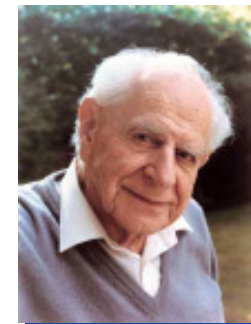


Unequal carry-over – “solved” in BE but still an Issue in Assessing Biosimilarity?

Helmut Schütz

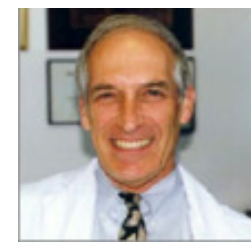
To bear in Remembrance...

Whenever a theory appears to you as the only possible one, take this as a sign that you have neither understood the theory nor the problem which it was intended to solve.



Karl R. Popper

Even though it's *applied* science we're dealin' with, it still is – *science!*



Leslie Z. Benet

Sequence Effect

Better: Unequal carry-over

- **Standard 2×2×2 cross-over design**
 - Subjects' responses in the second period in sequence RT are different from the ones in sequence TR.
 - The sequence effect is confounded with
 - the carry-over effect, and
 - the formulation-by-period interaction.
- **Therefore, a statistically significant sequence effect could indicate that there is**
 - a true sequence effect,
 - a true carry-over effect,
 - true formulation by period interaction, or
 - a failure of randomization.

Sequence Effect

‘Two-stage analysis’¹

- Was applied in the past
 - Test at α 0.10 (low sensitivity since this is a between-subject term).
 - If $p < 0.1$, evaluation of the first period’s data as a parallel design.
 - Extreme loss in power.
 - Example: CV_w 0.25, CV_p 0.50, GMR 0.95, n 28
 - power of 2×2×2: 0.8074
 - power of first period’s data: 0.001585 (!)
- Procedure was demonstrated statistically flawed.²
 - Inflated Type I Error.
 - Biased estimate.

1. Grizzle JE. *The Two-Period Change-Over Design and Its Use in Clinical Trials*. Biometrics. 1965;21(2):467–80. [doi:10.2307/2528104](https://doi.org/10.2307/2528104).
2. Freeman P. *The performance of the two-stage analysis of two-treatment, two-period cross-over trials*. Stat Med. 1989;8(12):1421–32. [doi:10.1002/sim.4780081202](https://doi.org/10.1002/sim.4780081202).

Sequence Effect

Nuisance

- No procedure exists to correct for a true sequence / unequal carry-over effect.^{2,3}
- Significant sequence effects were found in a large metastudy⁴ at about the level of the test, both for AUC and C_{max}
 - $2 \times 2 \times 2$ studies (n=324, α 0.10)

AUC	34 (10.5%)	C_{max}	37 (11.4%)
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 - 6×3 studies (n=96, α 0.05)

AUC	4 (4.2%)	C_{max}	4 (4.2%)
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 - As expected, the distribution of p values followed closely uniform [0, 1].
- Confirmed (20 studies from the public doamin and 165 from BEBAC's database; AUC).

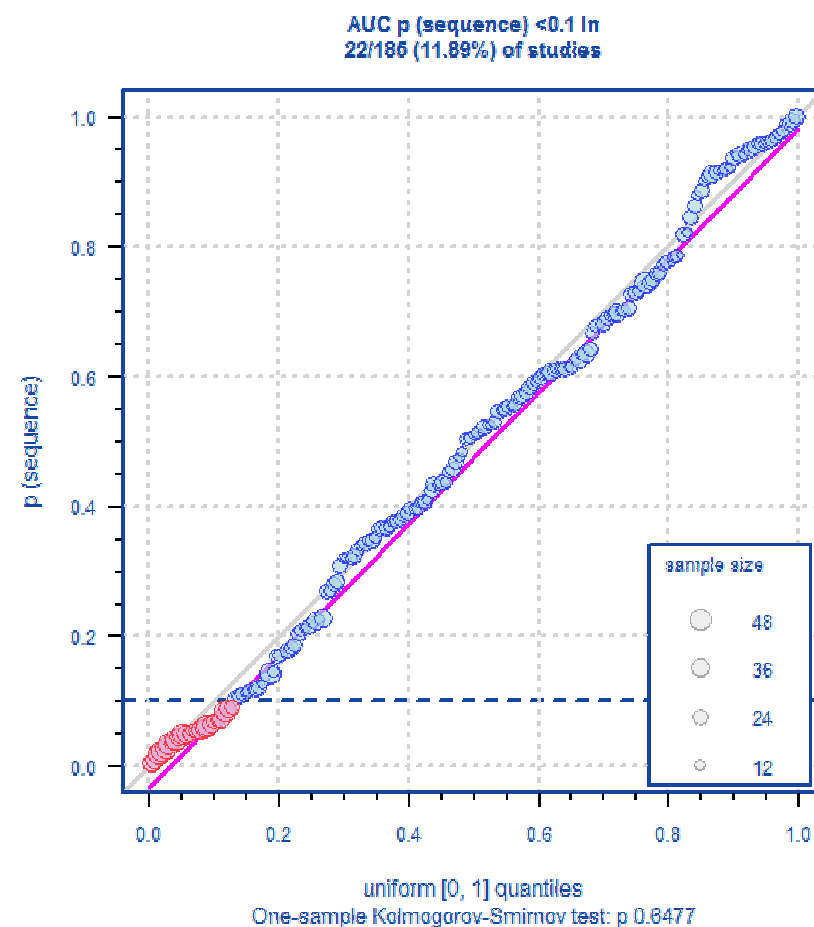
3. Senn S. *Cross-over Trials in Clinical Research*. Chichester: Wiley; 2nd ed. 2002.

4. D'Angelo G, Potvin D, Turgeon J. *Carry-over effects in bioequivalence studies*. *J Biopharm Stat.* 2001;11(1–2):35–43. [doi:10.1081/BIP-100104196](https://doi.org/10.1081/BIP-100104196).

Sequence Effect

Nuisance

- Significant sequence effects in properly planned studies could be considered a statistical artifact (significant results are likely false positives).
- A true sequence/carry-over is highly unlikely in a BE study if
 - the study is performed in healthy subjects,
 - the drug is not an endogenous entity, and
 - an adequate washout period was maintained.



Review of Guidelines

EMA

- **BE-GL (2010)**
 - A test for carry-over is not considered relevant and no decisions regarding the analysis (e.g. analysis of the first period only) should be made on the basis of such a test. The potential for carry-over can be directly addressed by examination of the pre-treatment plasma concentrations in period 2 (and beyond if applicable).
- **Clinical Investigation of the PK of Therapeutic Proteins (2005)**
 - The ordinary cross-over design is not appropriate for therapeutic proteins with a long half-life, e.g. therapeutic antibodies and pegylated proteins, or for proteins for which formation of anti-drug antibodies is likely.
- **However, in many of the product-specific guidelines a cross-over design is recommended.**

Recap

A true sequence/carry-over is highly unlikely if

- the study is performed in healthy subjects,
- the drug is not an endogenous entity ...

Always remember:

Pharmacokinetics may be simply defined as what the body does to the drug, as opposed to *pharmacodynamics* which may be defined as what the drug does to the body.⁵

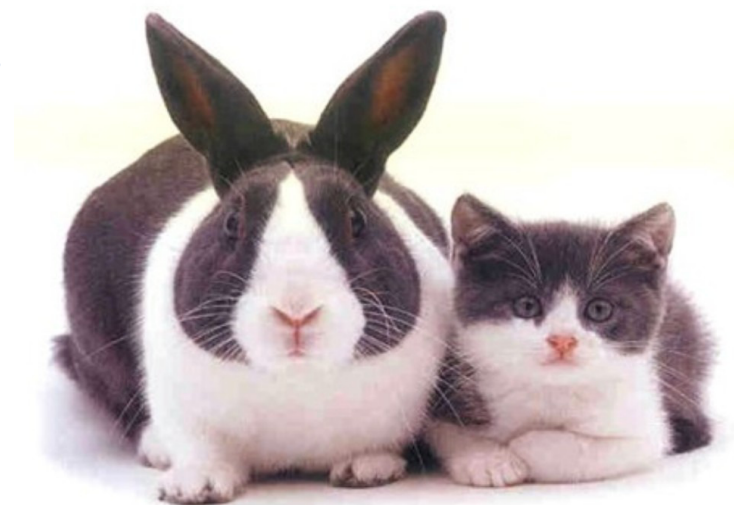
I'll give you my gun when you take it from my cold, dead hands.

5. Benet LZ. *Pharmacokinetics: Basic Principles and Its Use as a Tool in Drug Metabolism*. In: Mitchell JR, Horning MG, editors. *Drug Metabolism and Drug Toxicity*. New York: Raven Press; 1984. p. 199.

Observations / Concerns

Biosimilar Studies in a cross-over

- **Observations**
 - All I have seen showed a highly (!) significant sequence effect.
 - Almost in all a highly significant sequence effect was observed (János Borvendég, personal communication 2014).
- **Concerns**
 - I would be very wary performing studies of biosimilars in a cross-over – even if recommended in a product-specific guideline.
 - Absence of evidence \neq evidence of absence!
 - Assessing relevance? ⁶



6. Ocaña J, Sanchez O MP, Carrasco JL. *Carryover negligibility and relevance in bioequivalence studies.* Pharmaceut Stat. 2015;14:400–8. [doi:10.1002/pst.1699](https://doi.org/10.1002/pst.1699).

Are Parallel Designs the Solution?

In principle, yes.

- **Drawbacks**
 - Sample sizes much higher than in cross-overs.
 - Requires careful selection of subjects (anthropometric data, genotyping recommended, ...) in order to allow an unbiased estimate of the treatment effect.
 - Doubtful whether agencies would accept reference-scaling.
The current definition of HVD(P)s is based on within-subject variability.
- **For the courageous ones**
 - State in the SAP that you will evaluate the study as ‘matched pairs’ (suggested by Stephen Senn).
 - Power close to cross-over.
 - Scientific advisory meeting with the EMA mandatory.

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Thank You!
Open Questions?



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