



THE QUESTIONABLE ASSOCIATION OF VITAMIN E SUPPLEMENTATION AND MORTALITY – INCONSISTENT RESULTS OF DIFFERENT META-ANALYTIC APPROACHES

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Abstract – Medical research suggests benefits of vitamin E supplementation in treatment or prevention of cardiovascular disease, inflammatory joint diseases and cancer. Regardless of these benefits in a recently published meta analysis the authors drew the conclusion that high dose supplementation may cause a slight increase in mortality of the treated patients. The purpose of the present paper is to re-analyse the association of vitamin E supplementation and mortality. By means of augmented data sources as well as additional methodological approaches the results of the above mentioned meta analysis is to be either confirmed or called into question. In the above mentioned meta analysis 19 clinical trials comprising a total of 135967 participants were included. The dosages of vitamin E supplementation ranged from 16.5 to 2000 IU/d. In the present paper this data source was augmented and 10 additional trials were included (2495 additional participants receiving vitamin E doses from 136 to 5000 IU/d). Moreover in 2 of the originally included trials updated results of mortality at longer periods of follow-up were available. The present paper yields contradictory results regarding the association of vitamin E supplementation and mortality. Hierarchical logistic regression analyses confirm the former results showing an increased mortality of patients receiving high dose vitamin E. Furthermore a traditional methodological approach of meta-regression was applied to the same data source. Contrary to the former result it showed that the increased mortality odds ratio in certain trials is not due to the higher dose of vitamin E supplementation. Rather it can be explained by a higher proportion of male patients that were included in these trials compared to other trials. The causal relationship of vitamin E supplementation and increased mortality is questionable. Different methodological approaches of meta analysis yield contradictory results. Thus none of these results can be regarded to supply evidence in a statistical sense. In particular high dose vitamin E supplementation can not be regarded proved to increase mortality.

Key words: Vitamin E supplementation, all-cause mortality, meta analysis, meta regression, hierarchical logistic regression

INTRODUCTION

Medical research suggests beneficial effects of vitamin E supplementation in prevention of cardiovascular disease and cancer as well as treatment of inflammatory joint diseases and many others. Regardless of these benefits the authors of a recently published meta analysis draw the conclusion that high dose supplementation of vitamin E may cause a slight increase in mortality of the treated patients (22). In hierarchical logistic regression analyses the pooled all-cause mortality risk difference in high-

dose vitamin E trials compared to untreated patients was 39 per 10000 persons (95% CI, 3 to 74 per 10000 persons, $p=0.035$). A dose-response analysis by means of a quadratic-linear spline model showed a significant relationship between vitamin E dosage and all-cause mortality, with increased risk of dosages greater than 150 IU/d. The results obtained proved to be relatively stable under adjustment for covariates (gender distribution of trial participants, mean age, combination of vitamin E with other vitamins or minerals, average period of follow-up), so biased effect estimates due to confounding could be ruled out.

Abbreviations: IU/d, international units per day; CI, confidence interval; OR, odds ratio

The published results of Miller et al. (22) have been discussed highly controversially in circles of experts. Among the doubts that were

addressed regarding its validity the most important were the low general sanitary state of most trial participants as well as their old age. In neither of all trials included in the meta analysis all-cause mortality represented the primary end point. Furthermore individual trials had highly different sample sizes, so that a few single trials possibly exert a dominating influence on the final result of the meta analysis. The observed large heterogeneity of individual trial results may be explained by confounding covariates not included in the model. Furthermore the definition of high dosage vitamin E supplementation (≥ 400 IU/d) assumedly was not specified prior to the statistical analysis. Rather it seems to be chosen arbitrarily or data driven. Altogether the results obtained by Miller et al. (22) seem to be questionable and possibly can not be regarded representative.

The purpose of the present paper is to re-analyse the association of vitamin E supplementation and mortality. By means of augmented data sources as well as additional methodological approaches the results of the published meta analysis are to be either confirmed or called into question.

MATERIALS AND METHODS

In the meta analysis of Miller et al. (22) 19 clinical trials comprising a total of 135967 participants were included, where information on vitamin E supplementation and all-cause mortality was available. The trial selection process is documented in Miller et al. (22). In the present paper this data source was augmented and 10 additional trials were included (2495 additional participants). Moreover in 2 of the originally included trials updated results of mortality at longer periods of follow-up were available. Including the additional trials dosages of vitamin E supplementation ranged from 16.5 to 5000 IU/d. Table 1 shows the data source underlying the statistical analysis of the present paper.

In order to perform the statistical evaluation of meta analytic data several methodological approaches have been proposed in the literature. Comprehensive overviews of traditional approaches are presented in (24, 16, 31), including fixed effects models, random effects models and methods of meta regression of univariate effect measures. Beyond these traditional approaches in case of binary outcome variables hierarchical logistic regression models can be applied alternatively (see 28). In this case the outcome is modelled by means of a bivariate approach, i.e. the mortality risk in different treatment groups within a certain trial is modelled separately (but statistically dependent). In order to analyse the dose-response relationship of vitamin E supplementation and mortality both above basic approaches can either be formulated by means of a categorical model or by a continuous model. In the first case a certain fixed cutpoint separating high and low dosages has to be determined. A continuous model is

recommended to be formulated by means of a quadratic-linear spline proposed by Greenland (11).

Theoretical aspects of the applied statistical approaches are outlined in the appendix (see below). Data analyses of the present paper were performed using SAS version 8.2 (SAS Institute, Cary, NC), HLM 6.0 (Scientific Software International, Lincolnwood, Illinois), and S-PLUS version 6.1 (Insightful, Seattle, Washington). All statistical analyses are intended to be exploratory. P-values are not interpreted in confirmatory sense. No adjustment for multiplicity is performed, thus providing control of the comparisonwise type-I error rate rather than the experimentwise error rate.

RESULTS

In a re-analysis of the data provided by Miller et al. (22) overall the same results were obtained as reported by Miller et al. Therefore in the present paper only the augmented data base is reported.

Traditional approaches of meta analysis

In a pure random effects model (Ib, see appendix) the overall mortality odds ratio of vitamin E treatment compared to the control group pooled over all trials is $OR=0.99$ ($p=0.666$). This means that the mortality of vitamin E supplementation and control therapy does not show any differences.

Table 2 shows the results of the categorical dose-response model (Ic). In the first three model variants the cut-off values 200, 300, and 400 IU/d are used respectively in order to distinguish between high and low dose vitamin E treatment. In the former two cases no significant effects of vitamin E treatment are detected, whereas in the latter case patients receiving more than 400 IU/d seem to show an increased risk of mortality compared to control patients. In order to perform a sensitivity analysis this result is checked on the basis of a reduced data base. In particular the MRC trial (13) shows by far the largest number of participants among all high dose trials. So it might be suspected that the above result is driven entirely by that single trial dominating all others. This suspicion is only partially proven true. If the MRC trial is removed from the data base, the seeming effect of vitamin E treatment does not change, although the p-value of high dose treatment exceeds the 5% limit in this case. This is in contrast to Miller's statement. In further analyses the categorical dose-response model (Ic) is adjusted by covariates that possibly represent confounders and bias the results. Among the average period of follow-up (not reported), the mean age (not reported) and the gender distribution of trial participants the latter

represents a borderline significant covariate ($p=0.054$). If the model is adjusted by the percentage of male participants the above result of a seeming harmful effect of high dose vitamin E treatment compared to the control group is disproved. In the adjusted model both low and high dose vitamin E supplementation are estimated to have beneficial effects compared to control treatment, $OR_{LowDose \text{ vs. Control}}=0.88$ ($p=0.002$) and $OR_{HighDose \text{ vs. Control}}=0.96$ ($p=0.402$). Low Dose treatment significantly reduced the risk of all-cause mortality. In trials with high dose supplementation a minor beneficial effect compared to control treatment is found, possibly a null-effect merely, as shown by the non-significant p-value.

Hierarchical Logistic Regression

In a pure random effects model (IIa) the overall mortality odds ratio of vitamin E treatment compared to the control group pooled over all trials is $OR=1.01$ ($p=0.546$).

Table 3 shows the results of the categorical dose-response model (IIb). The two models using the cut-off dosages 200 or 300 IU/d of vitamin E supplementation respectively yield no significant effect estimates as in case of the corresponding traditional analyses. Using the cut-off value 400 IU/d high dose treatment is associated with a borderline significant increase in all-cause mortality ($p=0.060$) compared to control treatment. Interestingly and in contrast to the traditional analyses (!) this result can *not* be explained by confounding with covariates. Among the average period of follow-up (not reported), gender distribution (not reported) and mean age of trial participants the mean age proves significant ($p=0.005$). But in this case in an adjusted model the harmful effect estimate of high dose vitamin E treatment compared to the control group is confirmed to be (borderline) significant ($p=0.051$).

Table 4 shows the results of the quadratic-linear spline model (IIc). As in model (IIb) the mean age of trial participants proves a significant covariate ($p=0.006$). In the final adjusted model both parameter estimates γ_{11} and γ_{12} are significant ($p=0.018$ and $p=0.010$, respectively). This means that a significant association of vitamin E dosage and all-cause mortality is present. Figure 1 shows a graphical representation of the functional relation of vitamin E dosage and the all-cause mortality risk difference between treatment and control group. Increasing vitamin E dosage is associated with an

increasing mortality risk. Patients are at risk of higher mortality if vitamin E supplementation exceeds dosages of about 800 IU/d, as shown by the pointwise confidence intervals in Figure 1. This result is in line with the findings presented by Miller et al. (22) qualitatively. On the other hand it fundamentally contradicts the results of the traditional approaches reported above.

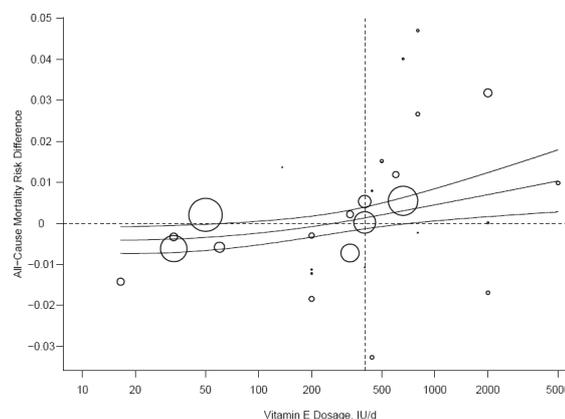


Figure 1. Hierarchical Logistic Regression, quadratic-linear dose response model (IIc), adjusted by mean age.

DISCUSSION

The above results show that the statement of Miller et al. (22) about a possible harmful effect of high dose supplementation of vitamin E is not consistent. Traditional meta analytic models and hierarchical logistic regression analyses yield contradictory results. The traditional approach finally does not indicate an enhanced mortality rate under high dose vitamin E treatment compared to the control group. A seeming harmful effect of vitamin E doses above 400 IU/d was identified not to be attributed causally to the vitamin E treatment. Instead of this, it proved to result from confounding and actually could be attributed to an unfavourable gender distribution that exists in high dose treatment groups. The protective effect of low dose treatment indeed is reduced or even removed if the vitamin E dosage is enhanced, but vitamin E treatment still does not turn to be harmful. Along with increasing dosage the mortality risk of vitamin E treatment changes from “better” to “as good as” control treatment. The hierarchical logistic regression approach applied in categorical model variants using the cut-off values 200IU/d or 300 IU/d in order to define high dose vitamin E supplementation shows no significant treatment effects. Only using the cut-off value 400 IU/d small harmful

Table 1. Data source underlying the meta analysis, for literary sources see references.

Nr.	Study	Year	Vitamin E dosage (IU/d)	Vitamin E No. deaths	Control Participants	Control No. deaths	Participants	Follow-up (years)	Mean age (years)	Men (%)	Type of Vitamin E	Source (see below)
(7)	AOX	1999	16.5	100	361	106	364	2.0	83.7	26	natural	(a)
(3)	LINXIAN-A	1993	33.0	1018	14792	1109	14792	5.3	55.0	45	synth.	(a)
(40)	MAX	2004	33.0	76	6481	98	6536	7.2	48.0	50	synth.	(b)
(2, 34)	ATBC	1994	50.0	1800	14564	1770	14569	6.1	57.0	100	synth.	(a)
(17)	LINXIAN-B	1993	60.0	157	1657	167	1661	6.0	54.0	44	synth.	(a)
(33)	EHDS	1995	136.0	1	73	0	74	6.0	47.0	40	natural	(c)
(29)	ARMDS	1996	200.0	2	39	2	32	1.5	72.0	93	natural	(c)
(38)	LINQU	2001	200.0	38	1706	43	1705	3.3	47.0	51	undef	(a)
(8)	ARTI	2002	200.0	3	164	5	164	1.2	73.0	50	synth.	(c)
(21)	VERTI	2004	200.0	39	311	44	306	1.0	85.0	27	synth.	(d)
(12)	GISSI	1999	330.0	488	5666	529	5668	3.5	59.4	85	synth.	(a)
(6)	PPP	2001	330.0	72	2231	68	2264	3.6	64.4	42	synth.	(a)
(39, 15)	HOPE-TOO	2000	400.0	799	4761	801	4780	7.0	66.0	73	natural	(b)
(1)	AREDS	2001	400.0	251	2370	240	2387	6.3	68.0	44	synth.	(a)
(35)	6STM1	2001	400.0	0	96	1	93	1.8	60.0	100	synth.	(c)
(19)	PRCP	1988	440.0	4	96	3	89	2.0	58.0	65	synth.	(c)
(10)	PPS	1990	440.0	15	433	29	431	4.0	61.0	79	natural	(a)
(37)	LCVKO	2002	500.0	1	67	0	69	2.0	64.0	45	natural	(c)
(20)	VECAT	2004	500.0	20	595	11	598	4.0	66.0	44	natural	(a)
(32, 23)	CHAOS	1996	600.0	68	1035	52	967	1.4	62.0	84	natural	(a)
(5)	REACT	2002	660.0	9	149	3	148	3.0	66.0	41	synth.	(a)
(13)	MRC	2002	660.0	1446	10269	1389	10267	5.0	65.0	75	synth.	(a)
(4)	SPACE	2000	800.0	31	97	29	99	1.4	65.0	69	natural	(a)
(14)	VEAPS	2001	800.0	1	79	1	67	3.0	53.0	87	natural	(c)
(36)	WAVE	2002	800.0	16	212	6	211	2.8	65.0	0	synth.	(a)
(30)	ADCS	1997	2000.0	19	170	22	171	2.0	73.0	35	synth.	(a)
(25, 18)	DATATOP	1998	2000.0	154	399	142	401	13.0	62.0	66	synth.	(b)
(26)	ADCSII	2005	2000.0	5	257	5	259	3.0	72.9	54	undef	(d)
(9)	E/ALS	2004	5000.0	31	83	28	77	1.5	58.0	65	synth.	(d)

(a) Trial data was originally taken from Miller et al. (22). (b) Information in Miller et al. (22) was corrected or updated on the basis of literature information. (c) Trials with <10 deaths that were excluded in Miller et al. (22). (d) Recently published trials that were not included in Miller et al. (22)

Table 2. Categorical dose-response model (Ic)

Model	OR Low Dose Vitamin E vs Control	OR High Dose Vitamin E vs Control
(Ic), cut-off=200IU/d	0.95 (p=0.208)	1.02 (p=0.467)
(Ic), cut-off=300IU/d	0.95 (p=0.144)	1.03 (p=0.322)
(Ic), cut-off=400IU/d	0.95 (p=0.104)	1.05 (p=0.018)
(Ic), cut-off=400IU/d, MRC trial removed	0.95 (p=0.077)	1.05 (p=0.156)
(Ic), cut-off=400IU/d, adjusted by pct male (OR=1.00 per unit increase, p=0.054)	0.88 (p=0.002)	0.96 (p=0.402)

Table 3. Categorical dose-response model (IIb).

Model	OR Low Dose Vitamin E vs Control	OR High Dose Vitamin E vs Control
(IIb), cut-off=200IU/d	0.99 (p=0.600)	1.02 (p=0.315)
(IIb), cut-off=300IU/d	0.99 (p=0.381)	1.02 (p=0.281)
(IIb), cut-off=400IU/d	0.98 (p=0.118)	1.04 (p=0.060)
(IIb), cut-off=400IU/d, MRC trial removed	0.97 (p=0.062)	1.04 (p=0.220)
(IIb), cut-off=400IU/d, adjusted by mean age (OR=1.04 per unit increase, p=0.005)	0.96 (p=0.057)	1.04 (p=0.051)

Table 4. Quadratic-linear spline model (IIc).

Model	γ_{11}	γ_{12}
(IIc), cut-off=400IU/d	-0.022 (p=0.034)	0.004 (p=0.019)
(IIc), cut-off=400IU/d, MRC trial removed	-0.025 (p=0.033)	0.005 (p=0.036)
(IIc), cut-off=400IU/d, adjusted by mean age (OR=1.04 per unit increase, p=0.006)	-0.033 (p=0.018)	0.006 (p=0.010)

effects of high dose treatment emerge. Dose-response analyses also show a significantly increased mortality rate along with vitamin E dosage compared to the control group. This relation can not be explained by confounding effects and thus potentially seems to be causal.

The reason for the fundamental contradiction of both above results could be the extremely small risk difference that possibly exists between vitamin E supplementation and control therapy. The possible harmful effect of high dose vitamin E treatment – if it exists at all – turns out to be almost negligibly small. Such small differences are irrelevant in epidemiology. Thus on the basis of the published trials there is no relevant increase in all-cause mortality caused by vitamin E supplementation. High dose vitamin E supplementation can not be regarded proved to increase mortality.

Appendix: Theoretical aspects of the established meta analytic models

Traditional approaches of meta analysis

In a traditional approach of meta analysis a univariate outcome measure is applied in order to describe the difference of two treatments with respect to a determined end-point. In the present case according to common recommendations this measure is chosen to be the log odds ratio $\ln OR$ of all-cause mortality of patients of the vitamin E group compared to untreated patients.

$$\ln OR = \ln \left(\frac{\text{Odds(Vita min E)}}{\text{Odds(Control)}} \right) = \beta_0 + r. \quad (I)$$

In each individual trial the log OR is calculated by (I). The respective empirical estimates are supposed to have a certain expected value β_0 . The parameter β_0 represents the so-called true effect size within a certain trial. Empirical deviations of these true effects are accounted for by means of an additional random component r in (I). The component r captures any kind of measurement error that occurs in data collection. Given the expected value β_0 the variance of the empirical log OR estimate of a certain trial is assumed to be known. Theoretical results show that it can be estimated by

$$\hat{\text{Var}}(\ln OR | \beta_0) = \frac{1}{\#Deaths(Vit.E)} + \frac{1}{\#Alive(Vit.E)} + \frac{1}{\#Deaths(Control)} + \frac{1}{\#Alive(Control)}$$

Computation of this formula obviously turns out to be problematic if any of the figures on the right hand side equals zero (number of deaths or number of patients alive in the vitamin E or control group). In order to avoid this problem and also to improve certain asymptotic properties of further analyses it is recommended to simply add the value 0.5 to each number on the right of the above formula.

In a second stage of model formulation the parameter β_0 is further modelled in several different ways.

(Ia) $\beta_0 = \gamma_{00}$ (pure fixed effects model)

Regardless of measurement error the true effect sizes β_0 of each individual trial coincide totally and take one common fixed value γ_{00} . In case of the present data the pure fixed effects model does not turn out to be appropriate. Obviously individual trial results differ markedly and this heterogeneity across individual trials is not accounted for.

(Ib) $\beta_0 = \gamma_{00} + u_0$ with $\text{Var}(u_0) = \tau_0^2$
(pure random effects model)

In the pure random effects model additional to measurement error another source of heterogeneity of individual trial results β_0 is modelled. This is represented by the random component u_0 with $\text{Var}(u_0) = \tau_0^2$.

(Ic) $\beta_0 = \gamma_{01} \cdot I_{\text{LowDose}} + \gamma_{02} \cdot I_{\text{HighDose}} + u_0$ with $\text{Var}(u_0) = \tau_0^2$
(categorical dose-response model)

The random effects model (Ib) can be extended by dropping the assumption of one single common expected value γ_{00} that holds for (the true effects sizes β_0 of) each individual trial. Instead of that depending on the administered vitamin E dose in a certain trial two different average effect sizes are modelled. Low dose trials are assumed to show an average size of γ_{01} , whereas in case of high dose trials a different expected value γ_{02} holds. In order to formally distinguish low and high dosages of vitamin E supplementation different cut-off values may be applied, as is discussed below.

All above variants of traditional meta analytic model formulations (Ia)-(Ic) can be extended in a way to account for covariates that possibly explain heterogeneity across individual trial results. This is simply done by adding a corresponding term of the respective covariate to one of the equations (Ia)-(Ic).

Parameter estimation in either of the formulated models is recommended to be done by restricted maximum likelihood methods (see 28).

Hierarchical Logistic Regression

Beyond the traditional meta analytic approach in case of binary end-points hierarchical logistic regression models can be applied alternatively. In this case in contrast to the above univariate effect measure $\ln(\text{OR})$ corresponding to each single trial a bivariate effect measure is calculated instead, i.e. the mortality risk of patients receiving vitamin E treatment as well as of untreated patients, respectively. Both levels of mortality risk within a certain trial are modelled separately by means of a logistic regression approach:

$$\ln \frac{p}{1-p} = \begin{cases} \beta_0 & \text{for untreated patients (control group)} \\ \beta_0 + \beta_1 & \text{for patients receiving vitamin E treatment} \end{cases}$$

$$\Leftrightarrow \ln \frac{p}{1-p} = \beta_0 + \beta_1 \cdot I_{\text{vit.E=yes}} \quad (\text{II})$$

In this model the term β_0 represents the average “basic” mortality risk holding for each patient regardless of which treatment group he corresponds to. The term β_1 represents an additional component that is added in the above formula in case of patients of the vitamin E group. It represents the possible change in basic mortality risk due to vitamin E treatment.

As in the traditional approach in a second stage of model formulation the (bivariate) term (β_0, β_1) is further modelled as follows.

$$(\text{IIa}) \begin{pmatrix} \beta_0 \\ \beta_1 \end{pmatrix} = \begin{pmatrix} \gamma_{00} \\ \gamma_{10} \end{pmatrix} + \begin{pmatrix} u_0 \\ u_1 \end{pmatrix} \quad \text{with} \quad \text{Var} \begin{pmatrix} u_0 \\ u_1 \end{pmatrix} = \begin{pmatrix} \tau_0^2 & \tau_{01} \\ \tau_{01} & \tau_1^2 \end{pmatrix}$$

(pure random effects model)

In the pure random effects model the basic mortality risk β_0 as well as the additional component of vitamin E treatment β_1 show a certain expected value γ_{00} or γ_{10} , respectively. Both values γ_{00} or γ_{10} are identical for each trial, i.e. in particular the size of the vitamin E component does not systematically depend on the administered dosage of treatment. The term τ_{01} in the above formula allows for correlations between the effects β_0 and β_1 within a certain trial, whereas observations coming from different trials are assumed to be stochastically independent.

$$(\text{IIb}) \begin{pmatrix} \beta_0 \\ \beta_1 \end{pmatrix} = \begin{pmatrix} \gamma_{00} \\ \gamma_{11} \cdot I_{\text{LowDose}} + \gamma_{12} \cdot I_{\text{HighDose}} \end{pmatrix} + \begin{pmatrix} u_0 \\ u_1 \end{pmatrix} \quad \text{with} \quad \text{Var} \begin{pmatrix} u_0 \\ u_1 \end{pmatrix} = \begin{pmatrix} \tau_0^2 & \tau_{01} \\ \tau_{01} & \tau_1^2 \end{pmatrix}$$

(categorical dose-response model)

In (IIb) the assumption of one single common expected value γ_{10} that holds for the vitamin E component of each individual trial is dropped. Instead of that depending on the administered vitamin E dose in a certain trial two different average effect sizes are modelled, γ_{11} in case of low dosage and γ_{12} in case of high dose trials.

$$(\text{IIc}) \begin{pmatrix} \beta_0 \\ \beta_1 \end{pmatrix} = \begin{pmatrix} \gamma_{00} \\ \gamma_{11} \cdot \ln(\text{DOS}) + \gamma_{12} \cdot \{[\ln(\text{DOS})]^2 - I_{\text{HighDose}} \cdot [\ln(\text{DOS}) - \ln(c)]^2\} \end{pmatrix} + \begin{pmatrix} u_0 \\ u_1 \end{pmatrix}$$

(quadratic-linear dose-response model)

In this case a continuous dose-response relationship is assumed, which is expressed in the form of a quadratic-linear spline, that has been proposed by Greenland (11). In case of low dose trials up to a determined cut-off value c , the second component in (IIc) reduces to

$$\beta_1 = \gamma_{11} \cdot \ln(\text{DOS}) + \gamma_{12} \cdot \{[\ln(\text{DOS})]^2\} + u_1,$$

i.e. a quadratic dose-response model. In case of all dosages larger than c the function on average effect size changes its shape and becomes

$$\beta_1 = [\gamma_{11} + 2\gamma_{12} \cdot \ln(c)] \cdot \ln(\text{DOS}) - \gamma_{12} \cdot [\ln(c)]^2 + u_1,$$

as can be derived by simple analytic arguments. This function corresponds to a linear relationship of log-dosage on average effect size β_1 . One special feature that is provided by a spline model like (IIc) is the fact that both parts of the function merge both continuously as well as continuously differentiable in mathematical sense. This kind of a smooth link between both parts of the curve obviously represents a sensible fact that meets biological behaviour in a realistic way. The choice of a quadratic-linear spline model is motivated in Miller et al. (22). The authors argue that the lower quadratic term of the model was selected to allow for nonlinear responses at low doses of vitamin E, while ensuring a null effect for 0 dose level. The upper term was restricted to be linear to avoid implausible shapes at high dosages.

As in case of the traditional meta analytic approach the above hierarchical logistic models can be extended with respect to adjustment for covariates as well. In order to model systematic heterogeneity between trials two different kinds of covariates have to be distinguished. Certain covariates affect the basic mortality risk of all

patients of a trial regardless of the treatment (e.g. common mean age of all treatment groups), whereas others obviously affect the vitamin E group exclusively (e.g. type of vitamin E, natural or synthetic). In the former case the model component $\beta_0 = \gamma_{00} + u_0$ is amended by the covariate, covariates of the second kind are inserted into the respective equation modelling β_1 .

Statistical analyses of all hierarchical logistic models are performed on the basis of population-average models (see 28). Thus all effects are to be interpreted to represent average values across trials. Robust standard errors are computed in order to provide insensitivity to model misspecification. Parameter estimation is done by restricted maximum likelihood methods.

After estimation of the parameters γ_{ij} in each of the above models the derived estimates can be transformed into common effect measures in order to compare the mortality risk between both treatment groups (odds ratio, risk ratio, risk difference). This is done by applying the error propagation law (delta method, see e.g. (27), chapter 6.a.2). The corresponding calculations are to be shown exemplarily in case of the most simple model (IIa). Parameter estimation via restricted maximum likelihood yields asymptotically normal estimates γ_{00} and γ_{10} as well as the corresponding variance-covariance matrix $\text{Var}(\gamma_{00}, \gamma_{10})$. According to the model formulation thus the expected mortality risk amounts

(i) in case of vitamin E treated patients

$$p_1 = \frac{e^{\gamma_{00} + \gamma_{10}}}{1 + e^{\gamma_{00} + \gamma_{10}}} \text{ and}$$

(ii) in case of untreated control patients:

$$p_0 = \frac{e^{\gamma_{00}}}{1 + e^{\gamma_{00}}} .$$

Applying (i) and (ii) the risk difference of treated vs. control patients $p_1 - p_0$ can be expressed as a function of γ_{00} and γ_{10} ,

$$g(\gamma_{00}, \gamma_{10}) := p_1 - p_0 = \frac{e^{\gamma_{00} + \gamma_{10}}}{1 + e^{\gamma_{00} + \gamma_{10}}} - \frac{e^{\gamma_{00}}}{1 + e^{\gamma_{00}}} .$$

This formula directly allows to derive a point estimate of the risk difference $p_1 - p_0$. In addition to this point estimate the error propagation law yields an estimate of its asymptotic variance that amounts

$$\text{Var}(\hat{p}_1 - \hat{p}_0) = \dot{G} \cdot \text{Var}(\hat{\gamma}_{00}, \hat{\gamma}_{10}) \cdot \dot{G}^T \quad \text{with}$$

$$\dot{G} := \left(\frac{\partial g(\gamma_{00}, \gamma_{10})}{\partial \gamma_{00}}, \frac{\partial g(\gamma_{00}, \gamma_{10})}{\partial \gamma_{10}} \right) \Big|_{(\hat{\gamma}_{00}, \hat{\gamma}_{10})} .$$

Under certain mild regularity conditions the derived estimate of the risk difference is asymptotically normal. Applying this result, significance tests and confidence intervals can be constructed for the risk difference $p_1 - p_0$.

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