Keys to executing good field research
开展良好生产研究的关键

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Evidence hierarchy证据层次结构

- Levels of evidence are based on the **value** of the **evidence** for making the **appropriate** decision.
- 证据的级别是基于做出适当决定的证据的价值。
- How to evaluate quality of scientific evidence?
- 如何评价科学证据的质量？
  - **Quality of the evidence 证据的质量**
    - Design 设计
  - **Risk of bias 误差风险**
    - **Bias** is a **systematic error**, or deviation from the truth.
    - 偏差是一种系统的错误，或偏离事实。
    - Different biases can lead to underestimation or overestimation.
    - 不同的偏见会导致低估或高估。
    - Selection bias, information bias, and confounding (similar concept to blocking factors).
    - 选择偏差、信息偏差和混杂因素（类似于阻断因素的概念）。

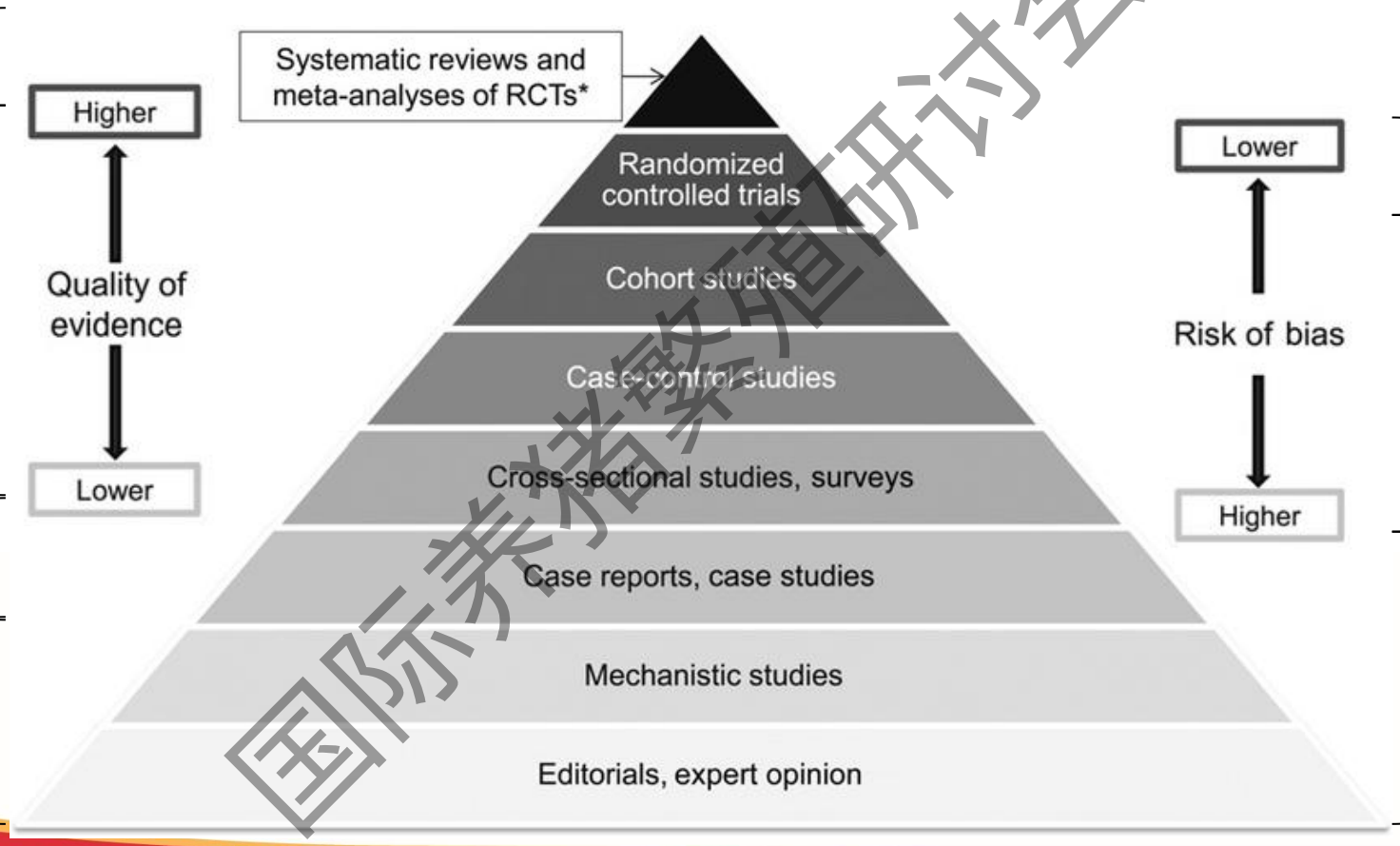
Overview on Trials designs试验设计概述

Summary of primary Studies
初步研究总结

Primary Studies
初步研究

Description of observations
描述观察

Opnions
观点

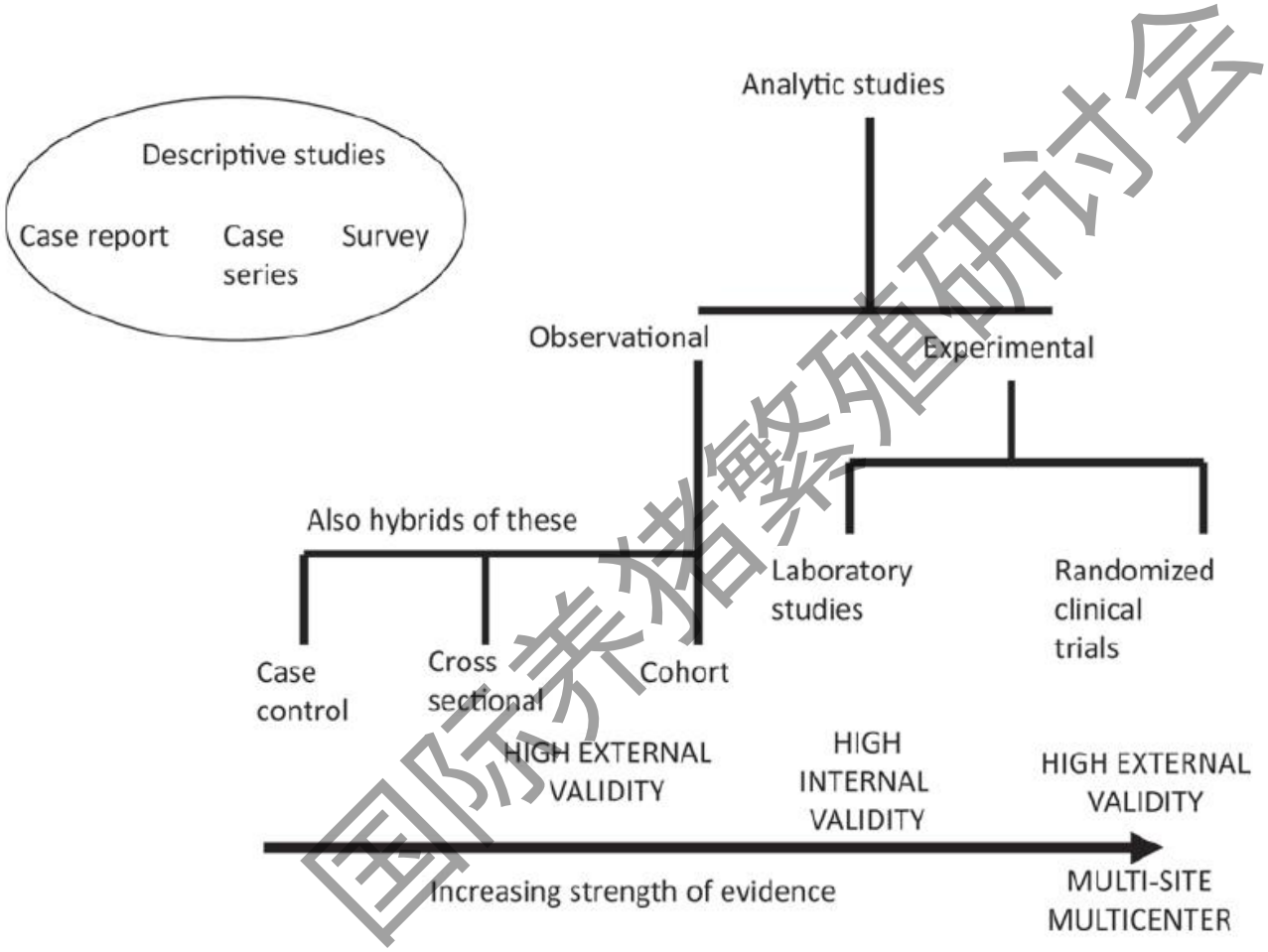


Experimental studies
实验研究

Observational studies
观察研究

Studies or info without design
没有设计的研究或信息

Overview of Epi studies 实验研究概述



Source: Lean et al, 2009. doi: 10.3168/jds.2009-2140

Descriptive studies 描述性研究

- Description of the nature and distribution of a outcome.
- 对结果的性质和分布的描述。
- **Limitations: 限制：**
  - **Don't have a comparison (control) group.**
  - **没有一个对照组（对照组）。**
  - Not appropriate for testing hypotheses (e.g. disease causation, risk factors or efficacy of interventions).
  - 不适合测试假设（例如，疾病原因、危险因素或干预措施的有效性）。

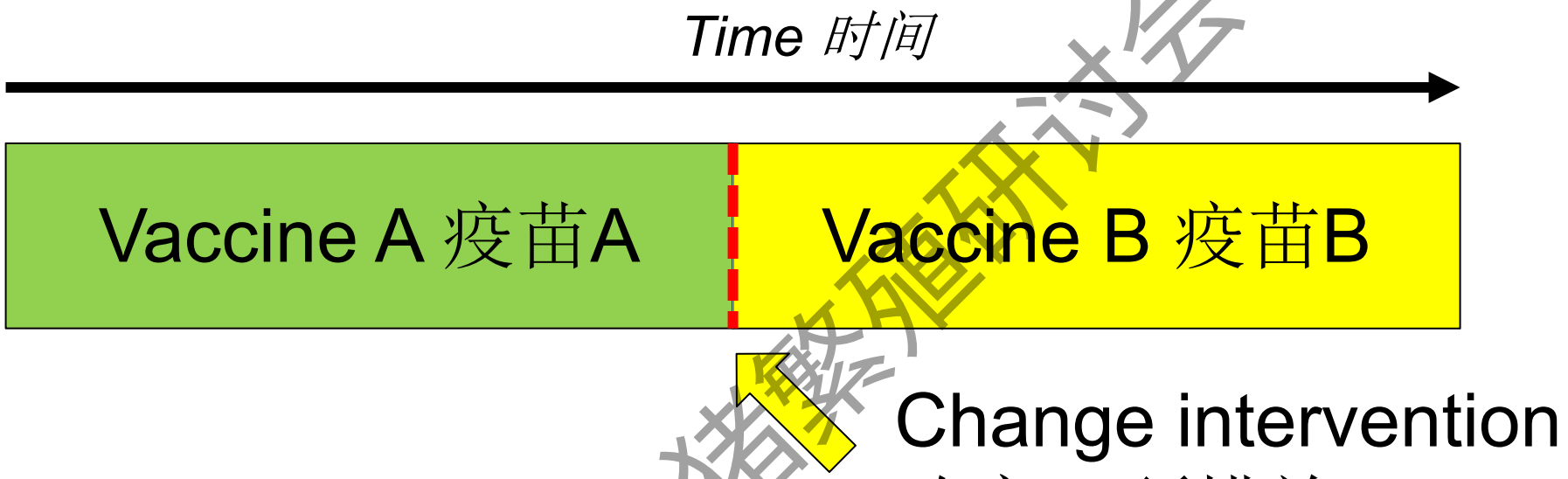
Case Reports and Case Studies
案例报告和案例研究

- **Applications:** 应用：
  - Describe what happened in a series of cases or events that shared common features. 描述在一系列具有共同特征的案例或事件中发生了什么。
  - Describe frequency of a disease or condition, or to describe features related to clinical presentation or disease progression and prognosis.
  - 描述疾病或病症的频率，或描述与临床表现或疾病进展和预后相关的特征。
  - **Hypothesis generating studies.** 假设生成研究。

Before and after trials 试验前后

- **Applications:** 应用：
  - Good for product screening.适合产品筛选。
  - Most useful in demonstrating the **immediate** impacts of short- term programs.
  - 在证明短期计划的**直接**影响方面最有用。
    - **Effect size (large)**of the treatment → may be not need a experimental trial.
    - 治疗效果大小（大）→可能不需要实验性试验。
  - Use **Statistical Process Control (SPC)** to apply some statistical rules.
  - 使用统计过程控制（SPC）来应用一些统计规则。
- **Limitations:** 限制：
  - Less useful for evaluating **longer** term interventions. 在评估长期干预措施方面的作用较小。
  - Validity of the measurement. 测量的有效性。
  - Can be biased by time-frame: season, group disease status, disease progress, **other events that impact outcomes**, etc.
  - 可能因时间框架而产生偏见：季节、群体疾病状态、疾病进展，**其他影响结果的事件**等。
  - **NO CONTROLS** 无控制

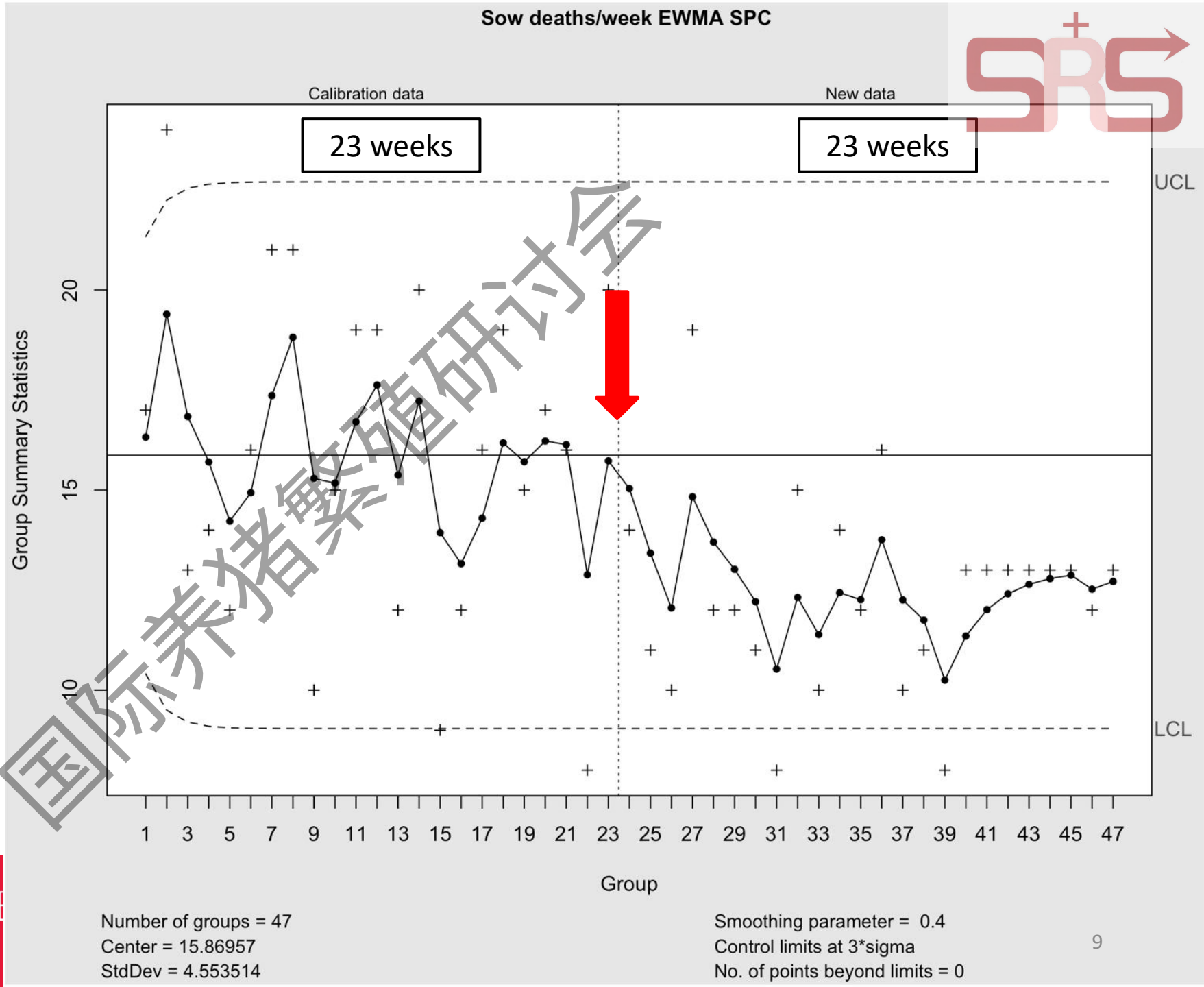
Design and examples设计和示例



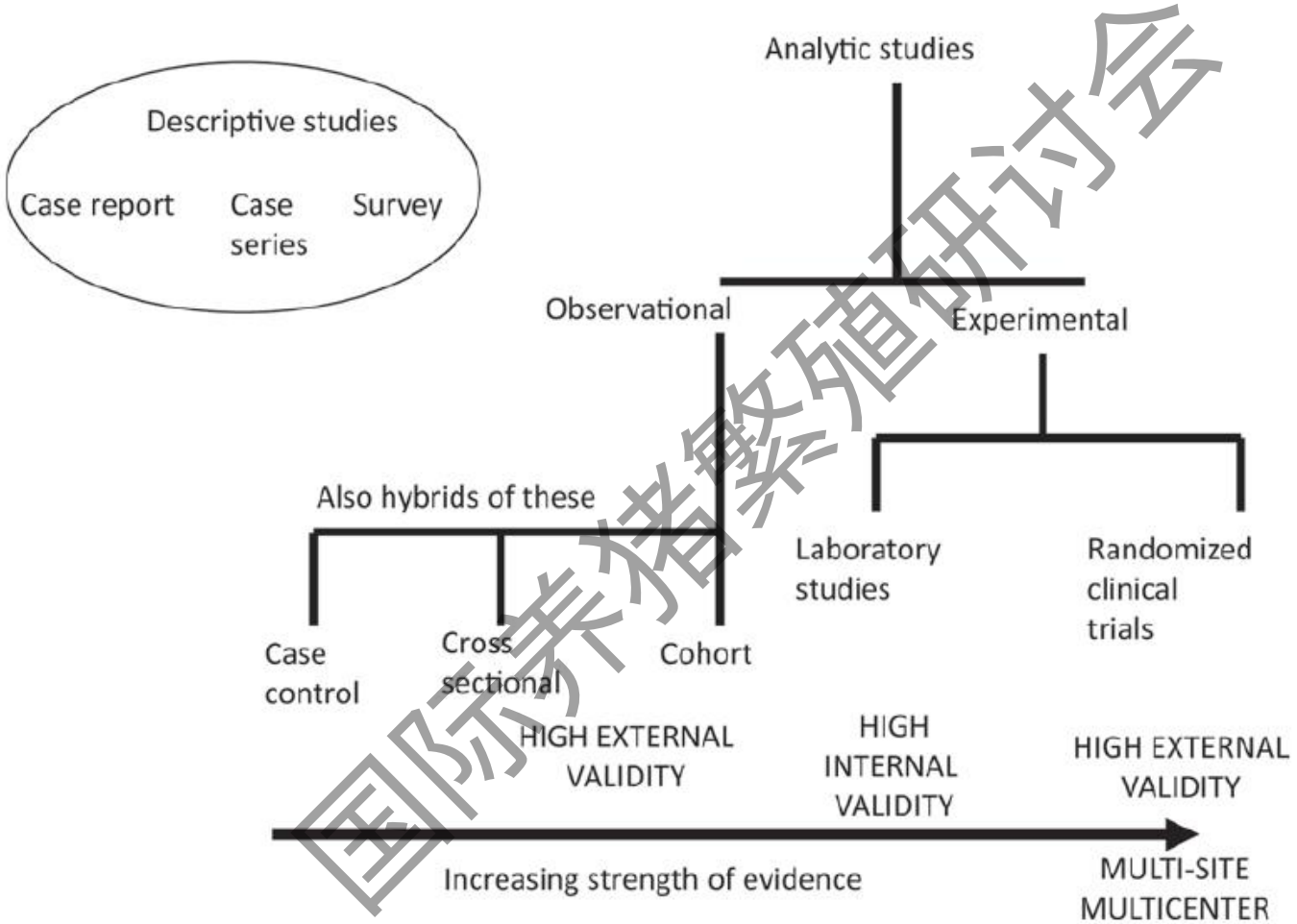
- Vaccines comparison. 疫苗比较。
- Changes on antibiotics. 抗生素的变化。
- Management changes. 管理变更。

Evaluation of Training 培训评估

- Weekly sow deaths per week
- 每周母猪死亡
  - **4.25% reduction in annualized sow mortality**
  - **每年母猪死亡率降低 4.25%**
    - **16.75% to 12.5%**
  - **Chi-squared test for proportions (before and after training)**
  - **比例卡方检验（训练前后）**
    - **p=0.007**



Overview of Epi studies 实验研究概述



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Big difference between an Experimental and Observational study
实验性研究和观察性研究之间的巨大差异

- How the **investigator manipulates** one or more factors (treatment, intervention or exposure) being studied. 研究者如何操纵一个或多个因素（治疗、干预或暴露）。
  - In experimental studies the investigator **manipulates** or **assigns** the treatment 在实验研究中，研究者操纵或分配治疗方法。
    - Treatment is under the control of the investigator. 治疗工作由研究者控制。
  - In observational studies the investigator is an **observer** rather than an agent who manipulates the treatments.
  - 在观察性研究中，研究者是一个观察者，而不是一个操纵治疗的代理人。

Experiments: from single barn to multiple sites
实验：从单个猪舍到多个地点

Allocated by investigator (*random process*)由研究者分配（随机流程）

Why is it important to have treatment allocated by investigator?
为什么采用由研究者分配的治疗方案很重要？

What to do if this is not possible?
如果这是不可能的，该怎么办？

Treatment group
治疗组

Control group
对照组

Follow over time
随时间变化

Measure outcomes
测量结果

Measure outcomes
测量结果

Compare outcomes
比较结果
- ✓ Time-to-event
- ✓ 事件发生时间
- ✓ Risk of event
- ✓ 事件风险
- ✓ Magnitude of event
- ✓ 事件的大小

Great design to: 伟大的设计：
- ✓ Define what is possible to happen under specific (controlled) conditions
- ✓ 定义在特定（控制）条件下可能发生的情况
- ✓ Define what happens under field conditions = clinical trials **[stronger external validity]**
- ✓ 定义=临床试验在现场条件下发生的情况[更强的外部有效性]

E.g., Effect of water soluble antibiotic treatment on barn mortality.
例如，水溶性抗生素处理对猪舍死亡率的影响。

Observational: cohort
观察性：队列

NOT allocated by investigator
未由研究者分配

Treatment not allocated by investigator
未由研究者分配的治疗方法
- Populations (exposed; not exposed) are **not** the same
- 人群（暴露，或未暴露）并不相同
- Adjust for effect of confounder variables such as age, gender, activity level, diet, …
- 调整混杂变量的影响，如年龄、性别、活动水平、饮食等。

Exposed group
暴露组

Unexposed group
未处理组

Follow over time
随时间变化

Measure outcomes
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比较结果
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Prospective or Retrospective design 前瞻性或回顾性设计

Great design to: 伟大的设计：
- ✓ Investigate rare exposures 调查罕见的暴露
- ✓ Investigate multiple outcomes due to a single exposure调查由于单一暴露而导致的多种结果
- ✓ *Prospective design*: more expensive, harder, more complex… also less prone to biases.
- ✓ 前瞻性设计：更昂贵，更困难，更复杂……也不太容易产生偏见。

E.g., Compare the incidence rate of PRRSV outbreaks between filtered farms vs. Non-filtered farms
E. g. , 比较经过过滤的农场与未过滤的农场

Observational: case-control
观察性：病例对照

NOT allocated by investigator
未由研究者分配

Compare outcomes

✓ Magnitude of association (e.g., odds ratios)关联的程度（例如，优势比）

Measure exposures
测量暴露量

Measure exposures
测量暴露量

Ask about prior exposure to 'risk factors'
询问之前接触过"风险因素"

Cases案例
(positive to specific outcome)
（对具体结果积极）

Controls对照
(negative for specific outcome)
（对具体结果阴性）

Great design to:

✓ Assess likelihood of multiple exposures to cause a specific outcome (e.g. disease) 评估多次暴露导致特定结果（如疾病）的可能性

✓ Retrospective design: inexpensive & quick compared to Cohort and Experiments 回顾性设计：与队列研究和实验相比，价格便宜、速度快

E.g., Odds of low-prevalence CNS disease on populations exposed to specific pathogens;
例如，暴露于特定病原体的人群中低流行的中枢神经系统疾病的几率；
Risk factors associated with disease outbreaks in a specific region of interest.
与特定感兴趣区域的疾病暴发相关的风险因素。

Principles of field studies 现场研究原理

- **Define THE question**. *As in experimental studies conducted in controlled conditions (=1 hypothesis at the time), field studies can & should be **focused on 1 question** to allow proper design with reasonable sample size.*
- *定义问题。与在控制条件下进行的实验研究（当时的=1假设）一样，实地研究可以而且应该集中在一个问题上，以允许以合理的样本量进行适当的设计。*
- **Define the metrics**. *What are you going to measure, and effect sizes (e.g., define 'efficacy') 定义指标。你要测量什么，以及效应大小（例如，定义"效能"）*
- **Define populations**. *Characterize external validity, and strategies for selecting the study pop'n) 定义人群。描述外部效度，以及选择研究流行的策略)*
- **Design the study**. *Controls, limit variation and confounders as much as possible at the design. Design what can be executed (limited resources). 设计研究。在设计中尽可能多地控制、限制变化和混杂因素。设计可执行的内容（资源有限）。*

Principles of field studies 现场研究原理

- **Very specific question in mind**... e.g., *prophylactic use of MLV vaccine in neonatal pigs of PRRSv-positive unstable herds that will be placed in high pig dense areas*.
- 这是一个非常具体的问题。例如，在PRRSV阳性的不稳定新生猪群中预防性使用MLV疫苗，这些猪将被放置在高猪密集地区。
    - Are results valid to preventive use of MLV in neonates?
    - 研究结果对新生儿预防性使用MLV是否有效？
    - Can results be extrapolated to vaccinating pigs at weaning?
    - 研究结果可以外推到在断奶时给猪接种疫苗吗？
    - Pigs from PRRSv positive stable herds?
    - 来自PRRSv阳性的稳定畜群的猪？
- Have the **outcome metric(s) in mind**: 要考虑到结果指标：
    - Mortality? ADG? Shedding? 死亡率？ADG吗？排毒？

Principles of field studies 现场研究原理

- Once the question & outcomes are clearly defined: time to **design the study, assuring to control for confounder variables**:
- 一旦问题和结果被明确定义：设计研究的时间，确保控制混杂变量：
  - E.g., if mortality is the outcome of interest: what are other key variables, in that system, playing major role in mortality? Adjust for those in the design… Some examples: 例如，如果死亡率是人们感兴趣的结果：在该系统中，还有哪些其他关键变量在死亡率中起着主要作用？根据设计中的那些因素进行调整。以下一些例子：
    - Randomize treatment, large sample size, close attention to eligibility and exclusion criteria, do not enroll herds clinically active with other non-endemic diseases affecting mortality (PEDV). 随机化治疗、大样本量、密切关注合格性和排除标准，不纳入临床活跃的其他影响死亡率的非地方性疾病（PEDV）的畜群。

Different questions, different needs...

不同的问题，不同的需求。。。

- Understand mode of action了解行动模式
- Understand potential了解潜力
- Test technologies测试技术
- High internal validity内部效度高

合并感染Co-infections

次优的环境条件Sub-optimal environmental conditions

多维问题Multi-dimensional problems

- Measure the true impact (w/confounders)
- 衡量真正的影响（带混杂因素）
- Targeted solutions
- 目标解决方案
- High external validity
- 外部有效性高

Isolation units
(~University settings, small scale)
隔离装置
（~大学设置，小规模）

Commercial research barns
(Controlled commercial barns)
商业研究猪舍
（受控商业猪舍）

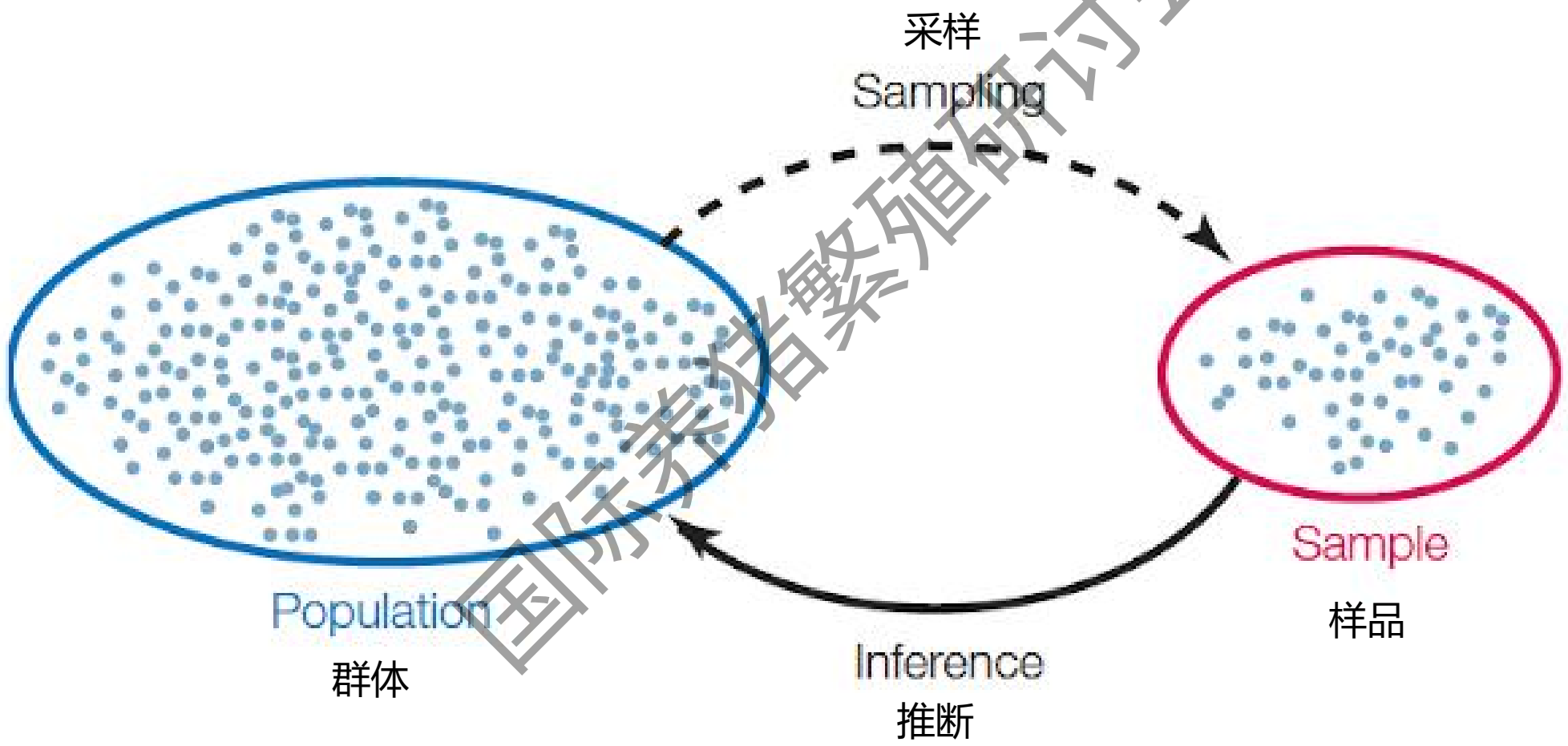
"The field"
(Commercial farms)
"临床"
商业场

Source: Dr. Daniel Linhares

Validity 有效性

- **Definition:** ability of a study to reflect the true state.
- 定义：研究反映真实状态的能力。
- In the context of inference there are two possible concepts of validity:
- 在推理的背景下，有两个可能的有效性概念：
  - **Internal:** results are valid for the source population.
  - 内部：结果对源猪群有效。
  - **External:** results can be generalized/extended to the target population. 外部：结果可以推广到目标。

Correct interpretation and extrapolation of the results! 正确的解释和推断的结果！



Challenges of field-based studies
基于临床研究所面临的挑战

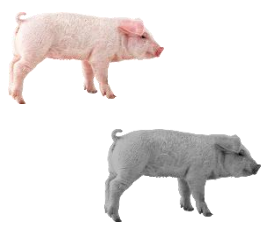
- **Sources of variation.** 变异的来源。
  - E.g. different farms or production flows, regions.
  - 例如，不同的农场或生产流程，地区。
- **Confounders** (factors associated w/ the outcome and treatment and/or covariates) 混杂因素（与结果、治疗和/或协变量相关的因素）
  - E.g. age, barn settings. 例如，年龄，猪舍设置。
- **Interactions**. 相互作用
  - E.g. disease status x age. 例如，疾病状态时的年龄。

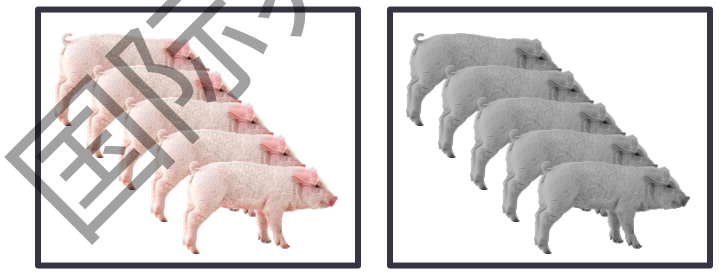
1. Outcome variables 结果变量

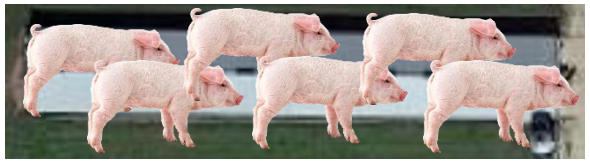
- The most **common issue** in field study design is trying to answer **too many questions** within a single study.
- 实地研究设计中最常见的问题是试图在单一研究中回答太多的问题。
  - Sometimes work as a double-edge sword. 有时是一把双刃剑。
- A good study should be designed to address a specific objective with **one primary variable** of interest.
- 一个好的研究应该设计来解决一个主要感兴趣的变量。
  - At least one variable should have power to detect the expected difference!
  - 至少有一个变量应该有能力检测到预期的差异！
- *Secondary variables* can be assessed, but too much peripheral data can cloud the results and distract from the real outcome.
- 次要变量可以评估，但过多的外围数据会影响结果，分散对真实结果的注意力。

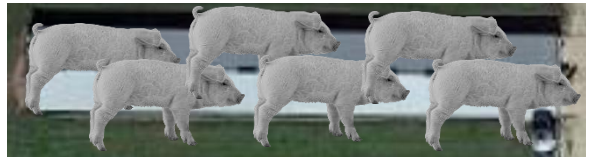
2. Experimental unit (EU) 实验单元（EU）

- The EU is the smallest physical entity to which a treatment is applied. 实验单元是适用于治疗的最小的物理实体。
  - Any experimental unit must be capable of receiving different treatments.
  - 任何实验单元都必须能够接受不同的处理。
  - It is also known as ***unit of analysis*** (observational and hybrid trials).
  - 它也被称为*分析单元*（观察性试验和混合试验）。
  - It is the unit used in the **statistical analysis**.
  - 它是在*统计分析*中使用的单位。









Adapted from Dr. Derald Holtkamp.

Experimental unit 实验单元

- The ***unit of measurement*** or ***observation unit*** are the entities on which the response variable is measured or observed.
- 测量单位或观察单位是用来测量或观察响应变量的实体。
- It is possible that your **experimental unit** and **observation unit** are the same.
- 你的实验单元和观察单元有可能是相同的。
  - E.g.: ADG measured at pen-level, close-out data. E.g.： ADG测量在栏的水平， 近距离数据。
- Sometimes they are different! 有时他们是不同的！
  - E.g.: Mortality measured at the pen or barn-level. E.g.：在栏或猪舍水平上测量的死亡率。
    - Observation unit = pig. 观察单元=猪。

Proper recognition of the **EU** requires a detailed understanding of the **design**.
正确承认实验单元需要对设计的详细理解。

Working example (1) 工作示例（1）

Scenario: you have access to two 2,400 head barns. There are 80 pens with 30 pigs/pen per barn. Pigs in barn 1 are vaccinated with vaccine A, pigs in barn 2 with Vaccine B.

场景：你可以访问两个2400头猪只猪舍。有80个猪圈，每个猪舍有30个猪圈。猪舍1的猪接种疫苗A，猪舍2的猪接种疫苗B。

What is the experimental unit (EU) 实验单元是什么？

A. Pig 猪
B. Pen 栏
C. Side of barn 猪舍旁
D. Barn 猪舍

N=1 per treatment

No replication so no statistical analysis can be performed (summary statistics only)

没有复制，因此无法执行统计分析（仅进行汇总统计）

Working example (2) 工作示例（2）

Scenario: you have access to a single barn constructed with separate feed lines and bins on each side of the barn. There are 20 pens with 25 pigs / pen on each side of the barn (500 / side). Diet A is fed to 20 pens on side 1, and Diet B is fed to 20 pen on side 2. 场景：您可以进入一个单独的猪舍，由单独的饲料线和箱子在猪舍的两侧。猪舍两侧有20个猪栏，25个猪圈(500只）。饲料A喂1边的20个猪栏，饲料B饲喂2侧的20个猪栏。

What is the experimental unit (EU) 实验单元是什么?

A. Pig 猪
B. Pen 栏
C. Side of barn 猪舍旁
D. Barn 猪舍

N=1 per treatment
No replication so no statistical analysis can be performed (summary statistics only)
没有复制，因此无法执行统计分析（仅进行汇总统计）

Working example (3) 工作示例（3）

Scenario: At placement in a wean-to-finish barn 1,000 pigs are randomly assigned to 40 pens (25 pigs / pen). Pens are then assigned to Vaccine A or Vaccine B (20 pens). 场景：在一个潮湿的猪舍中，1000头猪被随机分配到40个猪栏（25头猪/猪栏）。然后将栏分配给疫苗A或疫苗B (20栏）。

What is the experimental unit (EU) 实验单元是什么?

A. Pig 猪
B. Pen 栏
C. Side of barn 猪舍旁
D. Barn 猪舍

N=20

Study has replication, but N=20 pens instead of 500 pigs

研究有复制，但N=20栏，而不是500头猪

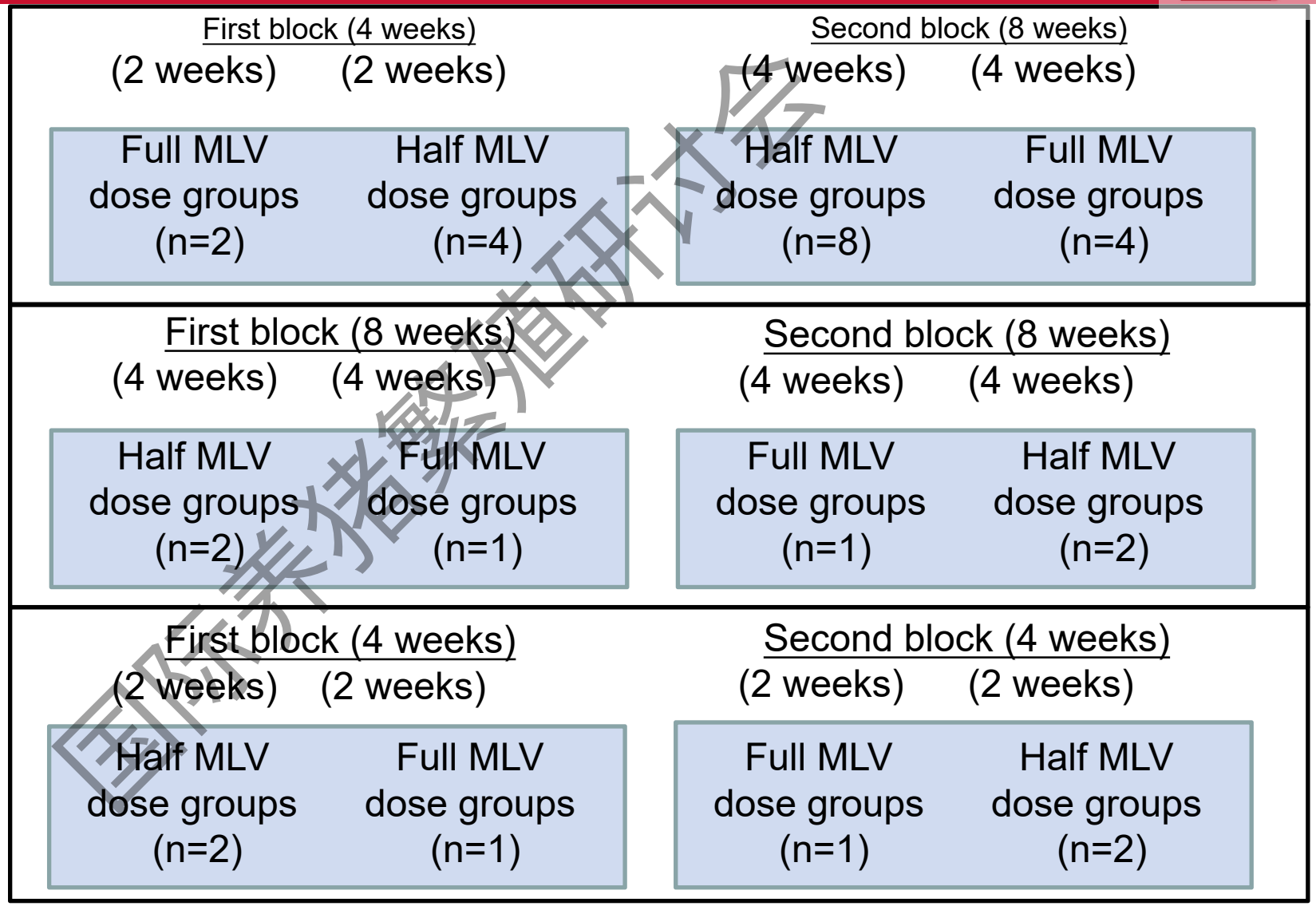
3. Replication of experimental units
复制实验单元：

- Replication = the number of experimental units per treatment group. 复制=：每个处理组的实验单位数。
- Replication is needed to overcome variation.
- 需要复制来克服变异。
- Power calculation: 功率计算：
  - Prior to conducting the trial. 在进行试验之前。
- Avoid **common mistake**: 避免常见错误：
  - The # observational units may not be = # of replicates (EU).
  - #观测单位可能不是重复次数（EU）。

4. Comparable experimental units
可比实验单元

- Another **common issue** is comparing the effect of interventions in different flows. 另一个**常见的问题**是比较在不同的资金流动中所采取的干预措施的效果。
  - Pigs from Farm A - Vaccine A **vs.** Pigs from Farm B - Vaccine B
  - 农场的猪疫苗A vs 农场B疫苗的猪
- Experimental units must be comparable: 实验单元必须具有可比性：
  - Different flows can have different health challenges.
  - 不同的资金流动可能会有不同的健康挑战。
  - Avoid confounders (e.g. weaning age, size of barn, health status).
  - 避免出现混杂因素（如断奶年龄、猪舍大小、健康状况）。
- Possible solutions to overcome this problem: 克服此问题的可能解决方案：
  - Create **pairs** (*blocks*) of experimental units from the same source (e.g. randomize based on weaning age, flow, parity).
  - 从同一来源创建实验单元对（块）（例如，根据断奶年龄、猪流、胎次进行随机化）。

Working example: 工作示例：



Source: Moura, 2021 (submitted).

5. P-value misconceptions P值误解

1. Not always a **significant** *p-value* is considered clinical or economical significant, and vice-versa. 并非总是显著的p值被认为具有临床或经济意义，反之亦然。
   - The outcome must help on the decision making process. 研究结果必须有助于决策过程。
2. If the *p-value* is greater than 5%, the treatment/intervention has **no effect**. 如果p值大于5%，则治疗/干预没有效果。
   - If the treatment has a smaller effect size, a study with a small sample may not have enough power to detect it. 如果治疗的效果较小，那么用小样本进行的研究可能没有足够的能力检测到它。
3. Having a **lower** *p-value (e.g. 0.00001)* does not mean a big difference between treatments. 具有较低的p值（例如0.000001）并不意味着处理之间的很大差异。
   - P-value is impacted by the sample size. P值受样本量的影响。

5. P-value P值

- **Avoid** looking at the *p-value* alone! 避免只看p值！
- **MUST LOOK** at the *clinical/economical significance*.
- 必须关注其临床/经济意义。
- Take your decision based on: 根据以下依据：
  - **Mean values** for each group. 每一组的平均值。
  - The **standard error** of the mean (SEM or SE). 平均值的标准误差（SEM或SE）
  - The **95% confidence interval. 95%置信区间。**
  - The ***p-value***. p值。
  - **Economics**. 经济

Take home message 总结

- Lack of attention when designing trials can waste resources and time.
- 在设计试验时缺乏关注可能会浪费资源和时间。
  - A trial needs to answer at least one good answer. 一个试验需要回答一个好的答案。
  - Sometimes experimental unit is not the same as observation unit. 有时实验单元和观测单元不一样。
  - Replication is needed to overcome biological variation. 需要复制来克服生物变异。
  - Experimental units need to be comparable to avoid add different sources of variation. 实验单元需要具有可比性，以避免增加不同的变异来源。
  - Don't focuses to much on p-values.不要太关注p值。

Questions?? 问题

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