Study Design and Basic Biostatistics

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Objectives
- Compare and contrast various study designs
- Describe basic statistical concepts
- Apply statistical testing to primary outcome
It all starts with “Why”?
• Are outcomes improved with rapid PCR and ID Pharmacist intervention in patients with Staph Aureus bacteremia?
• Is high dose dexmedetomidine more efficacious than low dose in critically ill patients?
• Does delaying IV lipids when starting TPN decrease infections in SICU patients?

Causal inference and Bias
• Causal inference
  – Strength of the association
  – Validity
• Bias
  – systematic error
  – Many different types
  – Difficult to quantify

Confounding
• Occurs when study association is mediated by a third factor
• Exists on the causal pathway
  • Example: smoking status when studying alcohol consumption and death
Types of Studies

• Descriptive
  – Case Reports
  – Case Studies/Series

• Observational
  – Case–Control
  – Cohort
    • Retrospective
    • Prospective
  – Cross-sectional

• Experimental
  – RCT
  – Quasi-experimental

• Meta-Analysis

Descriptive Studies

• Case Studies
  – Dexmedetomidine-associated bradycardia leading to Pulseless Electrical Activity. Pharmacother 2009

• Case Series
  – Vitamin B6 Deficiency: A Potential cause of refractory seizures in adults. JPEN 2011

• Not research as not testing hypothesis
• Used for rare events or new observations
• Hypothesis generating

Clinical Research Design

• 2 distinct types: Experimental or Observational

• Experimental
  – Investigator intervenes on population for sole purpose of evaluation

• Observational
  – Makes no attempt to intervene exclusively for investigational purpose
Experimental Studies
- Randomized Controlled Trials
  - Advantages
    - Best design to determine causality
    - Minimizes bias via randomization
    - May be parallel or crossover design
  - Disadvantages
    - Cost
    - Time
    - Ethical issues

Observational Studies
- Case-Control Studies
  - Study population defined by outcome of interest
- Cohort Studies
  - Study population defined by exposure
    - Retrospective
    - Prospective

Case-Control Studies
- Advantages
  - Relatively uncommon conditions
  - Allows for smaller n than cohort
  - Allows investigation of many exposures
  - Inexpensive and less time-consuming when outcomes are relatively rare
- Disadvantages
  - Selection of controls can be difficult and time-consuming
  - Confounding and bias are concerns
Case-Control Study

- Exposure?
  - YES = Case
  - NO = Control
- Outcome of interest? (death, recurrence, disease, etc.)
  - YES
  - NO

Cohort Studies
- Can be prospective or retrospective
- Advantages
  - Allows study of more than one disease/outcome
  - Less selection bias than case control
- Disadvantages
  - Expensive
  - Long time to conduct (prospective)
  - Loss to follow-up (missing data)
  - Larger n
  - Prone to information bias
  - Inefficient for evaluating rare diseases/outcomes
Examples
- Rapid PCR
  - Implemented new technology
  - Pre/post study or Cohort Study
- Dexmedetomidine
  - Study all patients and compared HDD > 0.7 mcg/kg/h to LDD < 0.7 mcg/kg/h
  - Case Control Study
- TPN
  - Implemented TPN w/o IVFE
  - Pre/post study or Cohort Study

Resident projects?

Statistics
- Methods for collecting, classifying, summarizing & analyzing data
- Descriptive
  - Frequency, Histogram, Measure central Tendency, Measure of spread, Scatter plot
- Inferential
  - Conclusion or generalization made about a population from study using a sample population
Variables
- Discrete or categorical
  - Nominal: classified into groups with no particular order or severity (Yes/NO)
    - E.g., Sex, mortality, Disease State
  - Ordinal: Ranked in specific order but no consistent level of magnitude between groups
    - E.g., NYHA class, Trauma scores, Likert scales

Variables
- Continuous
  - Interval: Data are ranked in specific order with constant change in magnitude with zero point arbitrary
    - E.g., Fahrenheit Temperature
  - Ratio: Like interval but with an absolute zero
    - E.g., Heart Rate, Age, Blood pressure

Normal (Gaussian) Distribution
- Bell Shape
- Mu: Mean
- Theta: Standard Deviation (SD)
  - 68% of population are within +/- 1 SD
  - 95% of population are within +/- 2 SD
Descriptive Statistics

- **Mean**: Average
  - Only for Continuous data
  - Sensitive to outliers
- **Median**: Point where half of observations fall below and above
  - Used with ordinal & continuous data
  - Insensitive to outliers
- **Mode**: Most common value
  - For all data types
  - Mode=Median=Mean for Normal Distribution

Descriptive Statistics, continued

- **Standard deviation (SD)**
  - Calculated to reflect range of samples
  - Appropriate for normal or nearly normal data
- **Standard error of the mean (SEM)**
  - Estimated from Standard deviation (=SD/√n)
  - 95% of sample means lie within ± 2 SEM
- **Range (minimum and maximum)**
- **Interquartile range (IQR)**
  - 75% of the population fall within this range

<table>
<thead>
<tr>
<th></th>
<th>Range (IQR)</th>
<th>SD</th>
<th>SEM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Interval/Ratio data</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Ordinal data</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
</tr>
<tr>
<td>Descriptive of sample variability</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
</tr>
<tr>
<td>Assists in Statistical Inference</td>
<td>No</td>
<td>Yes</td>
<td>Yes</td>
</tr>
<tr>
<td>Used to calculate Confidence Intervals</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
</tr>
</tbody>
</table>
Hypothesis Testing

- Used to determine if any observed difference between groups is due to chance or difference in study populations

1. Define research question
   - Does rPCR with an ID PharmD intervention shorten time to appropriate therapy in patient with S. aureus bacteremia? (Pre/post intervention study design)

2. Define null hypothesis ($H_0$):
   - $H_0$: No difference between pre and post intervention groups in time to appropriate therapy

3. Define alternative hypothesis ($H_a$)
   - $H_a$: Pre-intervention group ≠ Post-intervention group in time to appropriate therapy

Hypothesis testing, continued

- If $H_0$ is rejected = statistical difference between groups
- If fail to reject $H_0$ = no statistical difference

Note: statistical analysis can NEVER prove truth of a hypothesis, but rather provide evidence to support or refute
Hypothesis Testing

- Choose statistical test based on:
  - Type of data (nominal, ordinal, continuous)
  - Study design (matched, parallel, crossover)
  - Presence of confounding variables

- Depending on p-value, the investigator will reject or fail to reject the $H_0$

P-values (probability testing)

- Probability testing measures how likely the observed difference (if any) is due to chance.
- Related to sample size and effect size (size of the difference between groups)
- Report exact p-values (not S or NS)
- P-values do NOT indicate clinical significance

Statistical Test

<table>
<thead>
<tr>
<th>Type of variable</th>
<th>Statistical test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nominal</td>
<td>Chi squared</td>
</tr>
<tr>
<td></td>
<td>Fishers Exact Test</td>
</tr>
<tr>
<td>Ordinal (nonparametric)</td>
<td>Wilcoxon Rank Sum</td>
</tr>
<tr>
<td></td>
<td>Mann Whitney U</td>
</tr>
<tr>
<td></td>
<td>Kruskal-Wallis ($\geq 3$ groups)</td>
</tr>
<tr>
<td>Continuous (parametric)</td>
<td>Student’s t-test</td>
</tr>
<tr>
<td></td>
<td>ANOVA ($\geq 3$ groups)</td>
</tr>
</tbody>
</table>
Example: Staph PCR

- Are outcomes improved with rapid PCR and ID Pharmacist intervention in patients with *Staph aureus* bacteremia?
- Endpoint: Time to appropriate antibiotic for MSSA
- What type of data?

Example: Staph PCR

<table>
<thead>
<tr>
<th>variable</th>
<th>min</th>
<th>25%</th>
<th>median</th>
<th>75%</th>
<th>max</th>
<th>mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>icu days</td>
<td>1</td>
<td>1.5</td>
<td>2</td>
<td>2.5</td>
<td>5</td>
<td>2</td>
<td>0.725</td>
</tr>
</tbody>
</table>

Answers

- Type of Data: Continuous (days)
- Biostats Test: Chi Square since normally distributed
• Is high dose dexmedetomidine more efficacious than low dose in critically ill patients?
• Primary endpoint: % time at goal RASS, undersedated, oversedated?
• What type of data?

Example: High dose Dexmedetomidine

<table>
<thead>
<tr>
<th>% Time undersedated</th>
<th>min</th>
<th>p25</th>
<th>median</th>
<th>p75</th>
<th>max</th>
<th>mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>% time</td>
<td>0</td>
<td>0</td>
<td>4</td>
<td>7</td>
<td>22</td>
<td>4.97</td>
<td>5.96</td>
</tr>
</tbody>
</table>

Example: High dose Dexmedetomidine

Answers
• Type of Data: Continuous (% of time at goal RASS)
• Biostats Test: since non-normally distributed Mann-Whitney U
Example: Withholding lipids in TPN

- Does delaying IV lipids when starting TPN decrease infections in SICU patients?
- Primary endpoint: Development of Infectious complications after TPN started
- What type of data?

Example: Withholding lipids in TPN

<table>
<thead>
<tr>
<th>Total</th>
<th>Pneumonia</th>
<th>BSI</th>
<th>CRBSI</th>
<th>UTI</th>
</tr>
</thead>
<tbody>
<tr>
<td>IVFE</td>
<td>Withhold IVFE</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Answers

- Type of Data: Nominal
- Statistical test: chi square or Fisher’s exact test
Power
- Probability that the statistical test can detect a difference when a true difference exists
  \[ = 1 - \beta \]
- Dependent on:
  - \( \alpha \) (probably of wrongly concluding statistical difference between groups)
  - \( \beta \) (probably of wrongly concluding groups equal)
  - Sample size
    - Dependent on ethics, cost, availability, etc.
  - Effect size
    - Based on prior studies, clinical judgment
  - One-tailed vs two-tailed tests
  - Parametric vs non-parametric tests
  - Parametric tests are more powerful
  - Study design

Sample Size Determination
- Well educated guess
- Factors affecting sample size:
  - \( \alpha \)
  - Power
  - Effect size
- Consult statistician
- No "magic number"

Sample Size Example
Research question: Does extended-infusion piperacillin/tazobactam lead to improved mortality rates compared to intermittent-infusion?

<table>
<thead>
<tr>
<th>Estimates of Mortality (20% in Intermittent group, 10% in Extended group)</th>
<th>Power (%)</th>
<th>alpha</th>
<th>Sample size (Total)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Extended Infusion</td>
<td>Intermittent Infusion</td>
<td>Extended Infusion</td>
</tr>
<tr>
<td>70</td>
<td>0.05</td>
<td>352</td>
<td>176</td>
</tr>
<tr>
<td>70</td>
<td>0.1</td>
<td>278</td>
<td>139</td>
</tr>
<tr>
<td>70</td>
<td>0.01</td>
<td>528</td>
<td>264</td>
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<tr>
<td>80</td>
<td>0.05</td>
<td>352</td>
<td>176</td>
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<td>0.01</td>
<td>528</td>
<td>264</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Estimates of Mortality (40% in Intermittent group, 30% in Extended group)</th>
<th>Power (%)</th>
<th>alpha</th>
<th>Sample size (Total)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Extended Infusion</td>
<td>Intermittent Infusion</td>
<td>Extended Infusion</td>
</tr>
<tr>
<td>80</td>
<td>0.05</td>
<td>752</td>
<td>376</td>
</tr>
<tr>
<td>80</td>
<td>0.1</td>
<td>600</td>
<td>300</td>
</tr>
</tbody>
</table>

Add reference
Example

- Is inpatient hypertension associated with worse outcomes in trauma patients ≥45 years?
- No data on inpatients hypotension
- Hypothesis: Inpatient hypertension is associated with increased incidence of composite of MI, Stroke, VTE, AKI and Afib.
- Preliminary data n=107
  - 43 inpatient HTN, 63 no HTN
  - composite in 16.3% w HTN and 6.3% without, P=0.11
- Sample size calculation for significance 60 per group

Statistical Significance

- Size of p-value not related to importance of results
  - Smaller p-values mean less likely chance explains the difference
- Statistical significance does not mean clinically significant
- Lack of statistical difference does not mean results are not important
  - Lancet 2000;356:2139-43 Dopamine in ARF
  - Publication bias

Other Statistical Parameters

- Confidence Intervals
- Relative Risks/Odds Ratios
- Correlation (does not answer chicken or egg)
- Regression (Predictive)
- Survival Curves
  - Kaplan Meier
  - Cox Proportional Hazard Ratio
“Likeness to truth is not the same thing as truth”
Socrates

Resources
- Ann Emerg Med 1990;591-7
- Graphpad.com
- Bmj.com/collections/statbk/index.shtml
- Biostats Lecture on Pharmacy Intranet Operations>Training/Competency>Pharmacist Programs
- Critical Care 2002;6:335-341

Confidence Intervals (CI)
- Does your point estimate exist in the population?
- CI give idea of magnitude of difference with point estimate
  \[ 95\% \text{ CI } = x \pm 1.96 \left( \frac{SD}{\sqrt{n}} \right) \]
  or \[ x \pm 1.96 \times \text{SEM} \]
- CIs that include zero interpreted as p>0.05
- Changes in MAP for a drug is 95% CI –12 mmHg, (-22 to –10)
  - Means with 95% confidence drug a reduces MAP between 22 and 10 mmHg with a best point estimate of 12 mmHg
Correlation

- Estimate strength of relationship between 2 variables
- R is correlation coefficient range -1 to +1
- -1 is perfect negative correlation or indirect relationship
- +1 is perfect positive correlation or direct relationship
- Does not imply causation (chicken or egg)
- Pearson r, Spearman r

Regression

- Regression PREDICTIVE, correlation is not
- Mathematical method to describe relationship with goal to develop equation for prediction of one variable from one or more variables
  - Linear regression (OLS)
    - \( Y = MX + B \)
    - X= independent variable, Y= dependent variable, R\(^2\) statistic
    - Y can be continuous and must be normally distributed
  - Intensive Care Med 2004;30:1537-43
    - BIS XP and RASS R\(^2\) 0.36 , p=0.011
    - Means 36% of time BIS predicted RASS score

Regression, continued

- Logistic
  - Dependent variable is binary, no assumptions about independent variables
- Multiple
  - Multiple dependent variables
- Proportional hazards (discussed later)
RR can not be directly calculated for most case control studies
- Use RR for Cohort Studies
  \[ RR = \frac{(A/A+B)}{(C/C+D)} \]
- Use OR for Case-Control Studies
  \[ OR = \frac{(A/C)}{(B/D)} \]

Survival Analysis
- Time-to-event analysis
  - Example: time to death after disease diagnosis, time from beginning to end of remission period
  - Assumes HR is constant over time
- Censoring
- Kaplan-Meier Curve
  - Estimates survival time
  - No underlying distribution assumption
- Log-Rank Test

Survival Analysis, continued
- Cox proportional hazards regression
  - Allows adjustment for confounding variables
- Hazard Ratio (HR)
  - Probability of experiencing the event (death, no longer in remission) given the patient has survived
  - Interpreted similar to OR or RR