https://www.fda.gov/downloads/AnimalVeterinary/NewsEvents/WorkshopsConferencesMeetings/UCM444127.pdf

In Vitro Bioequivalence (BE) Pathways

Marilyn Martinez, *Ph.D., Senior Biomedical Research Scientist, Office of New Animal Drug Evaluation, Center for Veterinary Medicine, FDA*

Agenda for Today's Discussion

Goal:

1. Discuss the foundational principles for using in vitro release methods and product physico-chemical understanding as a mechanism for assessing product BE for non-systemically absorbed dosage forms.
2. To present a roadmap for determining in vitro BE for the purpose of providing a starting point for discussion.

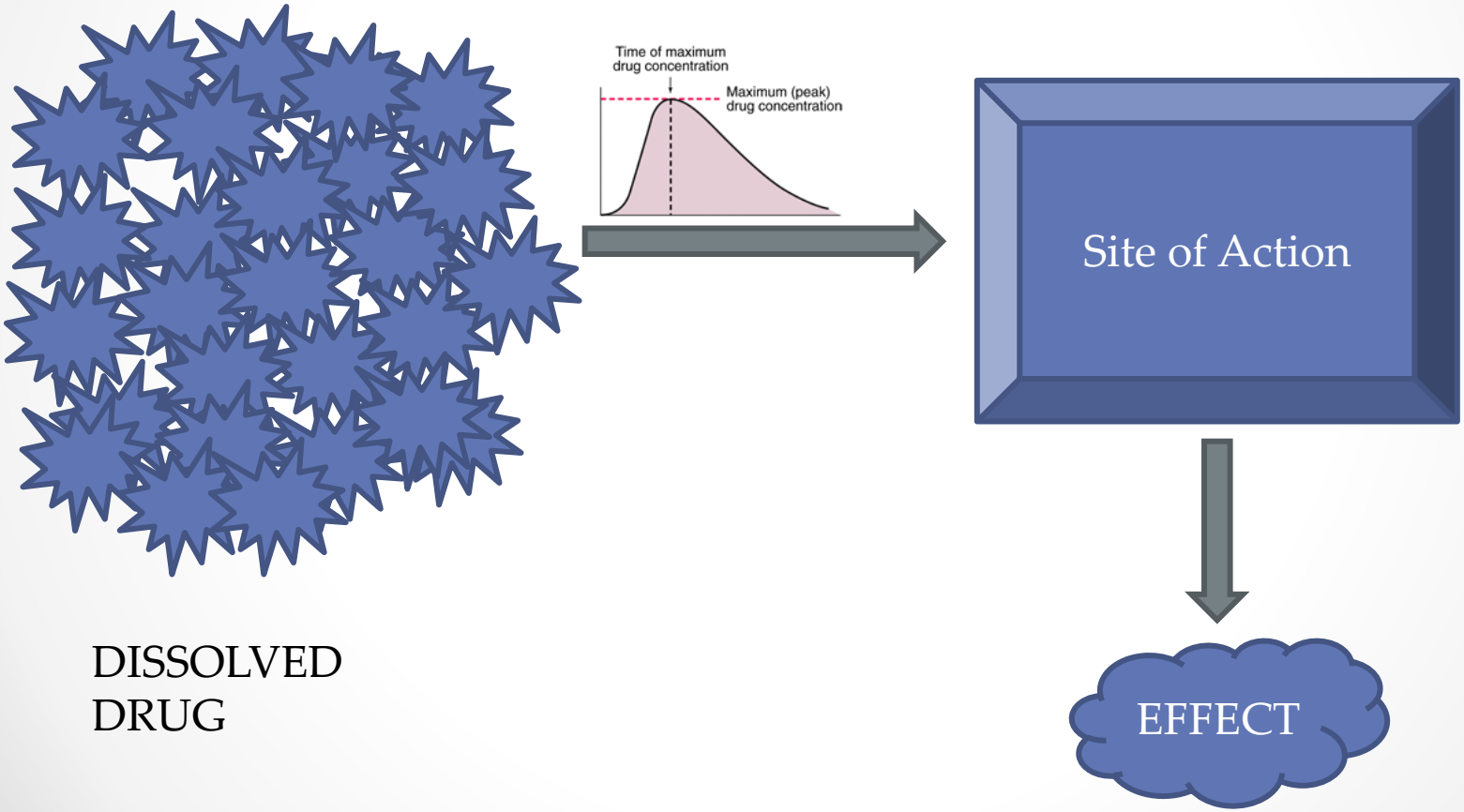
Background

Legal Considerations: Bioavailability Definition
Human Drug Regulations

21 CRF §320.1: Bioavailability is the rate and extent to which the active ingredient or active moiety is absorbed from a drug product and becomes available at the site of action.

Blood level (pharmacokinetic) studies are the "gold standard" for comparing products that are systemically absorbed.

Underlying Concept in Bioavailability Testing

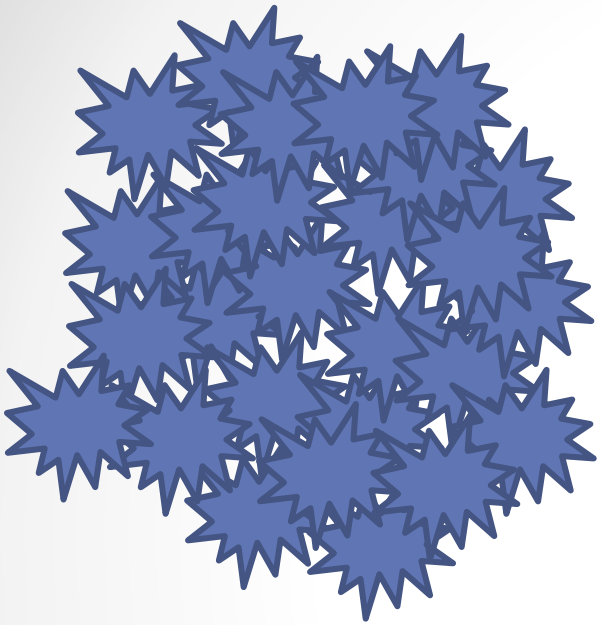


Legal Considerations: Bioavailability Definition
Human Drug Regulations

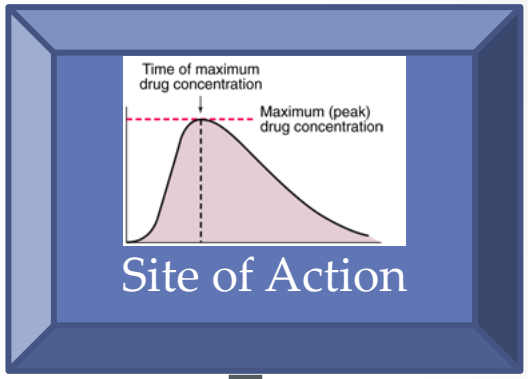
21 CRF §320.1: For drug products that are not intended to be absorbed into the bloodstream, bioavailability may be assessed by measurements intended to reflect the rate and extent to which the active ingredient or moiety becomes available at the site of action.

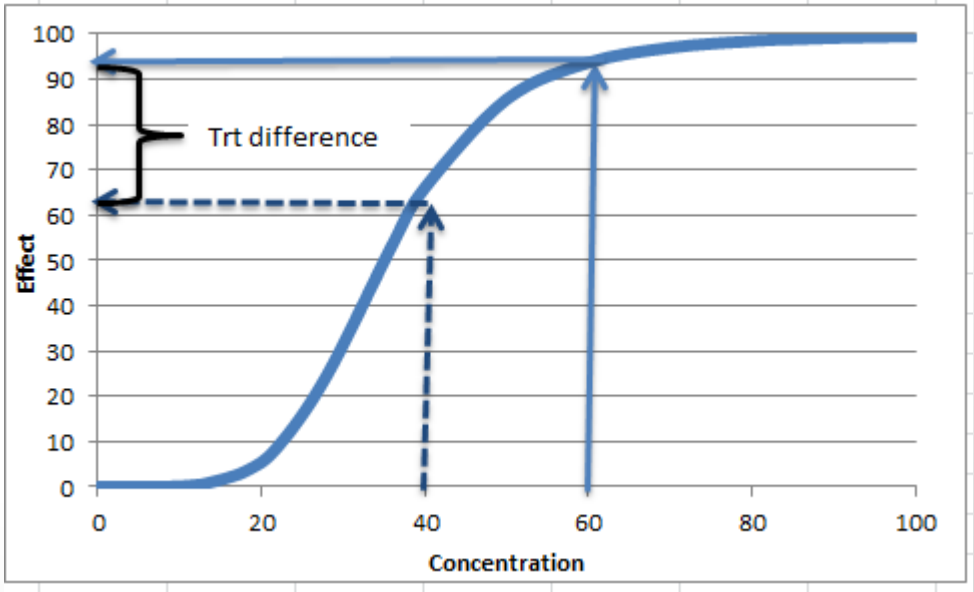
Within CVM, the evaluation of BE for nonsystemically absorbed drug products has typically been accomplished through the submission of clinical endpoint BE trials.

Non-Systemically Absorbed Drugs



DISSOLVED DRUG



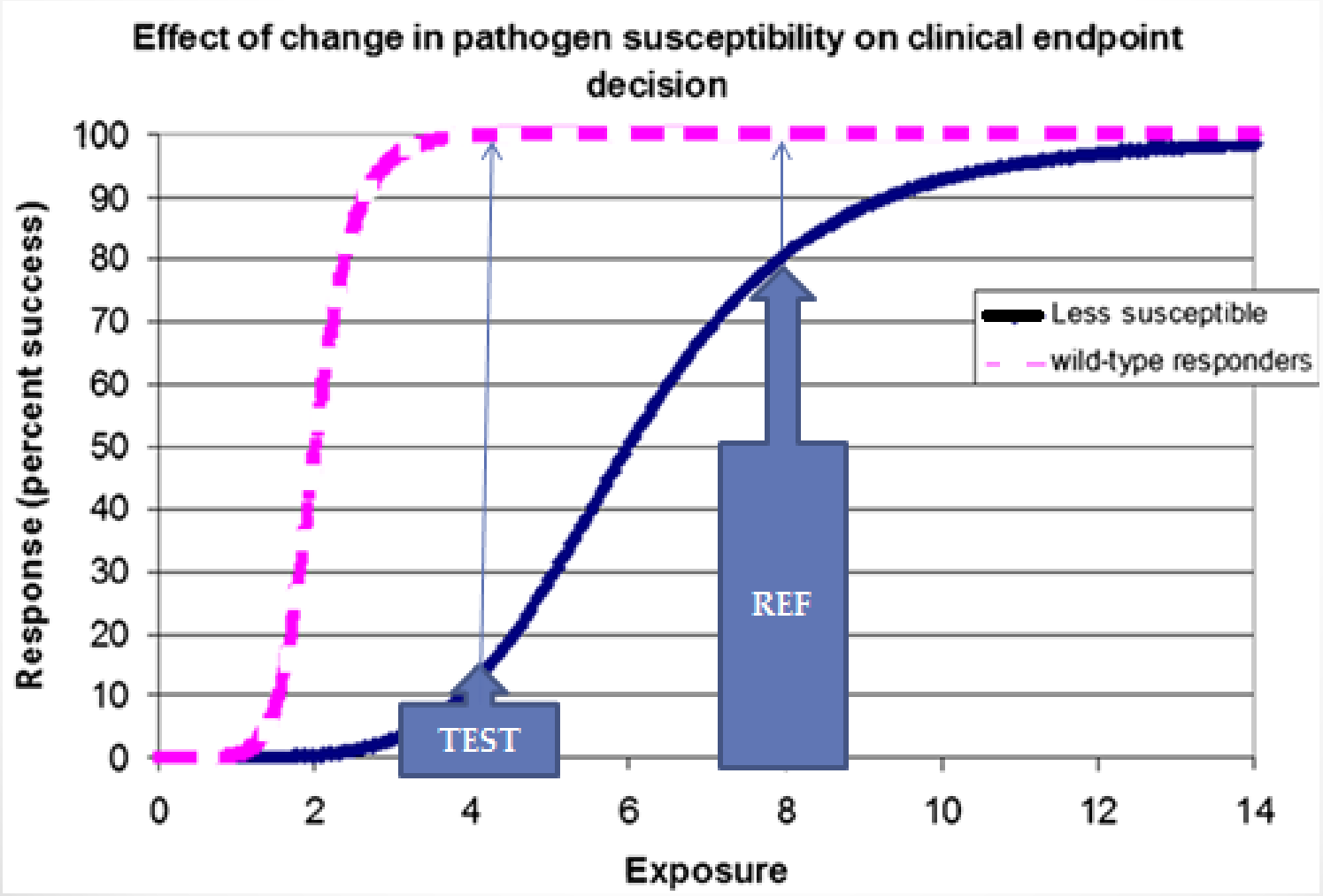


Challenges of Clinical Endpoint Studies

- Clinical endpoints more variable than PK but must meet the established BE limits.
- Difficult to achieve consistency between studies
  - study design
  - study population
  - BE endpoints
- Some products require multiple studies
- May require several hundred patients
- Study duration may be several weeks depending upon the approved labeling
- Very expensive to conduct

Based upon summary presented by Dena R. Hixon, M.D., Associate Director of Medical Affairs, Office of Generic Drugs, CDER. But equally applicable to veterinary medicine.

Another Challenge: Potential Shift in Exposure-Effect Relationship can Alter BE Conclusions:



Statutory Support for Alternative BE Criteria (human drugs)

A 2003 addition to the Federal FD & C Act at Section 505 (j)(8)(A)(ii) indicates that “For a drug that is not intended to be absorbed into the bloodstream, the Secretary may assess bioavailability by scientifically valid measurements to reflect the rate and extent to which the active ingredient or therapeutic ingredient becomes available at the site of drug action”.

CVM Initiative for Addressing this Challenge

By combining an understanding of drug physico-chemical properties, in vivo drug and drug product solubilization, and formulation characteristics, we can determine whether or not two formulations of a nonsystemically absorbed drug (active pharmaceutical ingredient) will be biologically equivalent **WITHOUT** the need for a clinical endpoint BE trial.

Points For Discussion

Perspectives

- The following portion of this presentation provides **perspectives** on this novel in vitro BE approach[1].
  - o When it can be applied
  - o Conditions for testing
  - o Criteria for determining product BE

We invite your insights and views on these proposals.

[1] The following slides provide suggestions and are intended to serve as a springboard for discussion. Unless otherwise indicated, the contents of these slides should not be construed as FDA-CVM guidance or recommendations .

Roadmap for the In Vitro BE Approach

- Confirm that the drug is not systemically absorbed.
- Describe the physico-chemical characteristics of the drug and the drug in the dosage form (this includes in vitro drug release characteristics).
- Compare the proposed product formulation to the reference formulation.

Potential Situations where an In Vitro BE Determination May be of Use

- Generic drug applications.
- When the innovator wishes modify an existing formulation.
- When BE needs to be determined for a combination drug product where one component is systemically absorbed (i.e., necessitating an in vivo blood level BE assessment) and the other is a locally acting compound.

What In Vitro BE Covers

- Target animal safety
- Clinical effectiveness

What In Vitro BE Does Not Cover:

- Human food safety
- Environmental assessment

Benefits of an In Vitro BE Approach

Removes roadblocks to the approval of locally acting animal drugs while maintaining a high level of scientific scrutiny to ensure “sameness”:

- Generics
- Pioneer products with modifications appropriate for the application of this approach

CVM Proposed Approach

Stage 1: Manufacturing (formulation) and physico-chemical characterization. This is based upon evaluation of “Q1, Q2, and Q3” (to be discussed by Dr. Fahmy).

Once the criteria for demonstrating comparability based upon physico-chemical characterization are met, the assessment can move to Phase 2.

CVM Proposed Approach

Stage 2: Demonstrating comparable product release characteristics.

The basis of this in vitro approach is that through an understanding of the in vivo fluid composition and the hydrodynamics of the in vivo and in vitro systems, we can define a set of in vitro conditions that allows for the discrimination of inequivalent formulations.

When used to meet the criteria of in vitro BE, the in vitro dissolution study must be conducted in accordance with GLP regulations.

Candidates for In Vitro BE Applications

- Disintegrated dosage forms (e.g., Type A medicated articles)
- Suspensions
- Emulsions
- Ointments
- Creams

Thus, the in vitro BE approach can be used for products intended to be delivered orally, topically, and by intramammary infusion when these products are associated with little to no systemic absorption.

How Might We Generate the Equivalence Evaluation of In Vitro Drug Release?

Suggested Paradigm for Discussion

- To determine the appropriate test conditions for comparing two products, sponsors generate in vitro release profiles of the reference product across a range of conditions.
- Based upon these results, the sponsor proposes the in vitro conditions under which the pivotal product comparisons will be generated.

Point for discussion: how many conditions need to be tested to insure that the two products will perform comparably across the range of potential in vivo environments?

Suggested Paradigm for Discussion

- Selected conditions should insure formulation-limited dissolution (i.e., conducted under sink conditions).
- For one of these conditions, ≥85% of the reference product should be dissolved (timeframe for achieving this is based upon dosage form stability under the conditions of the assessment).
  - Profiles should follow an Emax or sigmoidal Emax model (approximately 4 or more timepoints prior to peak % dissolved).
  - Test of discriminative characteristics through test of multiple formulations rarely feasible.

Suggested Paradigm for Discussion

- The dissolution media should reflect the range of in vivo conditions to which the dosage form will be exposed (e.g., pH).
- The use of surfactants is acceptable for poorly soluble compounds (beware of foaming). See Noory, C et al., 2000 http://www.dissolutiontech.com/DTresour/200articles/200art3.html

Suggested Paradigm for Discussion

Numbers of lots needed are a point for discussion.

- Based on the selected pivotal test conditions, the in vitro dissolution profiles for the reference product would be evaluated across multiple lots (to characterize reference product inter-lot variability).
- If possible, multiple lots of the test product (or beginning, middle and end of a production run) would also be evaluated.
- For medicated articles, it is recognized that multiple test product lots may not be available. Accordingly, replication of “stock samples” can reflect bags that have been exposed to shipping conditions, with samples of these bags taken from the top, middle and bottom portions of these bags.

Dissolution Considerations

Method:
Apparatus: There are numerous USP apparatus's that can be appropriate for testing the veterinary dosage form. Method used should reflect the dosage form and the physico-chemical characteristics of the drug.

Description and potential concerns:
http://www2.aaps.org/uploadedFiles/Content/Sections_and_Groups/Focus_Groups/In_Vitro_Release_and_Dissolution_Testing/Resources/IVRDTFGStippler2011.pdf

USP general chapter 711
http://www.usp.org/sites/default/files/usp_pdf/EN/USPNF/revisions/m99470-gc_711.pdf

Compendial Apparatus' (USP <711>

UPS Apparatus	Common Name	Drug Formulation Tested	Reference
I	Rotating Basket	Floating dosage forms such as encapsulated products. Also used for swelling dosage forms, bead formulations, coated and uncoated tablets, suppositories, and some immediate and modified release formulations	Crist and Walker. http://www.chem.agilent.com/Library/articlereprints/Public/Back_to_Basics2.pdf
II	Paddle	Solid dosage forms such as tablets and capsules, or particulate dosage forms such as suspensions and powders	Erika Stippler http://www2.aaps.org/uploadedFiles/Content/Sections_and_Groups/Focus_Groups/In_Vitro_Release_and_Dissolution_Testing/Resources/IVRDTFGStippler2011.pdf
III	Reciprocating Cylinder	Tablets, capsules, beads, chewable products, animal feeds. Not suitable for products that disintegrate into small particles	Ken Boda https://www.chem.agilent.com/Library/eseminars/Public/Developing%20Methods%20for%20Apparatus%203%20and%207.pdf
IV	Flow Through Cell	Different types of cells are available for testing tablets, suspensions, powders, suppositories, hard- and soft-gelatin capsules, implants, semisolids, suppositories, and drug-eluting stents.	Nikoletta Fotaki http://www.dissolutiontech.com/DTresour/201111Articles/DT201111_A06.pdf

Dissolution Considerations

Conditions:

- Temperature: 37^0 C. or higher as relevant for the particular animal species.
- Agitation speed:
  - USP Apparatus 1: Generally maintained between 75 -150 rpm
  - USP Apparatus 2: Generally maintained between 50 -100 rpm.
  - Other USP Apparatus': Justification for conditions selected should be provided in the application.

Suggested Test Considerations

For each lot or “stock sample”:

- The in vitro data should be generated on 12 replicates per treatment (see the CDER’s 1997 dissolution guidance). http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM070237.pdf,
- The dissolution profiles would be characterized under each of the selected in vitro test conditions (e.g., 3 conditions?).

Pivotal In Vitro Test

Across the multiple lots of test and reference product (i.e., reflecting both inter and intra-lot variability), the criteria for applying the F2 approach is that the variability about the mean percent released for each product and at each timepoint must be ≤10% (≤20% at the first timepoint). If this criteria is met for the test and reference products , then comparability can be based upon use of the F2 metric. {Criteria and analysis for this test are based upon the 1997 CDER guidance}.

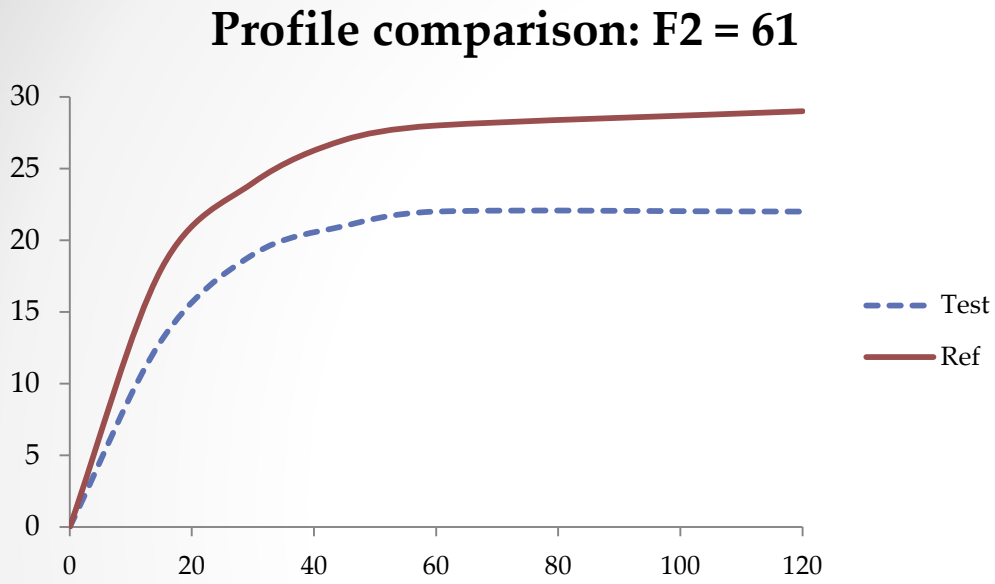
Pivotal In Vitro Test: F2

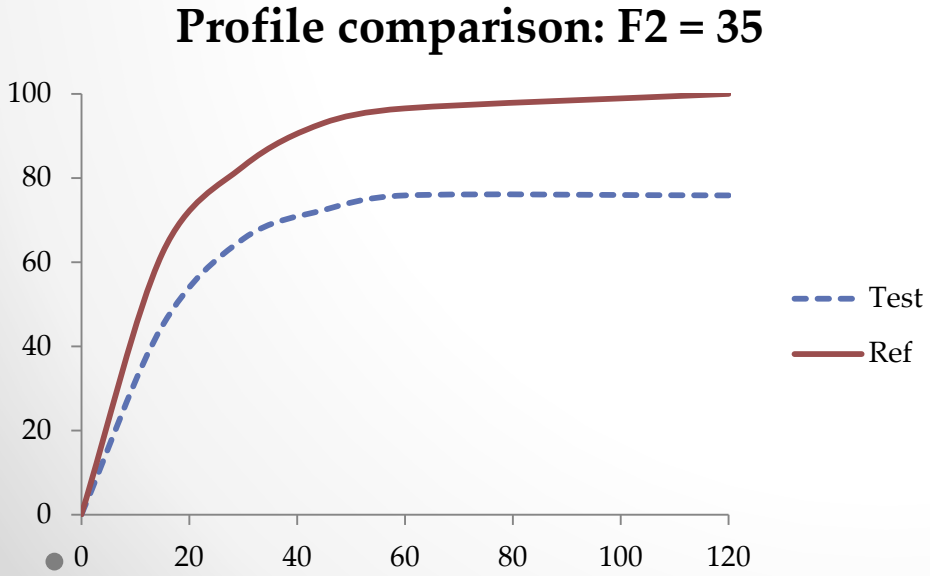
Within a given dissolution test condition, F2 can be calculated based upon the mean of all observations generated within a given formulation (e.g., # lots of the reference product * 12 replications per lot = number of observations included at each timepoint).

Word of Caution When Applying F2

When applying F2 to conditions associated with low in vitro release (<85% dissolved), the F2 value can inappropriately suggest comparability. Therefore, **we are suggesting that** in these situations, the maximum % dissolved values should be normalized to 100% and the corresponding correction made in all of the observed concentrations.

Example: Low %Dissolved and Inequivalent





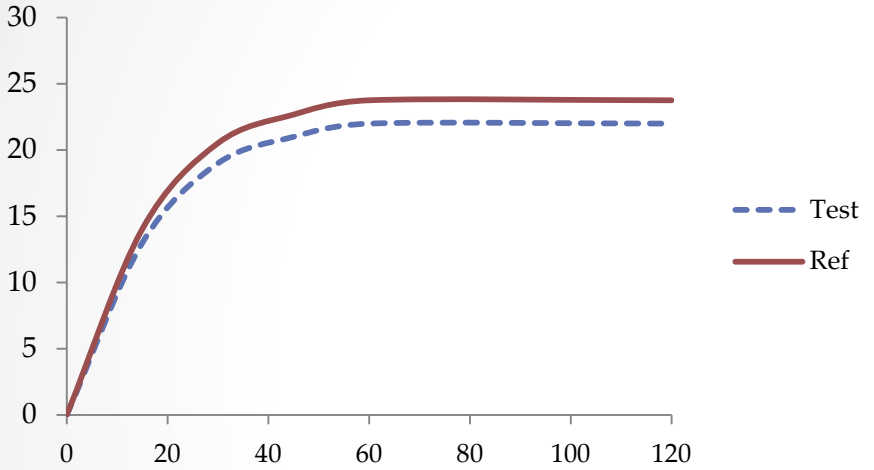
In the original dataset, the maximum amount dissolved (Ref) was 29%. Here we see marked differences in the test and reference profiles, but when applying F2 to the original data, the products would be declared equivalent.

In contrast, when we take the same dataset and correct each value by 100/29, we see that the relative profiles did not change, but the F2 metric clearly identifies the two profiles as being inequivalent.

Example: Low %Dissolved and Equivalent

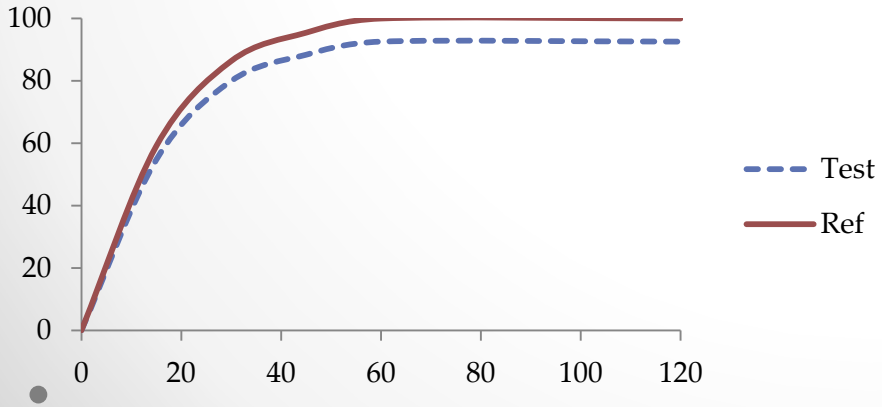
Profile comparison: F2 = 86

T/R at each point = 1.08



Profile comparison: F2 = 59

T/R at each point = 1.08



In the original dataset, the maximum amount dissolved (Ref) was 24%. Here we see that the test and reference profiles are similar (Test/Ref ratio at each point was 1.08. The F2 value was quite high, signifying equivalence when using the original data.

Similar, when we take the same dataset and correct each value by 100/24, we see that the relative profiles did not change, but the F2, although reduced, still identifies the two profiles as being equivalent.

Suggestions Continued: Pivotal In Vitro Release Test

- If the reference product meets the criteria for applying F2 but the test product does not, this constitutes a failure to demonstrate in vitro comparability.
- When the reference product itself exhibits extensive between-lot or within lot variability, the F2 metric should not be applied. In this situation, the test product needs to be compared to the reference product on the basis of alternative statistical methods (CDER in vitro dissolution guidance, 1997).

Suggestions Continued: Pivotal In Vitro Release Test

- Considering the need for the comparison of profiles across multiple test and reference lots, **we suggest** that when the F2 approach cannot be applied, the comparability assessment (under each of the pivotal in vitro test release conditions) be based upon the **90% tolerance interval (calculated for the test and for the reference formulations with 90% confidence)**.

What are the 90% Tolerance Limits?

- 90% Tolerance limits for a **measured quantity:** The limits within which **a specified portion of the population will fall**, where those limits are estimated with a 90% level of confidence.
- 90% Confidence interval for an **estimated parameter** *(e.g., mean)*: if the same population is sampled on numerous occasions and interval estimates are made on each occasion, the resulting **intervals would bracket the true population parameter** in approximately 90% of the cases.

NIST/Sematech *Engineering Statistics Handbook*, Section 7.2.6.3

What is a 90% Tolerance Interval?

- Tolerance interval **width** is a function of sampling error and the **true population variance**. As the sample size increases towards that of the true population, the tolerance interval will approach the true population variance.

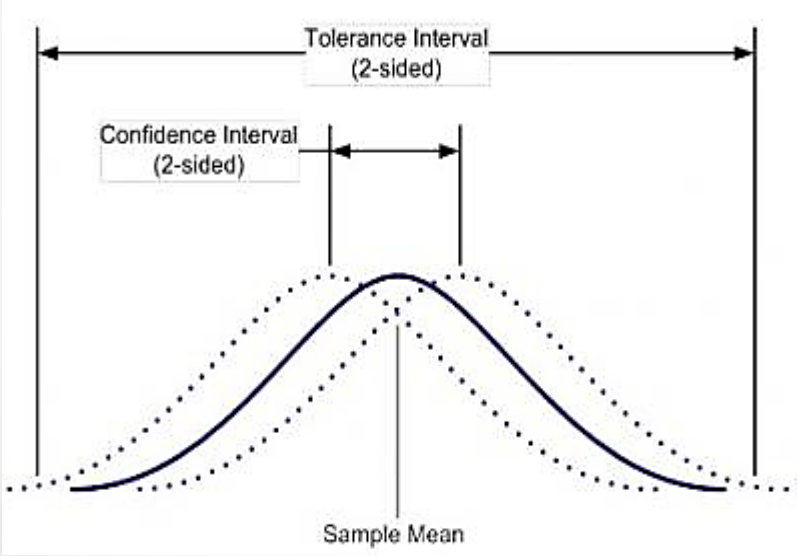
- Confidence interval **width** is a function of sampling error. As sample size increases towards that of the true population, the width of the confidence interval will approach zero.

NIST/Sematech *Engineering Statistics Handbook*, Section 7.2.6.3

Tolerance Intervals are Wider than Confidence Intervals

Solid line: true hypothetical population distribution.

Dotted lines: distributions reflecting the uncertainty of knowing the true location of the population mean.

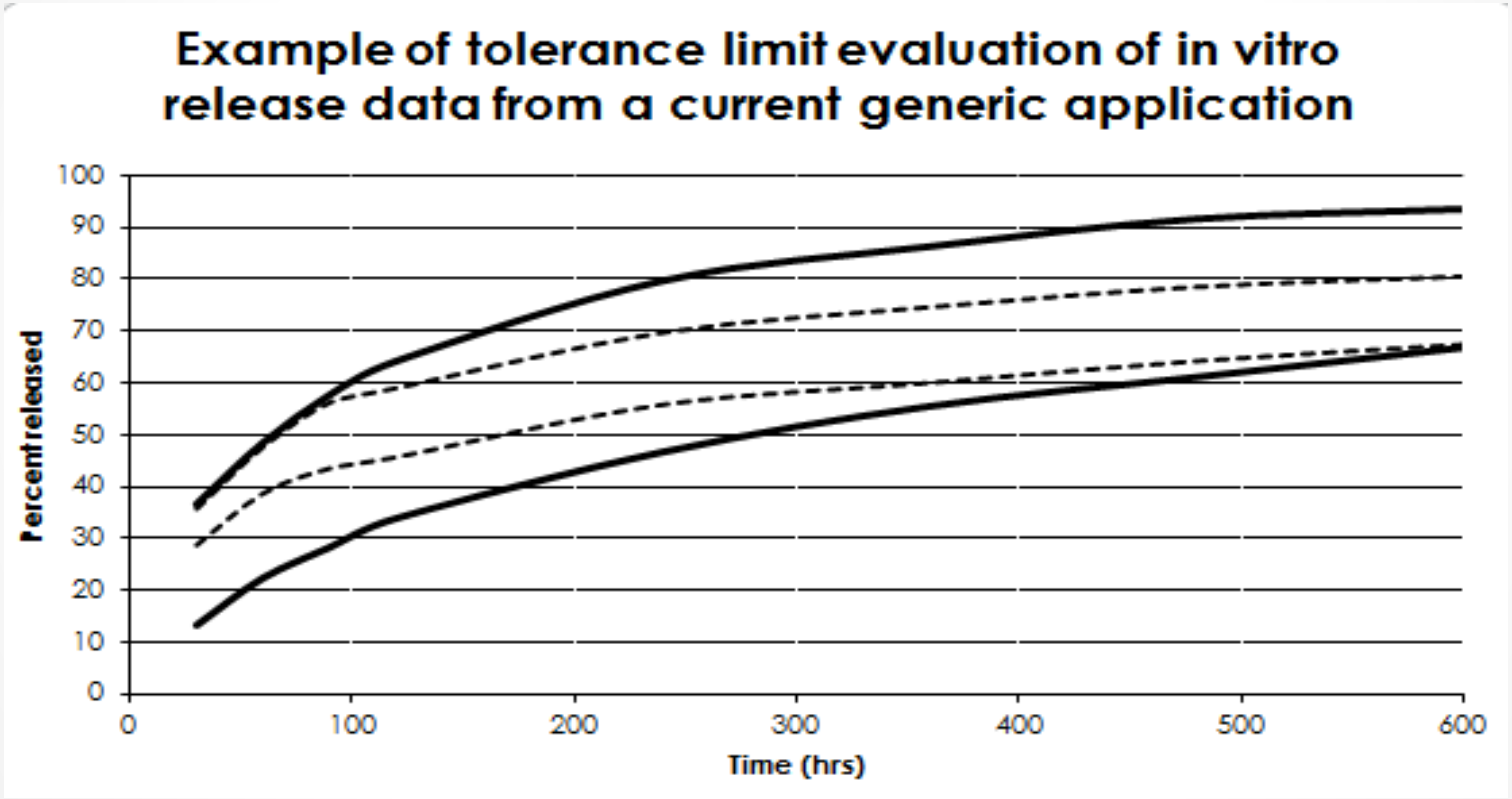


Confidence Interval: The difference in location of the mean due to this uncertainty is defined by the confidence interval.

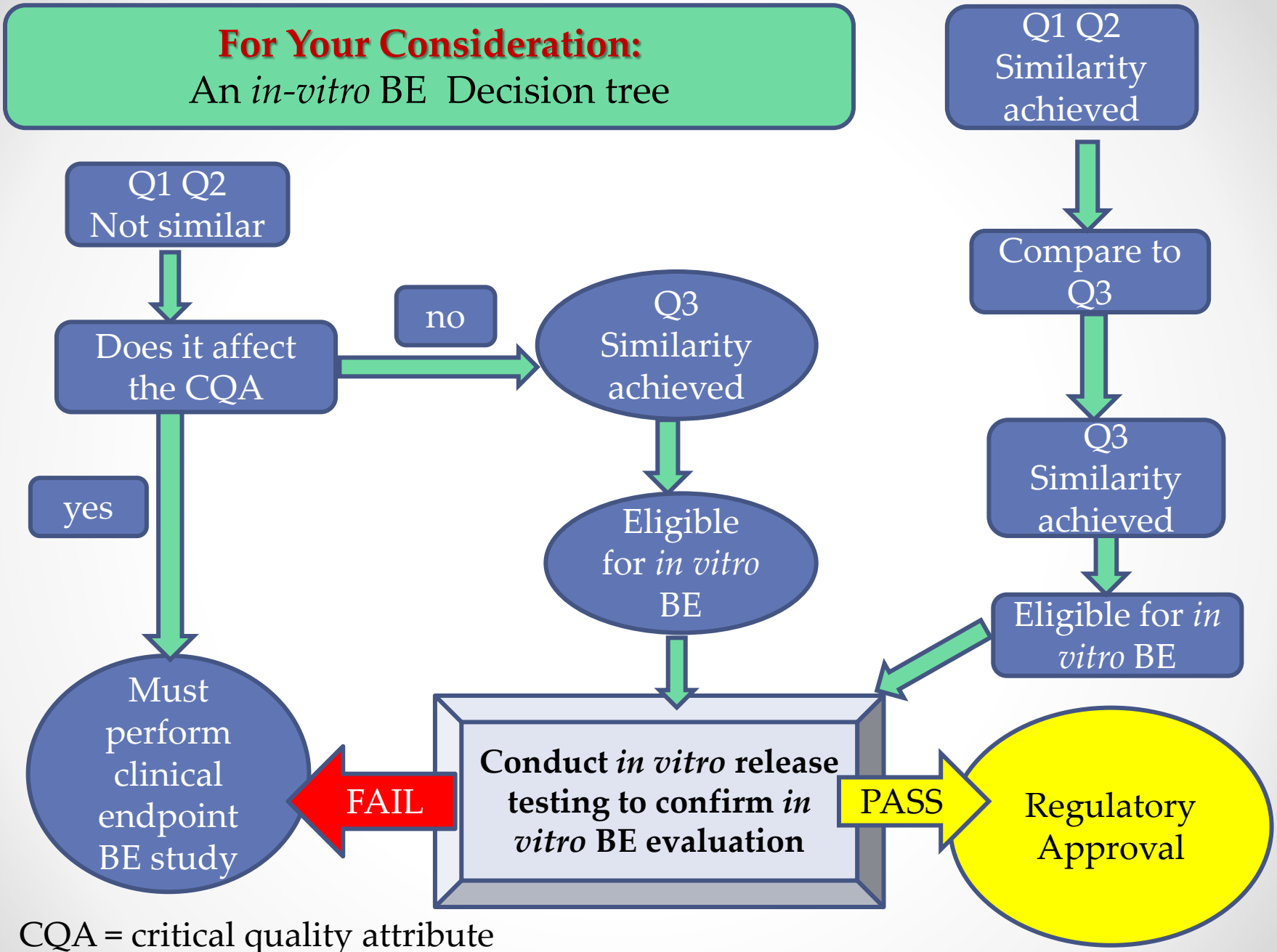
Tolerance interval: Accounts for both the uncertainty of knowing the true location of the mean of the population and the actual variability in the population

ProPharma Group Blog: http://www.propharmagroup.com/blog/understanding-statistical-intervals-part-iii-tolerance-intervals

Example of Tolerance Limit Approach



Solid lines = tolerance limits calculated for the reference product.
Hatched lines = tolerance limits calculated for the generic product


For Your Consideration:
An *in-vitro* BE Decision tree
Q1 Q2 Similarity achieved
Compare to Q3
Q3 Similarity achieved
Eligible for *in vitro* BE
Q1 Q2 Not similar
Does it affect the CQA
no
yes
Q3 Similarity achieved
Eligible for *in vitro* BE
Must perform clinical endpoint BE study
FAIL
Conduct *in vitro* release testing to confirm *in vitro* BE evaluation
PASS
Regulatory Approval
CQA = critical quality attribute

Next step: Physico-chemical characterization. Dr. Fahmy