken to be equal to 0.1 when time is measured in months. Sacrifice and

diagnosis are carried out at the endpoint of each age interval. For the observational plan with serial sacrifice the numbers of animals in each group were chosen in the ratios 1:4, 1:4:9 and 1:4:9:16 for two, three and four intervals, respectively. These ratios were chosen to make the efficiencies approximately equal in all age intervals.

The most striking result of Table 1 is the finding of very low efficiencies for the simple survival experiment, especially for the σ_j^*s. Note that even for the case with four age intervals, the probability of dying during a given interval is as high as 26 per cent such that this is not a very fine partitioning. Accordingly, the decrease in the efficiencies towards the nonidentifiability of the nonparametric case is rapid. The efficiencies for the σ_j^*s may be somewhat higher for other situations, but this is normally accompanied by a decrease in the efficiencies of the ν_j^*s. One example of this effect is shown in Table 2, where the situation is as in Table 1.d, with the exception that the ν_js are higher, corresponding to a more severe disease. We have studied other situations such as with a more or less common disease, or with increasing and decreasing intensities, but as long as the values of products $\mu_j l_j$, $\sigma_j l_j$, and $\nu_j l_j$ do not change too much, the efficiencies will be of the same order of magnitude.

In the lower left-hand part of Figure 2 we give the correlations between the estimators for the simple survival experiment for the situation of Table 1.d. (Note that for the simple survival experiment, the estimators for the first year are influenced by events occurring after this year.) The correlations are generally high. Note for example that all μ_j^*s are positively correlated with each other. The same is valid for the ν_j^*s, while the correlations between the μ_j^*s and the ν_j^*s are negative. This implies that the difference between the μ_js and ν_js may be either over- or under-

estimated in all age intervals, i.e., an overestimation (underestimation) of the difference in one interval is not "compensated" by an underestimation (overestimation) in the following interval. For the σ_j^*s, which are negatively correlated with each other, some "compensation" will occur, such that the integral over more than one interval will be relatively better estimated than the intensity for each of the intervals.

In studies using laboratory animals, an experiment with serial sacrifice will cause little extra work, and may even reduce the workload of the experimenter. Table 1 shows that it will also increase the efficiencies considerably. As shown in the upper right-hand part of Figure 2 the correlations between the estimators are far less than for the simple survival experiment. The efficiencies will depend on the ratios between the number of animals in each sacrifice group. By choosing an appropriate sacrifice design, one may obtain relatively high efficiencies for especially interesting age intervals. To illustrate this, Table 3 gives the efficiencies for two sacrifice designs for the situation of Table 1.d.

As illustrated, if no other factors in the experimental situation strongly motivate a simple survival experiment, a change to an observational plan with serial sacrifice is desirable. This, however, should not be permitted to overshadow the fact that even a plan with serial sacrifice may have rather low efficiencies. As revealed by Table 1, much is gained by changing to an observational plan with periodic diagnosis. When the intensities are not too high, and periodic diagnosis can be repeated at least three to four times, the loss in efficiency for this plan compared with the complete sampling scheme is negligible. Also, the correlations between the estimators are insignificant. (All were less than 1 per cent for the situation of Table 1.d.) The gain in efficiency when a sampling scheme with periodic diagnosis is used, must be evaluated against the extra work

needed per animal. It seems, however, that this observational plan ought to be used when complete observation is impossible, while diagnoses of live animals are possible, and the results from these not significantly less certain than those of post mortem examinations.

6. Development of lymphatic leukaemia in mice

A specific strain (AKR) of inbred mice shows a very high incidence of spontaneous lymphatic leukaemia, and this strain has therefore been widely used in studies of leukaemia development and treatment. In a sequence of investigations on leukaemia development in these mice, it was observed that the mean life spans of untreated mice dying with and without leukaemia were virtually equal (Ebbesen,1978). An explanation of this finding might be that the mice contract leukaemia in a limited age span, so that old animals are "safe" (Borgan et al., 1979). In view of the usual age-dependence of cancer incidence (see e.g. Doll, 1978), and the fact that the mice are strongly inbred and live in an unchanging environment, this explanation might provide interesting information on the mechanisms behind the development of leukaemia.

The above mentioned observation on life span was based on simple survival experiments including data from histological examinations of about 600 animals. We used these data to estimate the age-dependent leukaemia intensities for male and female AKR-mice. The analyses were carried out as described in Section 4.3. The estimated leukaemia risks were close to zero below an age of 4-5 months, but then increased sharply and reached a maximum at about an age of 8 months. For ages above one year they were again close to zero for both sexes (for details, see Borgan et al., 1979). In view of the results of Section 5, it is, however, evident that the estimates are

highly uncertain. We therefore decided to carry out a new experiment based on an improved observational plan.

The new experiment was designed primarily to provide reliable estimates of the leukaemia risk for untreated female mice, and in particular to determine whether the decreased intensity for higher ages was a real phenomenon. Moreover, since observational plans including periodic diagnosis are quite uncommon in animal experiments, it was of methodological interest to see whether such a design was applicable in the present situation. It is possible to obtain blood samples from a mouse several times without affecting it significantly. Thus, one can search blood smears for malignant cells and obtain counts of white blood cells. In addition, enlarged organs can detected by palpation. After some preliminary studies, we decided that these sources of informataion provided a possible way to diagnose the animals. The experiment was therefore based on a design with periodical diagnosis.

To determine the number of animals needed and the suitable ages for the periodic diagnoses, we used the results from the simple survival experiments, efficiency calculations similar to those given in Section 5, and some simulation studies. Since the results from the analysis of the simple survival experiments were uncertain, various sets of possible values of the intensities were tried out. For instance, we found that approximately 300 animals would be needed to detect a decrease in the leukaemia intensity from 0.5 at 7-8 months to 0.2 at 9-12 months with a probability of 95 per cent at the 5 per cent level (when the other intensities were assumed to be approximately as estimated from the simple survival experiments). Based on such considerations we decided to use about 500 female mice. The mice entered the study at the age of 3 months and were diagnosed (if still alive) at the ages 4, $5\frac{1}{2}$, 7, 9, 13 and 18 months. To obtain information as to how often a diagnosis based on examination of live

animals was at variance with a diagnosis obtained by histologic examination, 20 prespecified animals were sacrificed (if still alive) at each of the ages 4, $5\frac{1}{2}$, 7 and 9 months. All animals (7) still alive at the age of 18 months were sacrificed, and 9 mice were killed by accident.

It turned out that the periodic examinations resulted in some obviously erroneous diagnoses of cancer, especially in the early phase of the experiment. To minimize such errors a "blind" reexamination of nearly 100 out of the approximately 1700 blood slides was carried out. All cases with inconsistent or dubious results were included.

As could be expected, the results showed that the diagnosis of live animals detected leukaemia at a somewhat later stage than the histologic examinations. For a more detailed evaluation of the experimental design and the quality of the data, see Ebbesen et al. (1982). It should be stated here, however, that if we could have reconstructed the experimental design, we would have included larger groups for sacrifice at higher ages (9 months and older).

The data, based on diagnoses at death similar to those performed on live animals, are summarized in Table 4. (The data based on histologic examinations at death give 33 more cases of death with leukaemia, see Ebbesen et al., 1982.) The relatively high number of animals that have disappeared or have not been diagnosed at death is mostly due to rapid decay of animals following death, and cannibalism. However, with the present observational plan these animals still provide significant information.

The estimated intensities are given in Table 5, and the estimated leukaemia risk is illustrated in Figure 3. The estimated leukaemia intensity shows a marked peak around 7-9 months. Moreover, as seen by the estimated standard deviations, the fall in the leukaemia risk at higher ages is highly significant. We also note the high and

fairly constant death risk for mice with leukaemia. (Our estimates of this death intensity would, however, have been somewhat lower if the diagnosis of live animals could detect cancer at as early a stage as histologic examinations are capable of.) The rapid changes in the leukaemia risk imply that the assumption of piecewise constant intensities is not completely satisfactory, but the main pattern should nevertheless be fairly well illustrated, and the estimates should provide a fair basis for further experimental research.

For simple versions of the multistep hypothesis for cancer development (e.g. Doll, 1978), one may find mathematical expressions representing the age-dependence of the intensity. If it is assumed that the number of potentially malignant cells as well as the transition rates for each of the specific steps in each cell are independent of age, then the leukaemia intensity will (approximately) be of the form $\sigma(x)=k(x-a)^n$. Here, x is age and a is the minimum age at which development of leukaemia may start (Moolgavkar, 1978). The declining intensity for older AKR mice is clearly not in accordance with this strictly increasing functional form. However, in the present case one may assume that the intensity is affected by age-related changes such as loss of cells capabel of malignant changes (e.g. loss of thymocytes) or age-dependent changes in immune response. Moreover, assume that there is some randomly occuring event (e.g. in connection to the integration processes of viral DNA) which takes place in a limited age span, and which permanently changes either the number of potentially malignant cells or the transition rates for the steps in the multistage process. Then the simple model for the multistep hypothesis should be changed such that k in the expression for $\sigma(x)$ varies between the animals. To illustrate the possible effect of such a variation, we may assume that the value of k is, for each mouse, the outcome of a random variable K. This random variable is assumed to be zero with probability p. Given

that $K>0$, it follows a gamma distribution with mean $k_o/(1-p)$ and variance $k_o^2/[(1-p)^2\gamma]$, such that the expected value of K is k_o. Now, the functional form of the leukaemia intensity on the population level may be derived and this function may be fitted to the estimates of Table 5 by the modified minimum chi-square method (Hoem, 1976) for various integer valued n. (It is possible to fit the model directly to the data, but for the present purpose we have found this much simpler procedure satisfactory.) Figure 3 shows the result, and the model is seen to fit nicely. Thus, there are certain mathematically small and biologically plausible changes in the assumptions of the simple multistage model which imply a leukaemia intensity similar to the one observed.

ACKNOWLEDGEMENTS

The authors are grateful to Jan M. Hoem and Niels Keiding for helpful comments and discussions. Ørnulf Borgan and Knut Liestøl were supported by The Norwegian Research Council for Science and the Humanities. Ørnulf Borgan was also supported by The Association of Norwegian Insurance Companies. The experiment reported in Section 6 of the paper was financed by The Hafnia-Haand-i-Haand fund.

REFERENCES

Berlin, B., Brodsky, J. and Clifford, P. (1979). Testing disease dependence in survival experiments with serial sacrifice. Journal of the American Statistical Association 74, 5-14.

Borgan, Ø., Liestøl, K. and Ebbesen, P. (1979). Observational plans for the analysis of disease development and cause specific mortality. Working Paper No. 25, Laboratory of Actuarial Mathematics, University of Copenhagen.

Chiang, C.L. (1968). Introduction to stochastic processes in biostatistics. John Wiley & Sons, New York.

Clifford, P. (1977). Nonidentifiability in stochastic models of illness and death. Proceedings of the National Academy of Sciences 74, 1338-1340.

Doll, R. (1978). An epidemiological perspective of the biology of cancer. Cancer Research 38, 3573-3583.

Ebbesen, P. (1978). Life span, leukaemia and amyloid incidences of untreated and polycation treated AKR mice. British Journal of Cancer 37, 76-80.

Ebbesen, P., Borgan, Ø. and Liestøl, K. (1982). Decreasing leukemia intensity in old AKR mice. (Manuscript).

Fix, E. and Neyman, J. (1951). A simple stochastic model of recovery, relapse, death and loss of patients. Human Biology 23, 205-241.

Hoem, J.M. (1976). The statistical theory of demographic rates. A review of current developments (with discussion). Scandinavian Journal of Statistics 3, 169-185.

Kodell, R.L. and Nelson, C.J. (1980). An illness-death model for the study of the carcinogenic process using survival/sacrifice data. Biometrics 36, 267-277.

Mitchell, T.J. and Turnbull, B.W. (1979). Log-linear models in the analysis of disease prevalence data from survival/sacrifice experiments. Biometrics 35, 221-234.

Moolgavkar, S.H. (1978). The multistage theory of carcinogenesis and the age distribution of cancer in man. Journal of the National Cancer Institute 61, 49-52.

Neyman, J. (1980). Some memorable incidents in probabilistic/statistical studies. In Asymptotic theory of statistical tests and estimation, ed. I.M. Chakravarti, pp. 1-32, Academic Press New York.

Sverdrup, E. (1965). Estimates and test procedures in connection with stochastic models for deaths, recoveries and transfers between different states of health. Skandinavisk Aktuarietidskrift 48, 184-211.

Turnbull, B. and Mitchell, T.J. (1978). Exploratory analysis of disease prevalence data from survival/sacrifice experiments. Biometrics 34, 555-570.

APPENDIX

We here show how one may compute the asymptotic distribution of the maximum likelilhood estimators from a simple survival experiment. The computations for the observational plans 2 and 3 of Section 3 follow a similar scheme, see Borgan et al. (1979).

We introduce the following quantities:
Let N_j^{kl} be the number of transitions (0 or 1) direct from state k to state l experienced by a specific animal in the age interval $[x_{j-1},x_j>$, for $(k,l)=(0,2),(1,3)$. Let V_j^0 be the total time spent in state 0 by this animal in $[x_{j-1},x_j>$ if it dies without the disease A sooner or later. Let $V_j^0 = 0$ otherwise. Furthermore let V_j^L be the total time spent by this animal in the states $L=\{0,1\}$ in the jth interval if it dies with the disease sooner or later; $V_j^L = 0$ otherwise. Then the likelihood for this animal is

$$\Lambda = \prod_{j=1}^{K} \{\mu_j^{N_j^{02}} \nu_j^{N_j^{13}}\} \exp\{-\sum_{j=1}^{K} \alpha_j V_j^0\} P_{01}(0,V_\cdot^L)^{N_\cdot^{13}},$$

where $N_\cdot^{13} = \sum_{j=1}^{K} N_j^{13}$ and $V_\cdot^L = \sum_{j=1}^{K} V_j^L$. It follows by standard maximum likelihood theory that $(\mu_1^*,\ldots,\nu_K^*)$ is asymptotically multinormally distributed with the proper expectations and

$$\text{as.covm}(\mu_1^*,\ldots,\nu_K^*) = -\frac{1}{n}\{E\,\frac{\partial^2 \ln\Lambda}{\partial\theta\partial\rho}\}^{-1}_{\theta,\rho=\mu_1,\ldots,\nu_K}. \qquad (A.1)$$

Some calculation gives

$$E\frac{\partial^2 \ln\Lambda}{\partial\theta\ \partial\rho} = -\delta_{\theta\mu_j}\delta_{\rho\mu_j} EN_j^{02}/\mu_j^2 - \delta_{\theta\nu_j}\delta_{\rho\nu_j} EN_j^{13}/\nu_j^2 + E\{N_\cdot^{13} f(V_\cdot^L;\theta,\rho)\} \qquad (A.2)$$

for $\theta,\rho = \mu_1,\ldots,\nu_K$, where δ_{xy} is the Kronecker delta, and

$$f(v;\theta,\rho) = [\frac{\partial^2}{\partial\theta\partial\rho}\{P_{01}(0,v)\}P_{01}(0,v) - \frac{\partial}{\partial\theta}\{P_{01}(0,v)\}\frac{\partial}{\partial\rho}\{P_{01}(0,v)\}]/\{P_{01}(0,v)\}^2 .$$

Here

$$EN_j^{02} = P_{00}(0,x_{j-1})P_{02}(x_{j-1},x_j), \tag{A.3}$$

$$EN_j^{13} = P_{01}(0,x_{j-1})P_{13}(x_{j-1},x_j) + P_{00}(0,x_{j-1})P_{03}(x_{j-1},x_j), \tag{A.4}$$

and

$$E\{N_\cdot^{13} f(V_\cdot^L;\theta,\rho)\} = \sum_{j=1}^{K} \nu_j \int_0^{l_j} f(x_{j-1}+t;\theta,\rho)P_{01}(0,x_{j-1}+t)dt. \tag{A.5}$$

The integrals in (A.5) must be computed by numerical methods. For this we need the first and second order derivatives of $P_{01}(0,x_{j-1}+t)$ with $t \leq l_j$ for each j. By (4.2), $\frac{\partial}{\partial\theta} P_{01}(0,x_{j-1}+t)$ equals

$$\frac{\partial}{\partial\theta} P_{01}(0,x_{j-1})P_{11}(x_{j-1},x_{j-1}+t) + \frac{\partial}{\partial\theta} P_{00}(0,x_{j-1})P_{01}(x_{j-1},x_{j-1}+t)$$

for $\theta = \mu_m$, σ_m or ν_m with $m \leq j-1$; it equals

$$P_{01}(0,x_{j-1}) \frac{\partial}{\partial\theta} P_{11}(x_{j-1},x_{j-1}+t) + P_{00}(0,x_{j-1}) \frac{\partial}{\partial\theta} P_{01}(x_{j-1},x_{j-1}+t), \tag{A.6}$$

for $\theta = \mu_j$, σ_j or ν_j; and it is 0 otherwise.

Moreover, $\frac{\partial^2}{\partial\theta\partial\rho} P_{01}(0,x_{j-1}+t)$ equals

$$\frac{\partial^2}{\partial\theta\partial\rho} P_{01}(0,x_{j-1})P_{11}(x_{j-1},x_{j-1}+t) + \frac{\partial^2}{\partial\theta\partial\rho} P_{00}(0,x_{j-1})P_{01}(x_{j-1},x_{j-1}+t),$$

for $\theta,\rho = \mu_1,\ldots,\mu_{j-1},\sigma_1,\ldots,\sigma_{j-1},\nu_1,\ldots,\nu_{j-1}$; it equals

$$\frac{\partial}{\partial\theta} P_{01}(0,x_{j-1}) \frac{\partial}{\partial\rho} P_{11}(x_{j-1},x_{j-1}+t) + \frac{\partial}{\partial\theta} P_{00}(0,x_{j-1}) \frac{\partial}{\partial\rho} P_{01}(x_{j-1},x_{j-1}+t), \tag{A.7}$$

for $\theta=\mu_m,\sigma_m$ or ν_m with $m \leq j-1$ and $\rho=\mu_j,\sigma_j$ or ν_j; it is equal to

$$P_{01}(0,x_{j-1}) \frac{\partial^2}{\partial\theta\partial\rho} P_{11}(x_{j-1},x_{j-1}+t) + P_{00}(0,x_{j-1}) \frac{\partial^2}{\partial\theta\partial\rho} P_{01}(x_{j-1},x_{j-1}+t),$$

for $\theta,\rho = \mu_j$, σ_j or ν_j; and it is 0 otherwise.

Consequently, the first and second order derivatives of $P_{01}(0,x_{j-1}+t)$ may be found for any $t \leq l_j$; and any j by successive application of (A.6) and (A.7). In these computations, we need formulas for the derivatives of $P_{00}(0,x_{j-1})$, $P_{11}(x_{j-1},x_{j-1}+t)$ and $P_{01}(x_{j-1},x_{j-1}+t)$. These are found from (4.1) and (4.2) by straightforward differentiation, and are not given here. Having found the derivatives of $P_{01}(0,x_{j-1}+t)$ the expressions in (A.5) may be evaluated by numerical integration. Combining this with (A.3) and (A.4) we find by (A.2) $E \frac{\partial^2 \ln \Lambda}{\partial \theta \partial \rho}$ for $\theta,\rho = \mu_1,\ldots,\nu_K$, and, hence, by (A.1) the asymptotic covariance matrix of $\mu_1^*,\ldots,\nu_K^*$ results.

Table 1. Asymptotic relative efficiencies for the incomplete observational plans.

Age in whole months	Simple survival experiment			n·asymptotic variance a) Serial sacrifice			Periodic diagnosis			Complete observation		
	μ^*	σ^*	ν^*	$\tilde{\mu}$	$\tilde{\sigma}$	$\tilde{\nu}$	$\overset{\vee}{\mu}$	$\overset{\vee}{\sigma}$	$\overset{\vee}{\nu}$	$\hat{\mu}$	$\hat{\sigma}$	$\hat{\nu}$
				a. One age interval								
0-11	.509	.238	.357	.904	.904	.835	.904	.904	.835	.022	.022	.041
				b. Two age intervals								
0-5	.448	.065	.191	.817	.299	.565	.972	.972	.911	.029	.029	.098
6-11	.204	.064	.257	.626	.203	.664	.974	.974	.981	.095	.095	.070
				c. Three age intervals								
0-3	.448	.031	.138	.783	.128	.415	.987	.987	.939	.036	.036	.184
4-7	.168	.034	.137	.641	.093	.591	.988	.988	.985	.081	.081	.103
8-11	.167	.027	.251	.551	.130	.584	.988	.988	.993	.180	.180	.108
				d. Four age intervals								
0-2	.461	.019	.113	.768	.070	.331	.993	.993	.953	.044	.044	.298
3-5	.157	.021	.093	.617	.053	.476	.993	.993	.987	.081	.081	.147
6-8	.126	.017	.136	.647	.069	.659	.993	.993	.994	.147	.147	.135
9-11	.161	.014	.259	.477	.089	.501	.993	.993	.996	.268	.268	.147

a) The sacrifice designs in panels b - d are 1:4, 1:4:9, and 1:4:9:16, respectively. The notation 1:4 means that 1/5 of the animals are put in group No. 1 and 4/5 in group No. 2. Compare Section 3.

Table 2. Asymptotic relative efficiencies for the simple survival experiment, for three values of the ν_j's.

Values of (μ_j,σ_j,ν_j) [a]	Estimators											
	μ_1^*	μ_2^*	μ_3^*	μ_4^*	σ_1^*	σ_2^*	σ_3^*	σ_4^*	ν_1^*	ν_2^*	ν_3^*	ν_4^*
(.1,.1,.1)	.461	.157	.126	.160	.019	.021	.017	.014	.113	.093	.136	.258
(.1,.1,.25)	.462	.148	.117	.123	.019	.043	.038	.032	.042	.026	.032	.043
(.1,.1,.5)	.559	.239	.240	.194	.028	.135	.115	.119	.026	.015	.018	.025

a) The same values apply to all age intervals

Table 3. Asymptotic relative efficiencies for the observational plan with serial sacrifice for two sacrifice designs.

Sacrifice design [a]	Estimators											
	$\tilde{\mu}_1$	$\tilde{\mu}_2$	$\tilde{\mu}_3$	$\tilde{\mu}_4$	$\tilde{\sigma}_1$	$\tilde{\sigma}_2$	$\tilde{\sigma}_3$	$\tilde{\sigma}_4$	$\tilde{\nu}_1$	$\tilde{\nu}_2$	$\tilde{\nu}_3$	$\tilde{\nu}_4$
1:4:9:16	.768	.617	.647	.477	.070	.053	.069	.089	.331	.476	.659	.501
1:1:1:1	.944	.655	.437	.229	.229	.115	.074	.053	.716	.594	.442	.238

a) The notion 1:4:9:16 means that 1/30 of the animals are in group no.1, 4/30 in group 2, etc. Compare Section 3.

Table 4. Survey of data from an experiment with periodic diagnosis with female AKR mice

Number of animals	Age in months					
	3-4	4-5½	5½-7	7-9	9-13	13-18
Alive without leukaemia[a)]	477	425	287	118	31	6
Alive with leukaemia[a)]	1	10	15	13	1	1
Sacrificed without leukaemia[b)]	22	19	15	4	2	6
Sacrificed with leukaemia[b)]	0	0	3	6	0	1
Occurrence of leukaemia, 0→1[c)]	1	10	15	12	1	0
Death without leukaemia, 0→2[c)]	0	2	14	8	14	5
Occurrence of and death with leukaemia, 0→3[c)]	0	10	53	102	45	3
Death with leukaemia, 1→3[c)]	0	1	7	10	6	1
Disappeared or not diagnosed[c)]	2	8	40	33	24	14

a) Number alive at the end of the age interval and before sacrifice.

b) Number sacrificed at the end of the age interval.

c) Number of occurrences during the age interval. The notation 0→1 etc. refers to Fig. 1.

Table 5. Estimated death intensities and leukaemia risk for female AKR mice with one standard deviation based on an experiment with periodic diagnosis.

Intensity	Age in months					
	3-4	4-5½	5½-7	7-9	9-13	13-18
Death without leukaemia	.000 -	.004 ±.003	.036 ±.009	.031 ±.010	.068 ±.017	.188 ±.053
Leukaemia risk	.002 ±.002	.034 ±.007	.162 ±.018	.381 ±.033	.231 ±.032	.108 ±.053
Death with leukaemia	-	1.050 ±.338	2.124 ±.309	2.354 ±.274	1.819 ±.293	.543 ±.322

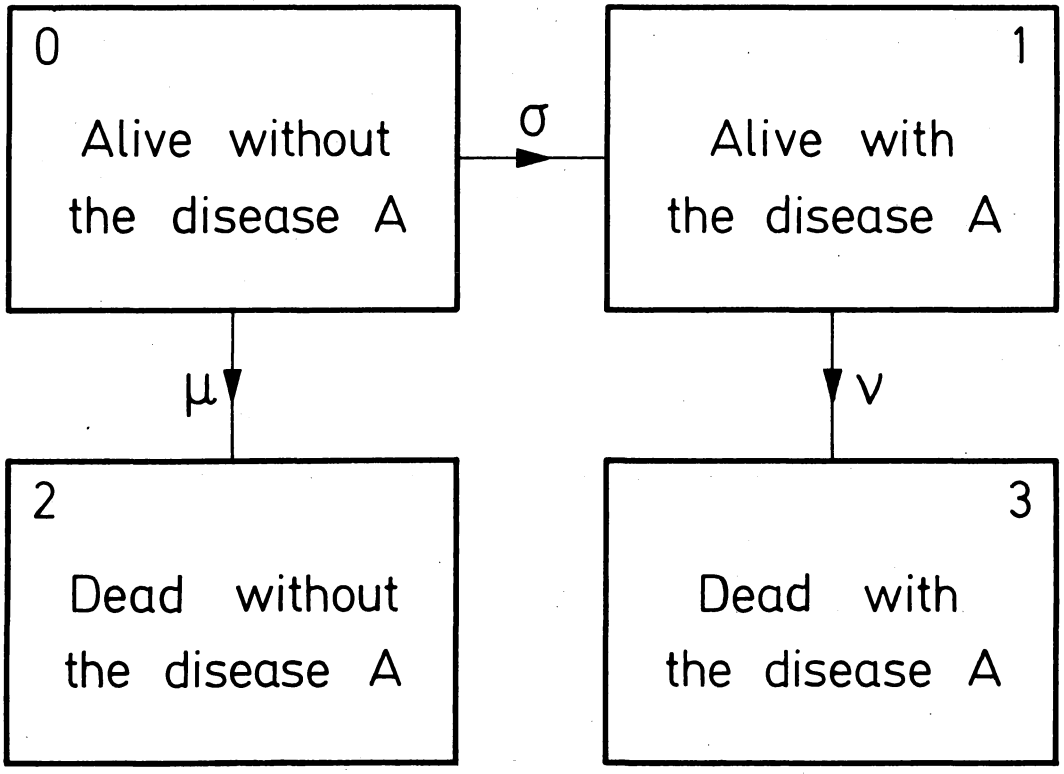

Figure 1. The progressive Markov illness-death model with one disease.

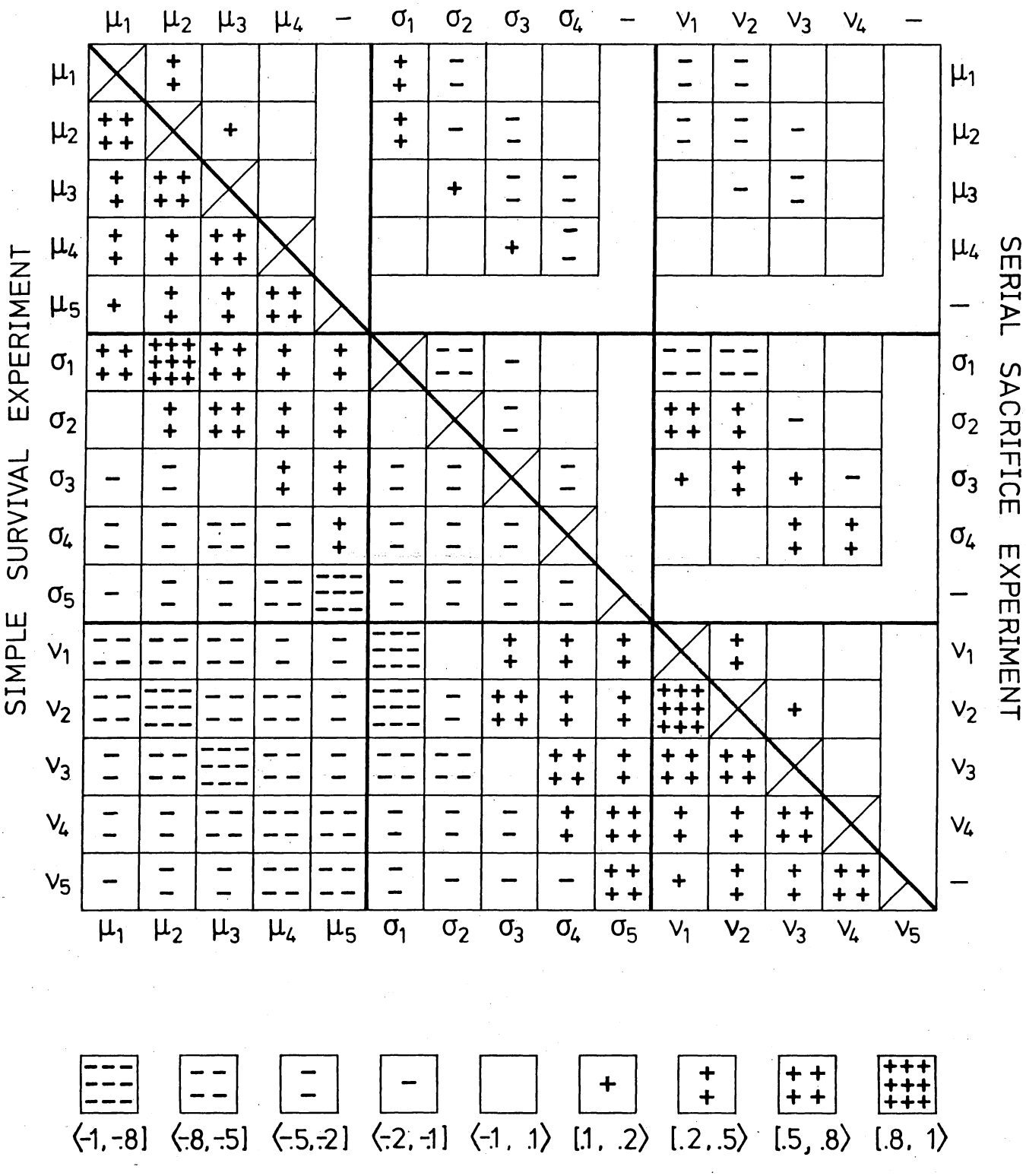

Figure 2. Correlations between the estimators for the simple survival experiment and for the experiment with serial sacrifice for the situation of Table 1.d.

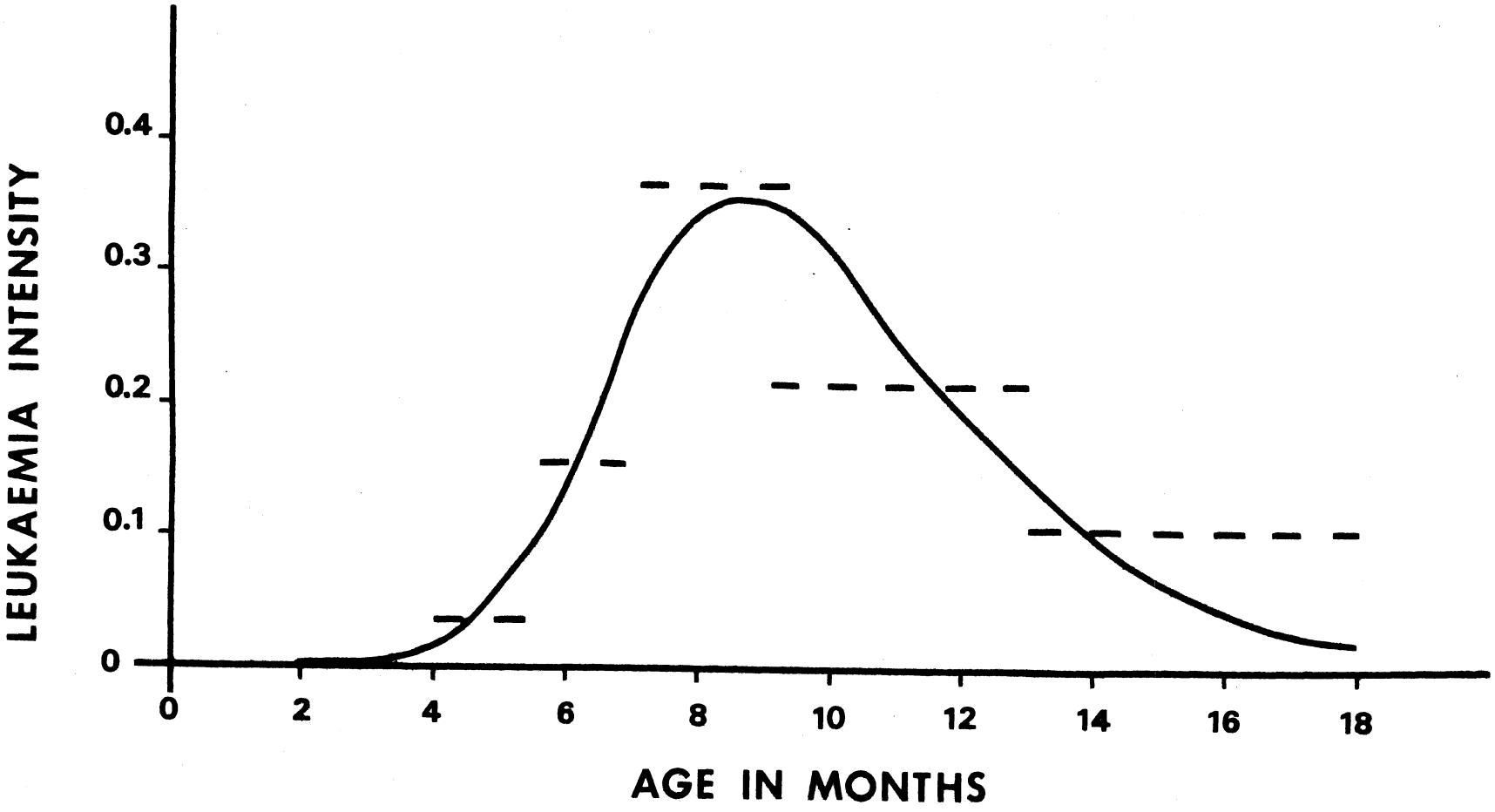

Figure 3. Estimated leukaemia risk according to Table 5 (-----) and fitted curve based on an inhomogeneity assumption (———) ($k_o = 1.0 \cdot 10^{-4}$, $p = 0.09$, $\gamma = 0.71$, $a = 1.59$, $n = 5$).