Faculty of Health Sciences



Day 5: binary responses and 2×2 tables

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DEPARTMENT OF BIOSTATISTIC

Outline

Preliminaries

Group comparisor

Sample size and power calculation

Confounding

Cohort vs case-control study

Screening: jargor



Binary outcome

$$Y = \begin{cases} 1 & \text{event / positive / disease} \\ 0 & \text{no event / negative / non-disease} \end{cases}$$



Binary outcome

$$Y = \begin{cases} 1 & \text{event / positive / disease} \\ 0 & \text{no event / negative / non-disease} \end{cases}$$

Parameters

Prevalence: proportion of the population with event at fixed time point.

How many have the disease right now?

- ► Incidence/hazard rate: number of event relative to time unit: How many per year newly acquire the disease?
- ▶ Risk: probability that event occurs in given time period: How likely will a subject acquire the disease within 1-year?

Statistical inference

Estimating risks and prevalence

$$\widehat{p} = \text{Relative frequency} = \frac{\text{Number of events}}{\text{Number of subjects}} = \frac{x}{n}$$

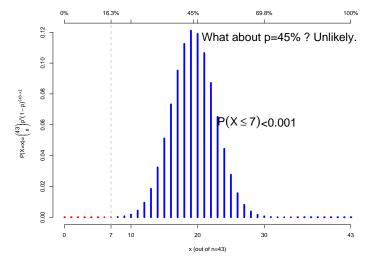
Confidence limits: normal approximation ("large" n^1)

$$\left[\widehat{p} - 1.96\sqrt{\frac{\widehat{p}(1-\widehat{p})}{n}}; \widehat{p} + 1.96\sqrt{\frac{\widehat{p}(1-\widehat{p})}{n}}\right]$$

Confidence limits: "exact" (any n)

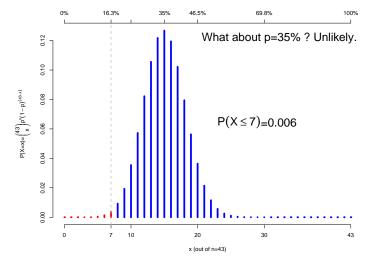


 $^{^{4/52}}$ 1rule of thumb: when both $x \ge 5$ and $n-x \ge 5$



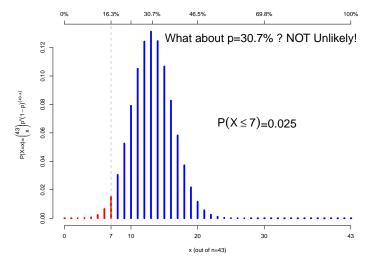






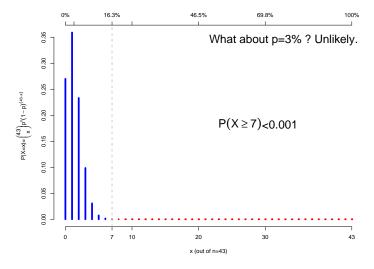






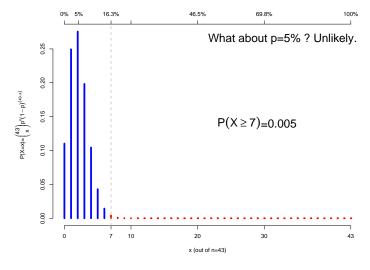
 \blacktriangleright x=7 and n=43 leads to $\hat{p}=16.3\%$ and 95% CI= [6.8;30.7].





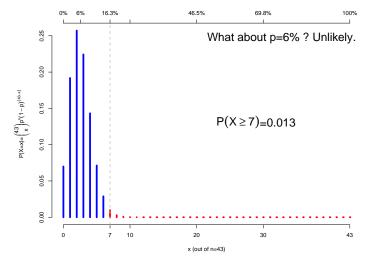






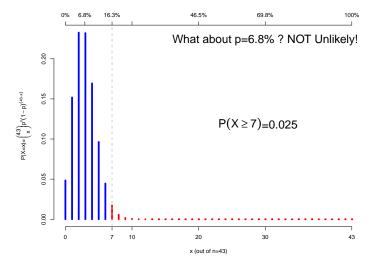








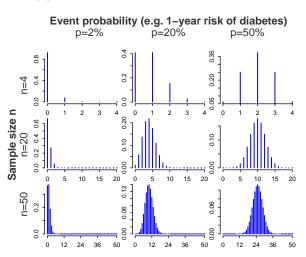








Normal approximation

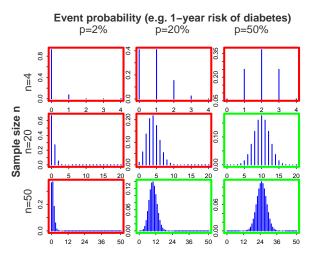


Binomial distribution: $P(X=x) = \binom{N}{x} p^x (1-p)^{N-x}$



x=7 and n=43 leads to $\hat{p}=16.3\%$ and 95% CI= [5.2;27.3]

Normal approximation



^{• &}quot;good"approximation if $np \ge 5$ and $n(1-p) \ge 5$.





Case: clinical trial on Dalteparin ³

Data: n=85 diabetic patients with peripheral arterial occlusive disease and chronic foot ulcers, randmomized (double-blind) to:

- ▶ Placebo (n = 42)
- ▶ Dalteparin (n = 43)



Outcome:

Category ²	Label
intact skin	healed
decreased ulcer area $\geq 50\%$	improved
increased ulcer area $\geq 50\%$	impaired
decreased or increased ulcer area $< 50\%$	unchanged
amputation above/below ankle	amputation

Research question: Does Dalteparin improve the outcome?

²mutually exclusive





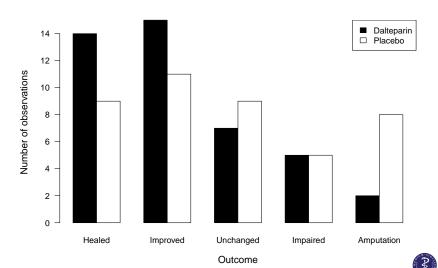
Frequency table

	Dalteparin	Placebo
Healed	14 (33%)	9 (21%)
Improved	15 (35%)	11 (26%)
Unchanged	7 (16%)	9 (21%)
Impaired	5 (12%)	5 (12%)
Amputation	2 (5%)	8 (19%)
total (100%)	43	42

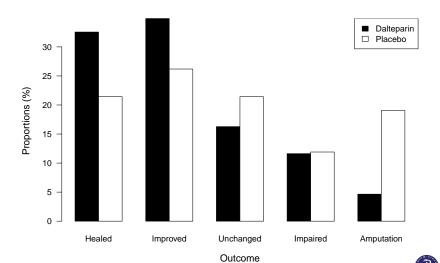
- ► Summarizes the outcome data.
- ► Prepare/Format data for analyzes.



Barplot (frequencies)



Barplot (proportions⁴)



Here we pool the outcome categories as follows

Category	Dichotomized outcome
intact skin	better
ulcer area decreased $\geq 50\%$	
decreased or increased ulcer area $< 50\%$	
increased ulcer area $\geq 50\%$	worse
amputation above/below ankle	

Important: this dichotomization should be **prespecified** (i.e. decision made before seeing the data).



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Group comparison

Placebo group

Risk of worse outcome
$$=\frac{22}{42}=\widehat{p}_1$$

Dalteparin group

Risk of worse outcome =
$$\frac{14}{43} = \hat{p}_2$$

Group comparison

Placebo group

Risk of worse outcome
$$=\frac{22}{42}=\widehat{p}_1$$

Dalteparin group

Risk of worse outcome =
$$\frac{14}{43} = \hat{p}_2$$

Association measures⁵

Relative risk:
$$\frac{\widehat{p}_1}{\widehat{p}_2}$$
 Odds ratio: $\frac{\widehat{p}_1}{\frac{1-\widehat{p}_1}{\widehat{p}_2}}$ Risk difference: $\widehat{p}_1-\widehat{p}_2$

$$\frac{1-\widehat{p_1}}{\widehat{p_2}}$$

$$\frac{1-\widehat{p_2}}{1-\widehat{p_2}}$$



⁵whenever possible, we prefer using risk ratios or risk differences to odds ratios. This is so much simpler to understand and to communicate!

2x2 contingency table

Response

Exposure –

	yes	no	total
yes	а	b	a+b
no	С	d	c+d
total	a+c	b+d	N

Risk estimates

$$\widehat{p}_1 = \frac{a}{a+b}$$
 $\widehat{p}_2 = \frac{c}{c+d}$



Relative risk

$$\widehat{RR} = \frac{a/(a+b)}{c/(c+d)}$$

Exposure

Response				
	yes	no	total	
yes	а	b	a+b	
no	С	d	c+d	
total	a+c	b+d	N	

Standard error of $\log(\widehat{RR})$ and confidence interval ⁶

$$\widehat{\sigma} = \sqrt{\frac{1}{a} - \frac{1}{a+b} + \frac{1}{c} - \frac{1}{c+d}}$$

$$CI_{95\%} = \left[\widehat{RR} \cdot \exp(-1.96\,\widehat{\sigma}) \; ; \; \widehat{RR} \cdot \exp(1.96\,\widehat{\sigma})\right]$$



Relative risk: placebo versus dalteparin

$$\widehat{RR} = \frac{22/42}{14/43} = 1.609$$

Treatment

	Outcome			
	worse	better	total	
placebo	22	20	42	
dalteparin	14	29	43	
total	36	49	85	

Standard error of $\log(\widehat{RR})$ and confidence interval

$$\hat{\sigma} = \sqrt{\frac{1}{22} - \frac{1}{42} + \frac{1}{14} - \frac{1}{43}} = 0.264$$

$$CI_{95\%} = [0.959; 2.7]$$
 (does include 1)



Risk difference

$$\widehat{\Delta} = \frac{a}{a+b} - \frac{c}{a+b}$$

Exposure

Response				
	yes	no	total	
yes	а	b	a+b	
no	С	d	c+d	
total	a+c	b+d	N	

Standard error of $\widehat{\Delta}$ and confidence interval 7

$$\widehat{\sigma} = \sqrt{ab/(a+b)^3 + cd/(c+d)^3}$$

$$CI_{95\%} = \left[\widehat{\Delta} - 1.96 \,\widehat{\sigma} \, ; \, \widehat{\Delta} - 1.96 \,\widehat{\sigma} \right]$$



Risk difference: placebo versus dalteparin

Treatment

Outcome

$\widehat{\Lambda}$		22		14		0.100
Δ	=	$\overline{42}$	_	$\overline{43}$	=	0.198

	worse	better	total
placebo	22	20	42
dalteparin	14	29	43
total	36	49	85

Standard error of $\widehat{\Delta}$ and confidence interval

$$\widehat{\sigma} = \sqrt{22 \cdot 20/42^3 + 14 \cdot 29/43^3} = 0.105$$

 $CI_{95\%} = [-0.008 ; 0.404]$ (does include 0)



Odds Ratio (OR)

Concept needed for

- case-control studies
- logistic regression

Odds: are defined as "risk of event divided by risk of no event"

$$\boxed{\mathsf{odds} = p/(1-p)} \ ,$$

and the risk can be computed back from the odds, $p=\operatorname{odds}/(1+\operatorname{odds})$.

Odds are difficult to interpret, but if risks are small, then risks \approx odds.



The Odds ratio (OR) is defined as the ratio of the odds,

$$OR = \frac{\mathsf{odds}_1}{\mathsf{odds}_2} = \frac{p_1/(1-p_1)}{p_2/(1-p_2)} \ .$$

OR are difficult to interpret, but from the equation...

$$RR = \frac{OR}{\left\{1 - p_2\right\} + p_2 OR},$$

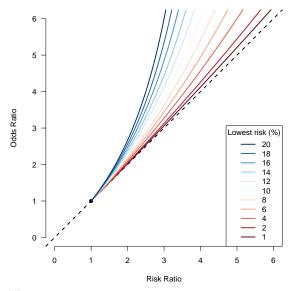
...we can first conclude:

- $ightharpoonup OR > 1 \Leftrightarrow RR > 1$
- $ightharpoonup OR = 1 \Leftrightarrow RR = 1$
- $ightharpoonup OR < 1 \Leftrightarrow RR < 1$

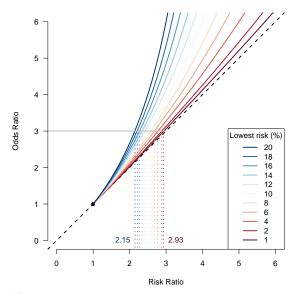
...and further conclude that

- ▶ the OR is sufficient to deduce whether a risk increases or decreases.
- ▶ if p_2 is small (e.g. rare disease), then $OR \approx RR$.

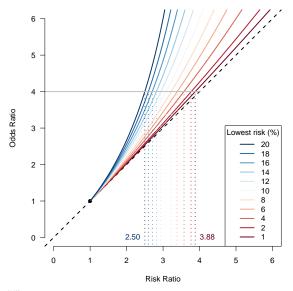




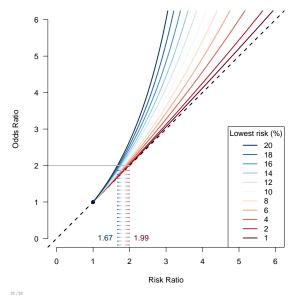














Odds ratio

$$\widehat{OR} = \frac{\frac{a/(a+b)}{b/(a+b)}}{\frac{c/(c+d)}{d/(c+d)}} = \frac{a \cdot d}{b \cdot c}$$

Exposure

Kesponse				
	yes	no	total	
yes	a	b	a+b	
no	С	d	c+d	
total	a+c	b+d	N	

Standard error of $\log(\widehat{OR})$ and confidence interval⁸

$$\widehat{\sigma} = \sqrt{\frac{1}{a} + \frac{1}{b} + \frac{1}{c} + \frac{1}{d}}$$

$$CI_{95\%} = \left[\widehat{OR} \cdot \exp(-1.96\,\widehat{\sigma}); \widehat{OR} \cdot \exp(1.96\,\widehat{\sigma})\right]$$



Odds ratio: placebo versus dalteparin

$\widehat{OR} = \frac{22 \cdot 29}{14 \cdot 20} = 2.279$

Treatment

	worse	better	total
placebo	22	20	42
dalteparin	14	29	43
total	36	49	85

Outcome

Standard error of $\log(\widehat{OR})$ and confidence interval

$$\widehat{\sigma} = \sqrt{\frac{1}{22} + \frac{1}{20} + \frac{1}{14} + \frac{1}{29}} = 0.449$$

$$CI_{95\%} = [0.946; 5.491]$$
 (does include 1)



Reporting results

The relative risk (of worsening) of group 1 (Dalteparin) versus group 2 (Placebo) is estimated as

$$RR = \frac{14/43}{22/42} = 0.622$$

Equivalent statements:

- ► The risk in group 2 is reduced by a factor 0.622 compared to group 1.
- ► The risk in group 2 is 37.8% lower than in group 1.9
- ▶ The risk in group 1 is 1.609 times higher than in group 2^{10}
- ► The risk in group 1 is 60.9% higher than in group 2.



Testing independence in a randomized clinical trial

Null hypothesis: the treatment has no effect.

$$\begin{array}{lll} \operatorname{Prob}(\operatorname{worse\ given\ dalteparin}) = \operatorname{Prob}(\operatorname{worse\ given\ placebo}) \\ \Leftrightarrow & p_1 - p_2 = 0 & (\operatorname{Difference} = 0) \\ \Leftrightarrow & \frac{p_1}{p_2} = 1 & (\operatorname{Relative\ risk} = 1) \\ \Leftrightarrow & \frac{p_1/(1-p_1)}{p_2/(1-p_2)} = 1 & (\operatorname{Odds\ ratio} = 1) \end{array}$$

Popular tests of independence between the treatment group and the outcome groups:

- $\triangleright \chi^2$ test (normal approximation)¹¹
- ► Fisher's exact test



^{25/52}11 This method is "good enough" with "large enough" sample sizes.

The χ^2 test statistic

$$\chi^2 = \sum \frac{(\text{observed counts} - \text{expected counts})^2}{\text{expected counts}}$$

Observed counts

		Response		
		yes	no	total
Exposure	yes	а	b	a+b
Exposure	no	С	d	c+d
	total	a+c	b+d	N

Response

Expected counts

	Response			
		yes	no	total
Exposure	yes	(a+b)(a+c)/N	(a+b)(b+d)/N	a+b
Lxposure	no	(c+d)(a+c)/N	(c+d)(b+d)/N	c+d
	total	a+c	b+d	N

 under the null hypothesis the groups are identical, hence data can be merged into a single group

in a population of size n, for a given risk of event p, we expect to see (on average) np events in this population

The expected counts are calculated under the null hypothesis.

Rule of thumb: a valid analysis requires that all expected counts are ≥ 5 .



Test results

Null hypothesis:

dalteparin treatment has no effect for chronic foot ulcers.

Test	p-value
Fisher's exact test	0.0808
Pearson's χ^2 test	0.0644
Pearson's χ^2 test with Yates' continuity correction ¹²	0.1032

R code:

```
tab <- rbind(c(22,20),c(14,29))
fisher.test(tab)
chisq.test(tab,correct=FALSE)
chisq.test(tab,correct=TRUE)</pre>
```

 $^{^{12}}$ Expected to be more precise than the usual Pearson's χ^2 test when the sample size is very small.

A note of caution

Because the (simple) formulas provided above for the 95% CI are based on large sample size approximations, they are not necessarily consistent with the result of the Fisher's exact test, with "very small" sample sizes.

Example:

	event	no event
exposed	5	12
non-exposed	8	3

- $\widehat{p}_1 = 8/11 = 0.73, \quad \widehat{p}_2 = 5/17 = 0.29.$
- \triangle $\hat{\Delta}$ = 0.43 (0.09; 0.77)
- $ightharpoonup \widehat{RR} = 2.47 \ (1.09 \ ; 5.62)$
- \triangleright \widehat{OR} = 6.40 (1.18; 34.61)
- ightharpoonup p-values from Fisher's exact test and Pearson's χ^2 (with and without Yates correction) are 0.051, 0.063 and 0.025, respectively.

Here the confidence intervals show a significant result, but not Fisher's test.



Advanced methods and software¹³ are available to avoid running into this kind of inconsistency between hypothesis test and confidence intervals.

Fortunately, it is rare that we run into this problem.... and even rarer that it matters for the interpretation.



Larger contigency tables (1/2)

If the table is not 2x2 but, e.g., 3x4 or 2x4, the χ^2 test and Fisher's exact test are testing an "ANOVA-like" null hypothesis similarly to what the F-test does to compare several means.

First example:

	underweight	normal	overweight	obese
no SCD	9	51	20	8
SCD	23	61	3	1

R code:

fisher.test(table(d\$SCD,d\$BMIgroup))

returns a p-value < 0.001, for the null hypothesis

 H_0 : "the prevalence of SCD is the same in all BMI groups"

(i.e. "no association between BMI and SCD").



Larger contigency tables (2/2)

Second example:

	underweight	normal	overweight	obese
age=[16, 25)	14	45	1	1
[25, 30)	3	25	3	1
[30, 67]	15	42	19	7

R code:

fisher.test(table(d\$ageGroup,d\$BMIgroup))

returns p-value=0.004, for the null hypothesis

H₀: "the prevalence of each BMI group is the same in all age groups"

(i.e. "no association between BMI and age").



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Textbook formula ("large n" approximation)

$$n = \frac{\left\{z_{\alpha/2}\sqrt{2\bar{p}(1-\bar{p})} + z_{\beta}\sqrt{p_1(1-p_1) + p_2(1-p_2)}\right\}^2}{(p_1 - p_2)^2}$$

- $z_{\alpha/2} = -1.96$ for $\alpha = 5\%.^{14}$
- $ightharpoonup z_{1-\beta} = 0.84$ and 1.28 for $1 \beta = 80\%$ and 90%.
- $\bar{p} = (p_1 + p_2)/2.$
- ▶ *n*: number of observations in **each** group.

Useful for computing:

- ▶ Sample size: n for given "guesses" of p_1 and p_2 and desired 1β and α .
- ▶ Power for a given budget/sample size: 1β for "guesses" of p_1 and p_2 and desired n and α .
- Least detectable difference (or ratio): $\delta=p_1-p_2$ (or $r=p_1/p_2$) for given n, "guess" of p_1 and desired α and minimal power $1-\beta$.



 $^{^{\}overline{33/52}}$ 14where z_{γ} is the γ -quantile of a standard normal distribution.

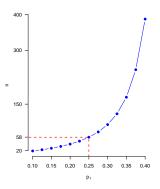
Sample size calculation

Standard software can be used, e.g. R:

```
power.prop.test(p1 = 0.25, p2 = 0.5, power=0.8)
```

Two-sample comparison of proportions power calculation

NOTE: n is number in *each* group



▶ n = 58 subjects needed in **each** group (i.e. 116 in total) to detect significant risk difference with a power of 80%, if the risks in the two groups are 25% and 50%.

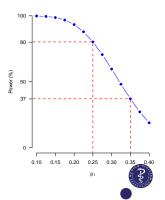
Power calculation

Example: an initial calculation suggests n=58 subjects per group (i.e. 116 in total), for detecting a difference of 25% survival between the two groups, assuming 50% survival in the placebo group (with 80% power). But what does the power become if we were too optimistic with the expected treatment effect? E.g. what if the difference in survival probability is only 15%?

```
power.prop.test(n=58, p1 = 0.35, p2 = 0.5)
```

Two-sample comparison of proportions power calculation

NOTE: n is number in *each* group



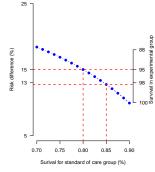
Least detectable difference

Example: My grant can finance a total sample size of n=150 (i.e. 75 per group). What is the smallest survival difference that I can hope to show with a decent power (e.g. 80%), if I expect 80% survival in the "standard of care" (i.e. control) group? And if I expect 85% in the "standard of care" group?

```
power.prop.test(n=75, p1 = 0.8, power=0.8)
```

Two-sample comparison of proportions power calculation

NOTE: n is number in *each* group



Note: you need to supply a value for p1, not p2, otherwise the software is looking for a lower risk and it returns 0.72.

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Confounding

"A simple definition of confounding is the confusion of effects. This definition implies that the effect of the exposure is mixed with the effect of another variable, leading to a bias." ¹⁵

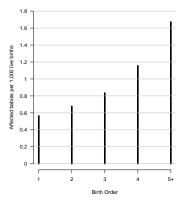
Failing to take a confounding variable into account can lead to a **false conclusion** that the outcome are in a **causal** relationship with the predictor variable.

Confounding variables are typically encountered in observational studies, but **not** in "ideal" randomized experiments.



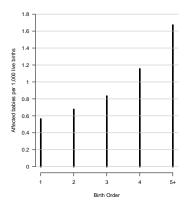
^{38/52}15 Rothman (2012), Epidemiology: an introduction.

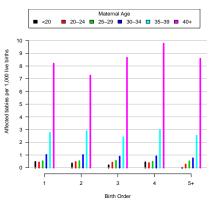
Confounding example (birth order and risk of Down syndrome 16)





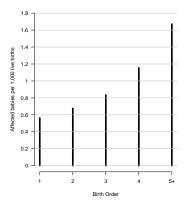
Confounding example (birth order and risk of Down syndrome 16)

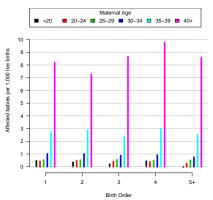




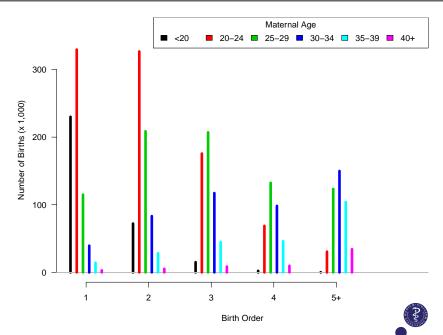


Confounding example (birth order and risk of Down syndrome 16)









When can association mean causation? (1/2)

We usually say that (statistical) association does not imply causation.

Confounding (among others things) can lead to non-causal associations.

There are however some exceptions, i.e., it exists some situations in which an association can be interpreted as a causal association. An example is the situation in which we analyze data from an "ideal" randomized experiments.



When can association mean causation? (2/2)

This is because the randomization ensures that the two compared groups are similar with respect to everything except the intervention / treatment under study. Hence, if a difference in outcome is observed between the two groups, then we can be confident that this is the consequence of this unique difference in exposure / treatment.

In non-randomized (or non "ideally" randomized) experiments the two compared groups will usually differ with respect to more than one characteristic. This generates multiple plausible explanations for the observation of the difference in outcome – some causal and some non causal.



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Adjusted analysis

Suppose that in addition to the outcome and the exposure group a categorical confounder variable (e.g. gender) is measured for each individual.

Subgroup analysis

Analyze 2x2 contingency tables separately in each strata defined by the confounder variable.

► Logistic regression (see Lecture 6)

To compute a "weighted" average of the subgroup analyses, assuming that the exposure-outcome association is the same in all subgroups. ¹⁷.



^{43/5217} Applicable also with continuous confounders.

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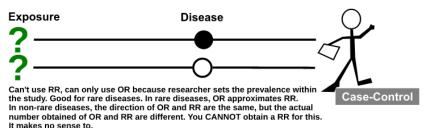
Observational study design

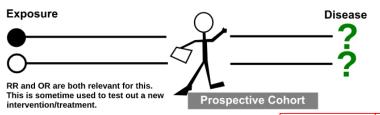
In a prospective **cohort study**, an outcome or disease-free study population is first identified by an exposure (e.g., onset of diabetes) or other inclusion criteria and followed in time until the disease or outcome of interest occurs.

Case-control studies identify subjects by outcome status at the outset of the investigation. First, subjects with outcome are identified and classified as cases. For each case a given number of controls (e.g., 4) are selected. A candidate control is a subject without the outcome but from the same source population.



Observational Study Designs: Case Control vs Cohort







Cohort study: example

For example consider the study from Egerup et al. (2020) 18

Infection within first year of life

		yes	no	total
Kidney- transplanted mother	yes	26	98	124
	no	133	1098	1231
	total	159	1196	1355

The estimated risk ratio is $\widehat{RR} = 1.94$ (Cl_{95%} = [1.33; 2.83]).



Case-control study: example

For example consider the case-control study of Frachon et al. 19 20

"explained" cardiac problem

Benfluorex use

		yes	no	total
-	yes	19	3	24
	no	8	51	59
	total	27	54	81

$$\widehat{OR} = 40.4$$
 (CI_{95%}: [9.7; 168])

The number of controls (here 2 per case) is defined by the study design. Hence we cannot estimate risks as one minus the proportions of controls among exposed and non-exposed...

- ▶ The statistic RR depends also on the ratio between controls and cases and should **not** be used for measuring association in case-control studies.
- ▶ The statistic \widehat{OR} works.

²20 See movie "150 Milligrams" (2016)



 $^{^{19}}$ Frachon et al. "Benfluorex and unexplained valvular heart disease: a case-control study." Plone 5.4 (2010).

Why does \widehat{OR} work? (1/2)

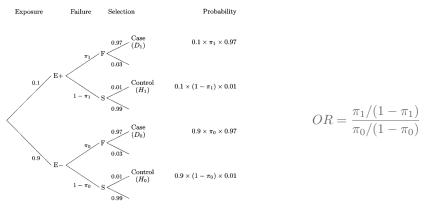


Fig. 16.1. The probability model in the study base.

- 97% of the cases are included in the case-control study and 1% of the "non cases" are selected as controls; all included "blinded" from exposure (i.e. before looking for the information on the exposure).
- Connection to notations of previous slides $\pi_1 = p_1$ and $\pi_0 = p_2$.
- ► E="exposure", F="Fail", S="Survive", D="Disease", H="Healthy".
- source: "Statistical models in Epidemiology", by Clayton and Hills, page 155.



Why does \widehat{OR} work? (2/2)

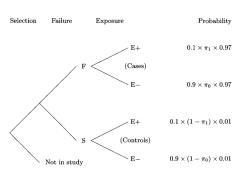


Fig. 16.2. The probability tree for the retrospective argument.

$$\widehat{OR} \approx \frac{\frac{0.1 \times \pi_1 \times 0.97}{0.1 \times (1 - \pi_1) \times 0.01}}{\frac{0.9 \times \pi_0 \times 0.97}{0.9 \times (1 - \pi_0) \times 0.01}}$$
$$= \frac{\pi_1 / (1 - \pi_1)}{\pi_0 / (1 - \pi_0)}$$

source: "Statistical models in Epidemiology", by Clayton and Hills, page 156.



Why does \widehat{OR} work? (2/2)

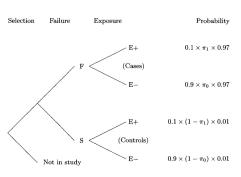


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$$\widehat{OR} \approx \frac{\frac{0.1 \times \pi_1 \times 0.97}{0.1 \times (1 - \pi_1) \times 0.01}}{\frac{0.9 \times \pi_0 \times 0.97}{0.9 \times (1 - \pi_0) \times 0.01}}$$

$$= \frac{\pi_1/(1 - \pi_1)}{\pi_0/(1 - \pi_0)}$$

but

$$\widehat{RR} \approx \frac{\frac{0.1 \times \pi_1 \times 0.97}{0.1 \times \pi_1 \times 0.97 + 0.1 \times (1 - \pi_1) \times 0.01}}{\frac{0.9 \times \pi_0 \times 0.97}{0.9 \times \pi_0 \times 0.97 + 0.9 \times (1 - \pi_0) \times 0.01}}$$

$$= \frac{\pi_1 / (\pi_1 \times 0.97 + (1 - \pi_1) \times 0.01)}{\pi_0 / (\pi_0 \times 0.97 + (1 - \pi_0) \times 0.01)}$$

$$\neq \frac{\pi_1}{\pi_0}$$



SITY OF COPENHAGEN

Outline

Preliminaries

Group comparisor

Sample size and power calculation

Confounding

Cohort vs case-control study

Screening: jargon



Medical test / screening: jargon

Y: Outcome (disease status) E.g. prostate cancer

$$X \colon \text{Test result (biomarker)}. \ \text{E.g.} \ X = \left\{ \begin{array}{ll} 1 & \text{positive if PSA} > 4.0\,\text{ng/mL} \\ 0 & \text{negative if PSA} \leq 4.0\,\text{ng/mL} \end{array} \right.$$

$$egin{array}{cccc} Y=1 & Y=0 \\ X=1 & {
m True\ positive} & {
m False\ positive} \\ X=0 & {
m False\ negative} & {
m True\ negative} \\ \end{array}$$

- ▶ True positive rate (sensitivity): $P(X = 1 \mid Y = 1)$
- ▶ True negative rate (specificity): $P(X = 0 \mid Y = 0)$
- ▶ False positive rate (1-specificity): $P(X = 1 \mid Y = 0)$
- ▶ The positive predictive value: $P(Y = 1 \mid X = 1)$
- ▶ The negative predictive value: $P(Y = 0 \mid X = 0)$

