About OMICS Group

OMICS Group is an amalgamation of Open Access Publications and worldwide international science conferences and events. Established in the year 2007 with the sole aim of making the information on Sciences and technology 'Open Access', OMICS Group publishes 500 online open access scholarly journals in all aspects of Science, Engineering, Management and Technology journals. OMICS Group has been instrumental in taking the knowledge on Science & technology to the doorsteps of ordinary men and women. Research Scholars, Students, Libraries, Educational Institutions, Research centers and the industry are main stakeholders that benefitted greatly from this knowledge dissemination. OMICS Group also organizes 500 International conferences annually across the globe, where knowledge transfer takes place through debates, round table discussions, poster presentations, workshops, symposia and exhibitions.

OMICS International

OMICS International is a pioneer and leading science event organizer, which publishes around 500 open access journals and conducts over 500 Medical, Clinical, Engineering, Life Sciences, Pharma scientific conferences all over the globe annually with the support of more than 1000 scientific associations and 30,000 editorial board members and 3.5 million followers to its credit.

OMICS Group has organized 500 conferences, workshops and national symposiums across the major cities including San Francisco, Las Vegas, San Antonio, Omaha, Orlando, Raleigh, Santa Clara, Chicago, Philadelphia, Baltimore, United Kingdom, Valencia, Dubai, Beijing, Hyderabad, Bengaluru and Mumbai.

6th World Congress on BA/BE

Bioequivalence of Topical Corticosteroids: Design and Data Analysis Challenges with the Vasoconstrictor Assay

Keith Gallicano, Ph.D.

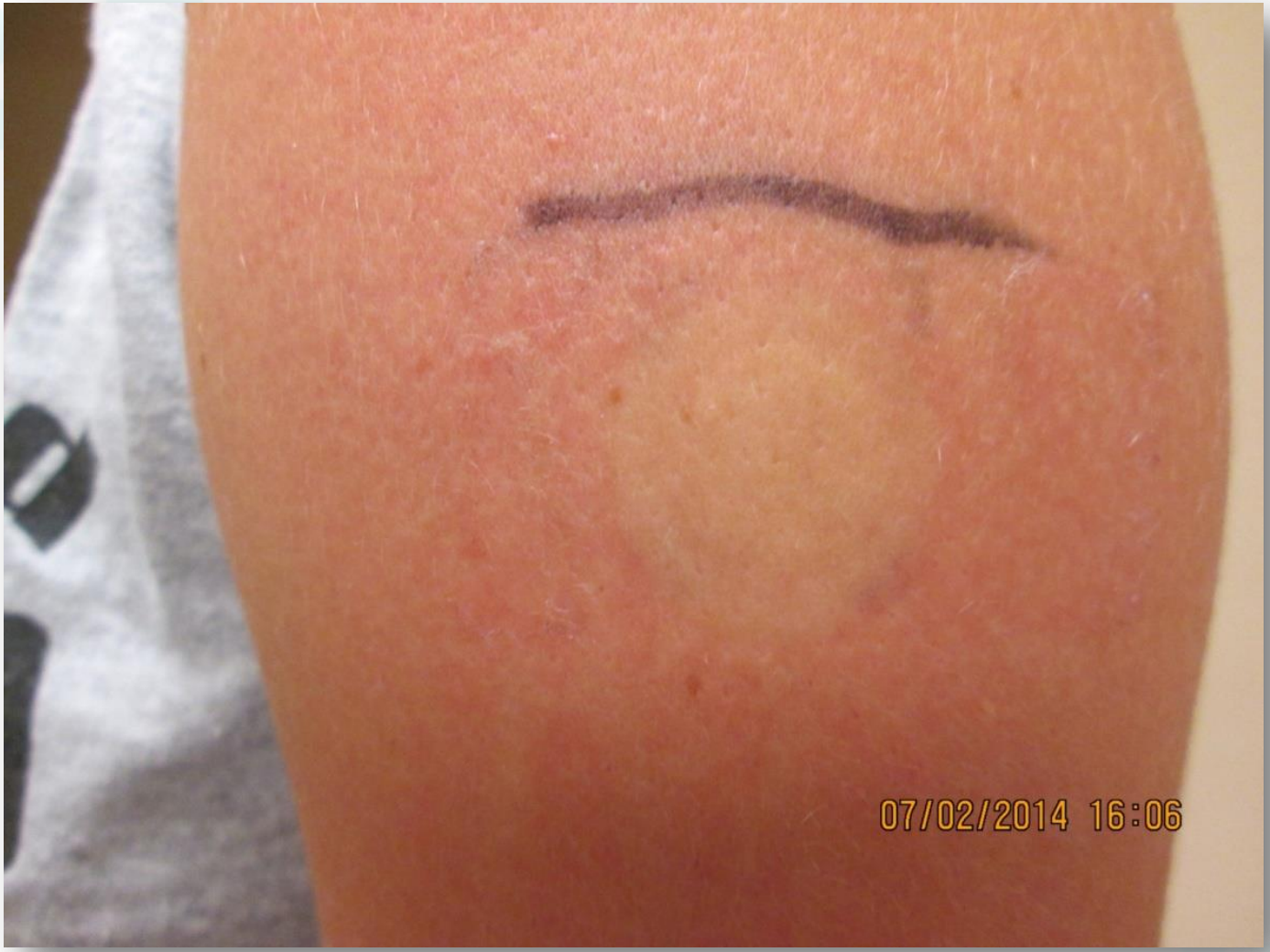
August 17, 2015

NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

Topics

- Operational Procedures and Challenges
- Types of Studies
- Data Analysis and Sample Size
- Design and Analysis Challenges and Issues
- BE Study Example
- Summary and Conclusions

Skin Blanching from Corticosteroids



Potency

- In 1985, Stoughton and Cornell classified corticosteroid potency according to their vasoconstrictive properties (vasoconstrictor assay)
- Determined by chemical structure, strength and formulation
- Higher potency more effective but higher rate of side effects; can be very significant in pediatrics

USA

Seven potency groups:

Super potent (*Class I*)

.

Least Potent (*Class VII = OTC*)

UK

Four potency groups:

Very Potent (*Class I*)

Potent (*Class II*),

Moderately potent (*Class III*),

Mild (*Class IV*)

Corticosteroid Activity

- Magnitude and duration of topical corticosteroid skin blanching in human skin is influenced by vehicle, treatment duration, and time of day of application
- Time of maximal decreased skin color generally occurs at midnight, independent of vehicle, treatment duration, or time of day of application
  - Coincides with lowest circulating cortisol concentrations
- In general, the higher the potency of the topical corticosteroid, the earlier the maximal effect is observed
  - This finding suggests that short application of highly potent agents might minimize systemic absorption without sacrificing efficacy

Pershing et al. J Invest Dermatol 1994;102:734-9

Potency by Formulation

- Typically, potency of gels and ointments are the greatest, followed by creams and lotions
- Lotions and solutions (sprays) cause a rapid and greater early response (a much quicker ED_{50} in the E_{max} dose duration-response curve)
- A shorter ED_{50} does not mean a more potent product
  - Faster absorption
- Occlusion (covering the site of application) significantly increases potency

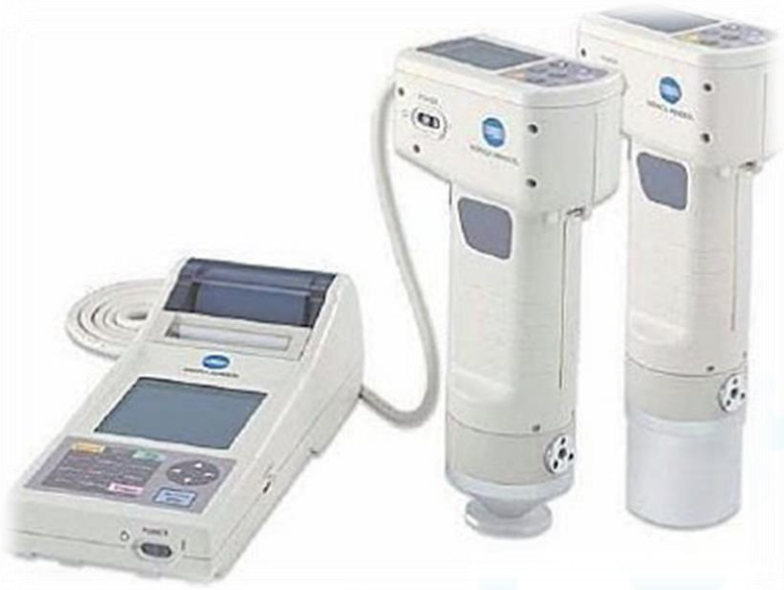
Use of Vasoconstrictor Response in Generic Evaluation

- When applied topically, corticosteroids are not systemically measurable (they are absorbed)
- The vasoconstrictor response is a “validated” pharmacodynamic measure of potency
- In 1992, OGD/FDA published an Interim Guidance on the use of the vasoconstrictor response to evaluate the bioequivalence of generic topical corticosteroids
- In 1995, OGD finalized the Guidance
- Only PD method currently “approved” by OGD for demonstrating BE
- Estimate ~ 100 generic topical steroid formulations approved by FDA using this method

Vasoconstrictor Assay Operational Procedures

- Primary objective is to measure the blanching response of the skin over time following a set dose
- Both the drug application, removal and the method of evaluation must be standardized and validated
- Important to try to reduce variability (noise) caused by the methodology

Chromameter is the equivalent of an LC/MS analyzer used in standard PK/BE studies

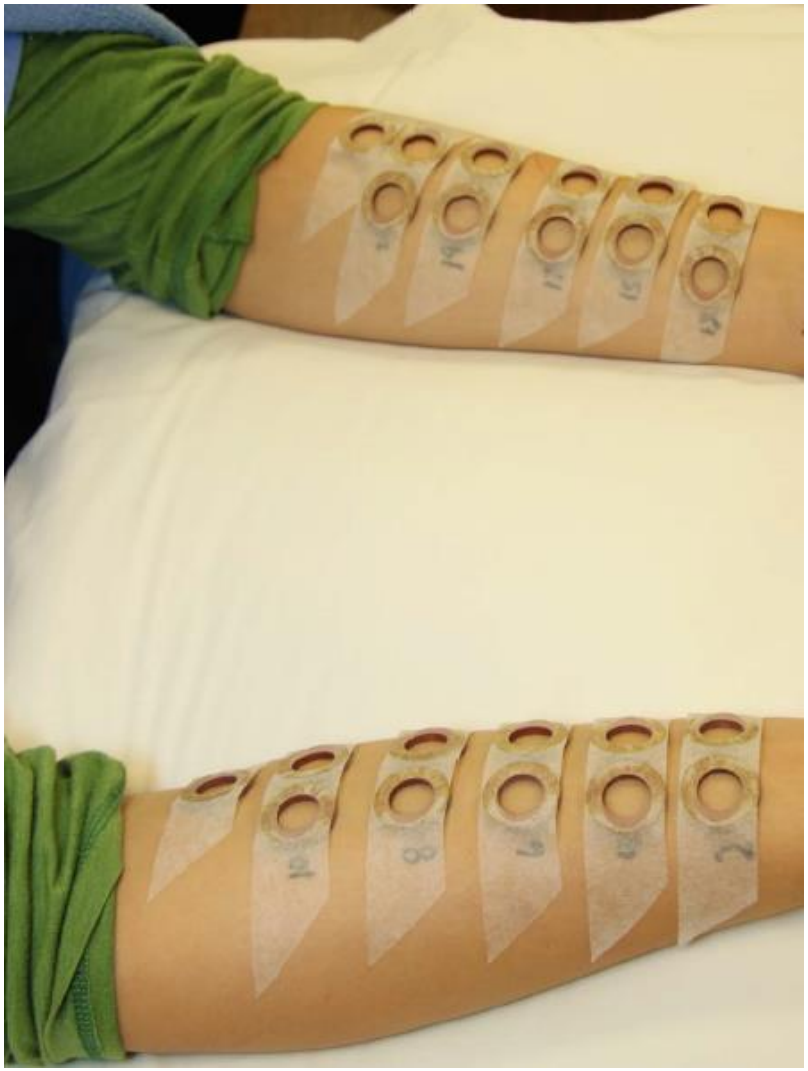


Vasoconstrictor Assay Operational Procedures

- Standardized application “dosing” is achieved using:
  - A standardized area of application
  - A standardized unit measure of dose
  - A standardized method of application and removal

- Inspect products to ensure they are homogenous
  - No separation of components

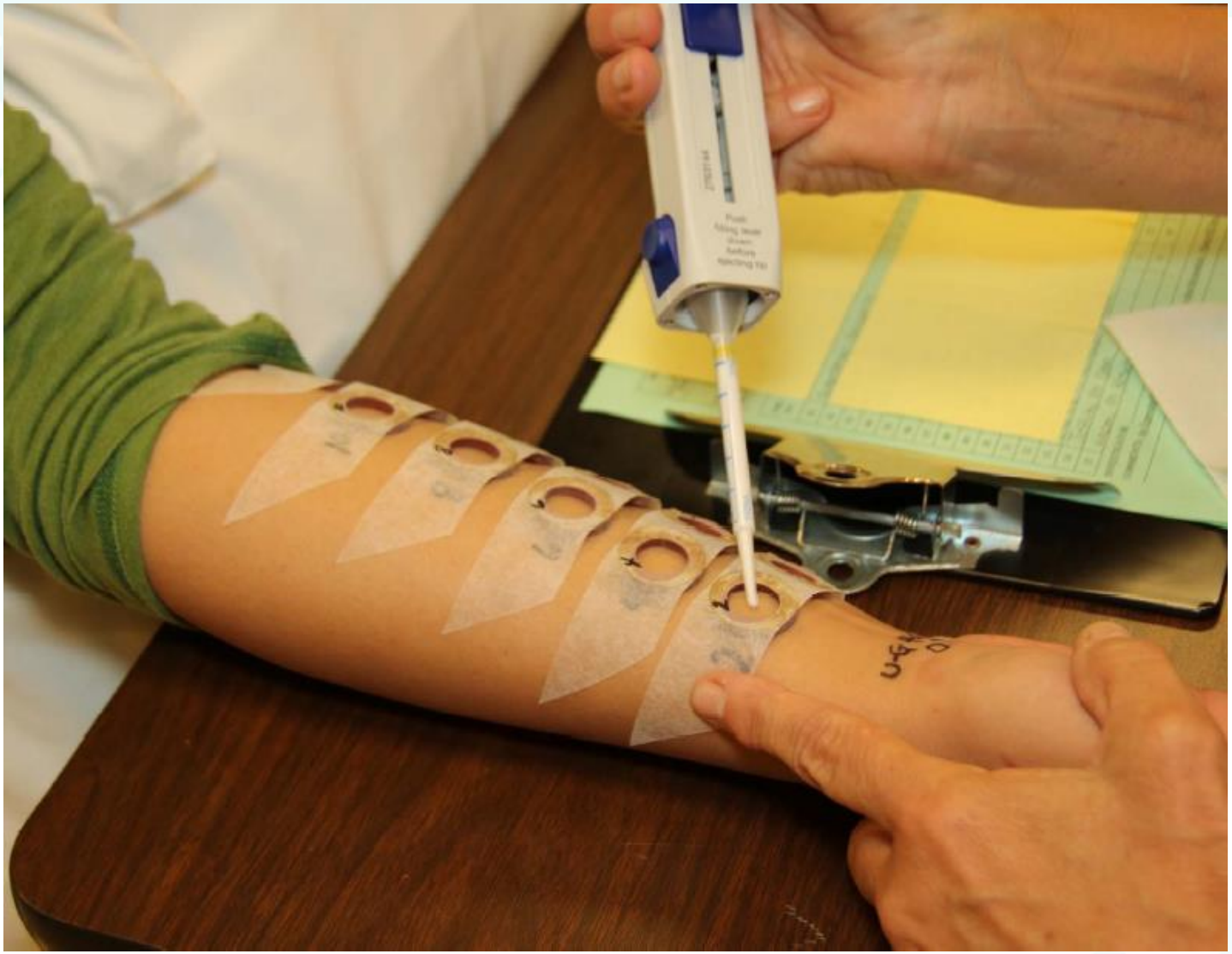
Standardized Area of Application



NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

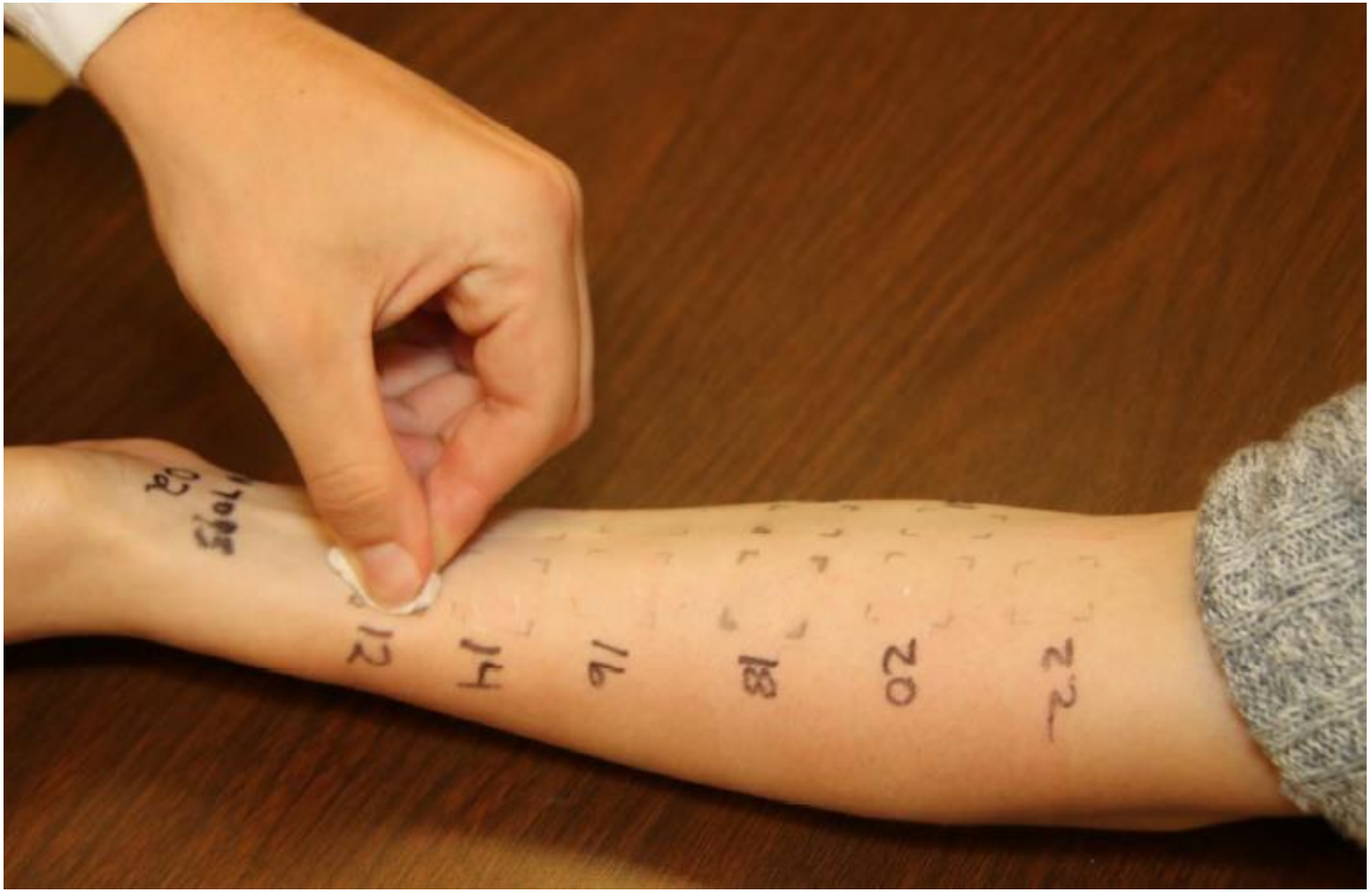
Standardized Unit Measure of Dose

Standardized Application



Standardized Application

Standardized Removal



Vasoconstrictor Assay Operational Procedures

- Standardized assessment/evaluation is achieved using:
  - An objective measuring device (Chromameter)
  - Pre-reading calibration / baseline correction
  - Validated operators
  - All Chromameters and operators must be cross validated for each study to ensure “within” and “between” reproducibility

Chromameter

Fitzpatrick Skin Types

SKIN TYPE	one	two	three	four	five	six
						
Hair	red, blonde	blonde, red, light brown	chestnut, dark blonde	brown, medium brown, dark brown	dark brown	black
Eyes	blue, grey, green	blue, grey, green, hazel	brown, blue, grey, green, hazel	hazel, brown	brown	brown
Skin	very pale white, pale white	pale white	white, light brown	medium brown, dark brown	dark brown	black
Tanning Ability	burns very easily, never tans	burns easily, rarely tans	sometimes burns, gradually tans	hardly ever burn, tans very easily	Rarely burns, tans easily and quickly darkens	Never burns, tans very dark

Basic Inclusion Criteria: Blanchers (Responders/Detectors)

- Only “subjects who have the capacity to vasoconstrict when dosed with the RLD”
- Inclusion of “nonresponders” would decrease the ability to detect differences between Test and RLD
- About 90% of the population will blanch
- 10-15% are super blanchers!
- Need to determine blanching status to every product (blanching depends on potency)
- Performed at screening on upper arm
- 2 hr duration or at estimated ED_{50} duration, whichever is longer
- Blanching (responder) status assessed visually 6-9 hr after removal

Four-Point Ordinal Visual Scale

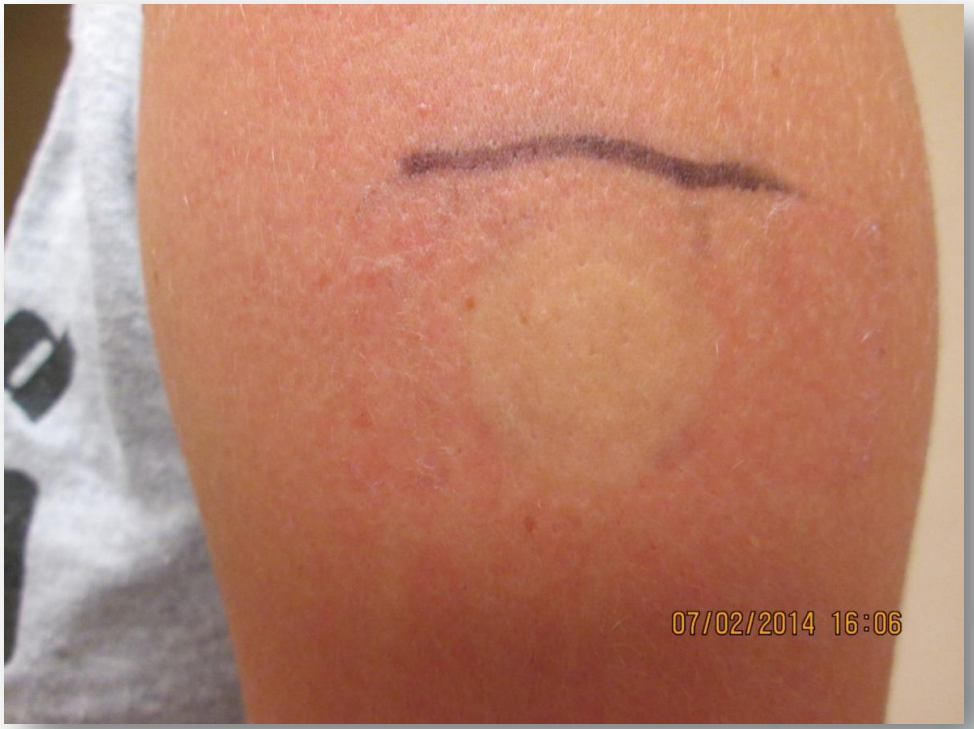
Assessments performed under standard fluorescent lighting and at room temperature.

0 = No pallor; no change from surrounding area.

1 = Mild pallor; slight or indistinct outline of application site.

2 = Moderate pallor; discernible outline of application site.

3 = Intense pallor; clean, distinct outline of application site.



During Study Procedures: Limit Variability

- Avoid extremes of temperature/humidity
- No caffeine/alcohol, etc. (flushing)
- No showers/washing arms at all during study
- Avoid activity
- No lying on arms during dosing or before readings
- Arms out straight for 5 minutes before readings
- Subjects must be under facility control throughout study

During Study Procedures: Challenges

- Summer months (suntans/availability)
- Too many sites/short arms (9-10 max)
- Very short duration periods (super potent products)
- Very long duration periods
- Drug “creeping” especially lotions

Types of Studies

- Dose duration-response study (ED_{50} or pilot study)
- Bioequivalence study
- Formulation screening study (more than one TEST lot)
- Potency-ranking study (NCEs or new formulations only)

FDA Guidance Quote

"…Because dose duration-response characteristics may vary with the particular drug of interest, as well as with study conditions, the Guidance encourages the performance of a pilot study to define appropriate parameters for the pivotal study…"

Guidance for Industry. Topical Dermatologic Corticosteroids: *In Vivo* Bioequivalence. U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER), 2 June 1995, Page 6.

Major Differences between ED_{50} and Bioequivalence Studies

ED_{50}

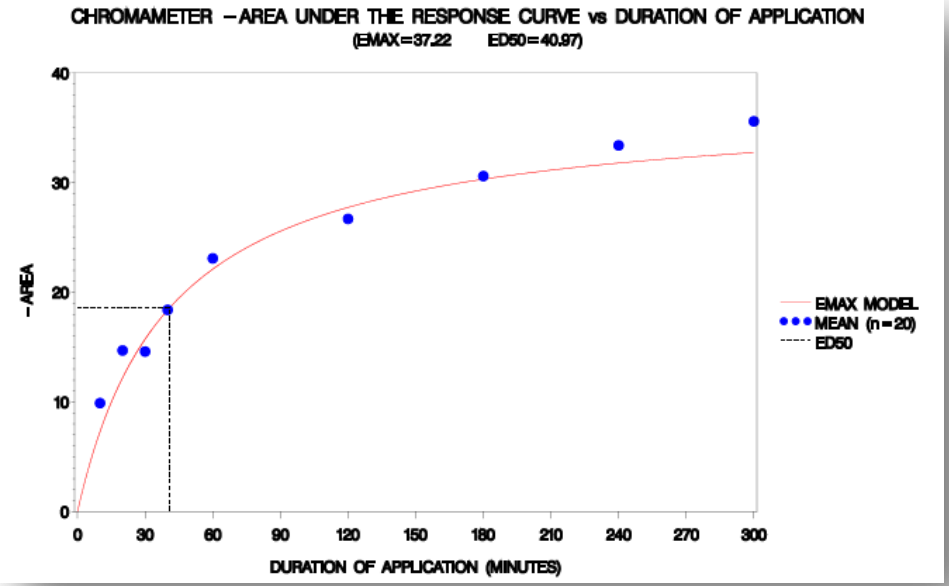
- Reference product only
- Apply for 7-8 durations
- Smaller subject #'s (20-24)
- Analysis on all subjects

Bioequivalence Study

- Reference and test
- Apply for 3 durations only
- Large number of subjects (100-200)
- Analysis on sub group of participants (qualifiers)

ED_{50} Study: Objective

- Objective is to calculate a duration of application (time) where 50% of maximal blanching occurs (i.e., the ED_{50})
  - This is the duration used to compare the test and reference product in the bioequivalence study because the responses are in the most sensitive (linear) part of the E_{max} dose duration-response curve to optimize discrimination of formulation differences



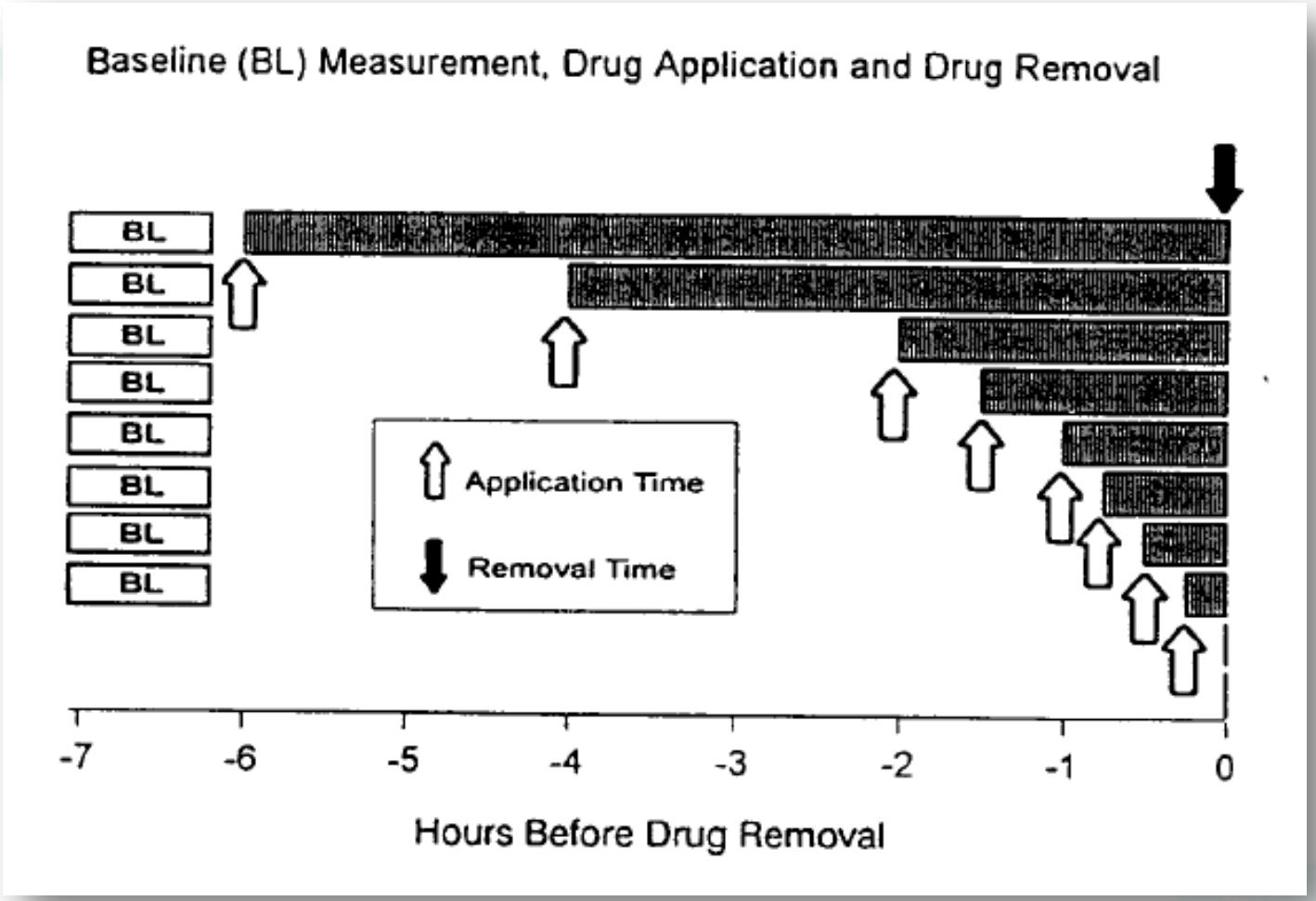
ED_{50} Study: Design

- Apply reference product for 7 to 8 different durations (range depends on potency of product)
- Class III/IV products: 5, 15, 30, 45, 60, 120, 180, 240 minutes
- Generally shorter for Super Potent; longer for Low Potency but ED_{50} time can easily be manipulated
- Two untreated sites per arm
- Evaluate area of blanching over 24 hours ($AUEC_{0.5\text{-}24hr}$) after removal for each duration (some exceptions)

ED_{50} Study: Application Schemes

- Two approaches for application of reference product
  - Staggered application with synchronized removal
  - Synchronized application with staggered removal

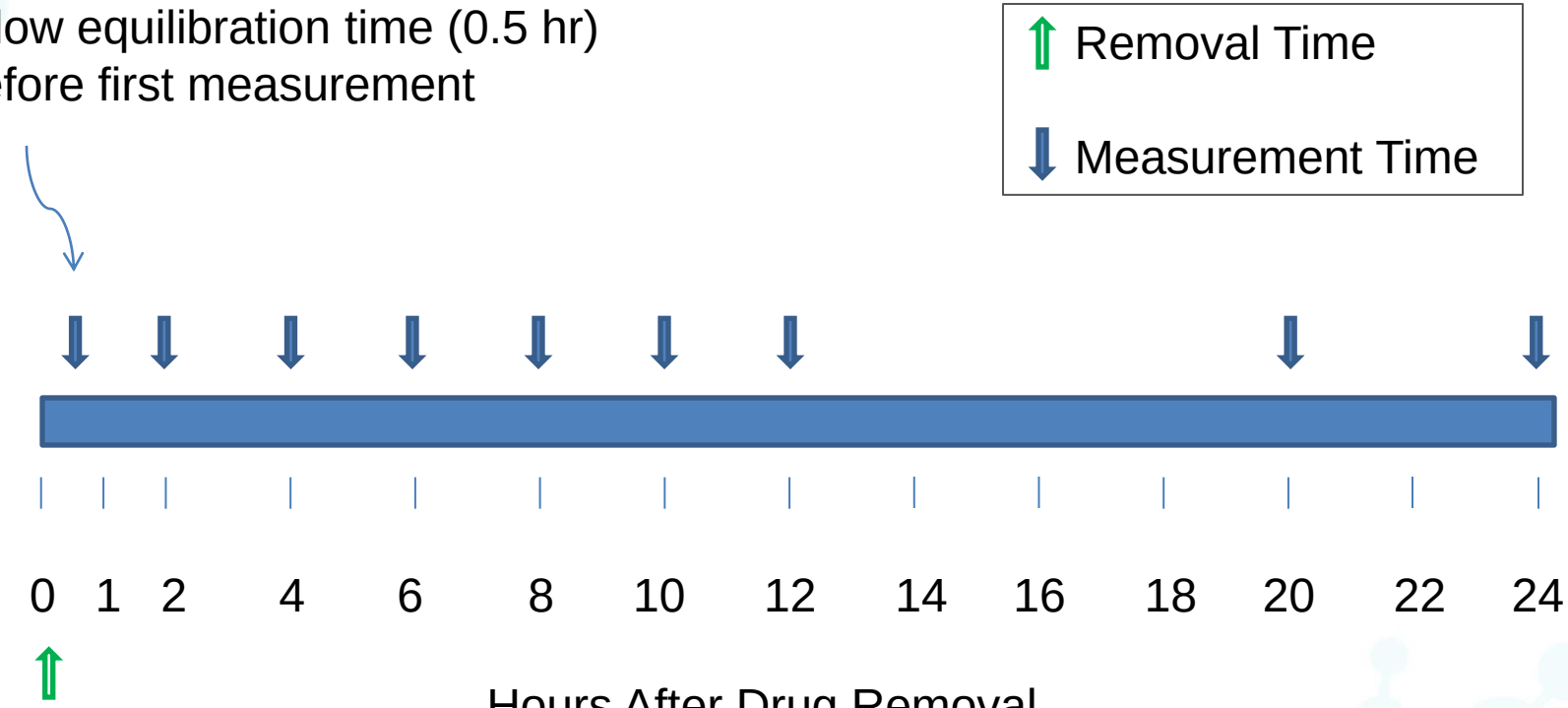
Staggered Application Synchronized Removal



Post-Removal Evaluation Scheme

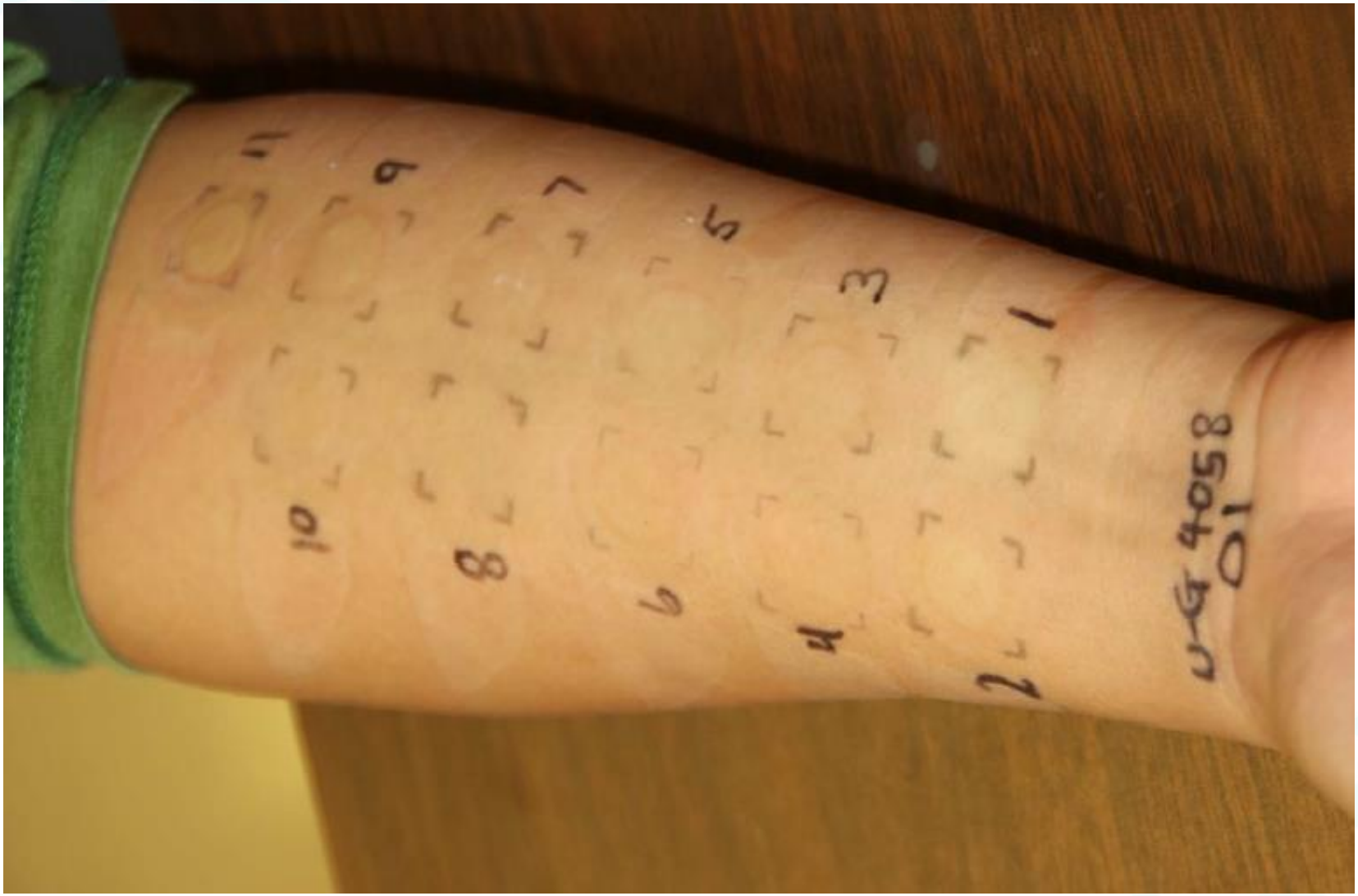
Skin Blanching Measurements

Allow equilibration time (0.5 hr) before first measurement

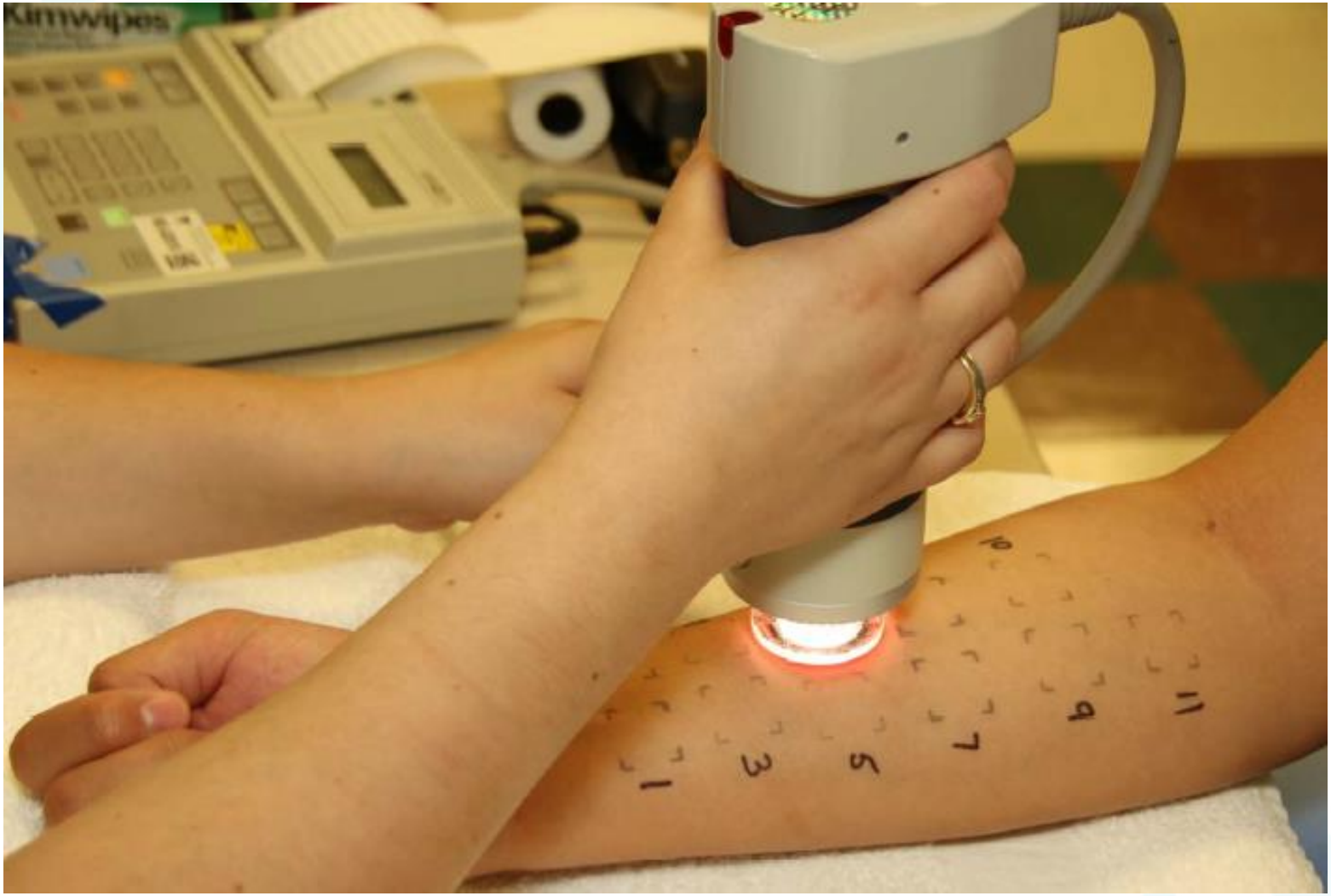


NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

Post-Removal Evaluation Sites



Chromameter Evaluation



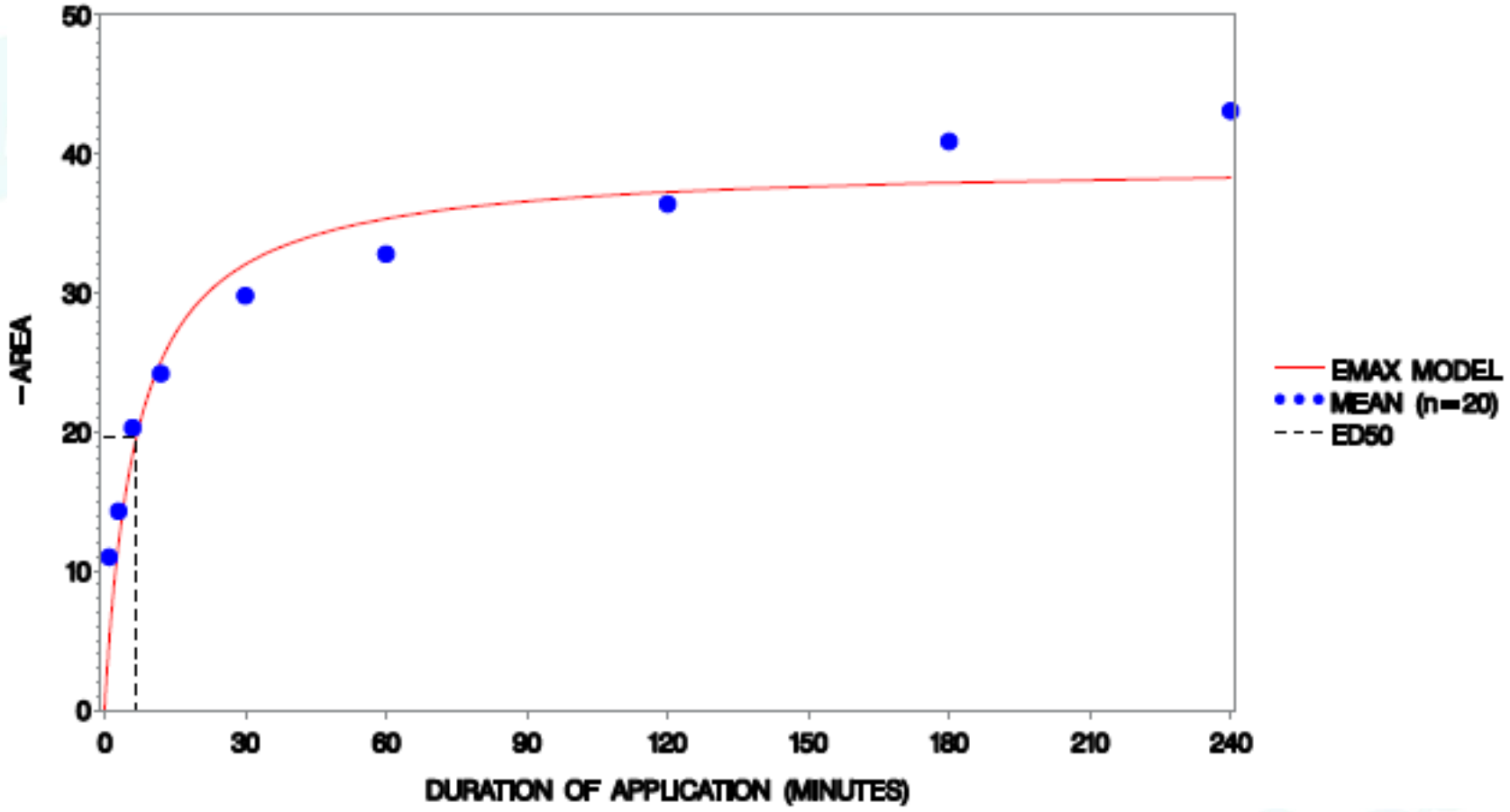
ED_{50} Study: Calculation of $AUEC_{0.5\text{-}24hr}$

- The post-dose Chromameter a-scale readings at each treated site are first corrected for the average of duplicate pre-dose (baseline) reading and then corrected for the average baseline-adjusted reading for the two untreated sites (on the same arm) at the corresponding post-dose reading time
- These "corrected" baseline-adjusted Chromameter values are used to calculate the area under the effect (response/time curve) ($AUEC_{0.5\text{-}24hr}$) for each site by the linear trapezoidal method
- Individual AUECs for each dose duration (single site on each ventral forearm (left/right)) are pooled across subjects
- A population model of the areas from all subjects is used to estimate the population dose duration-response (E_{max}) relationship

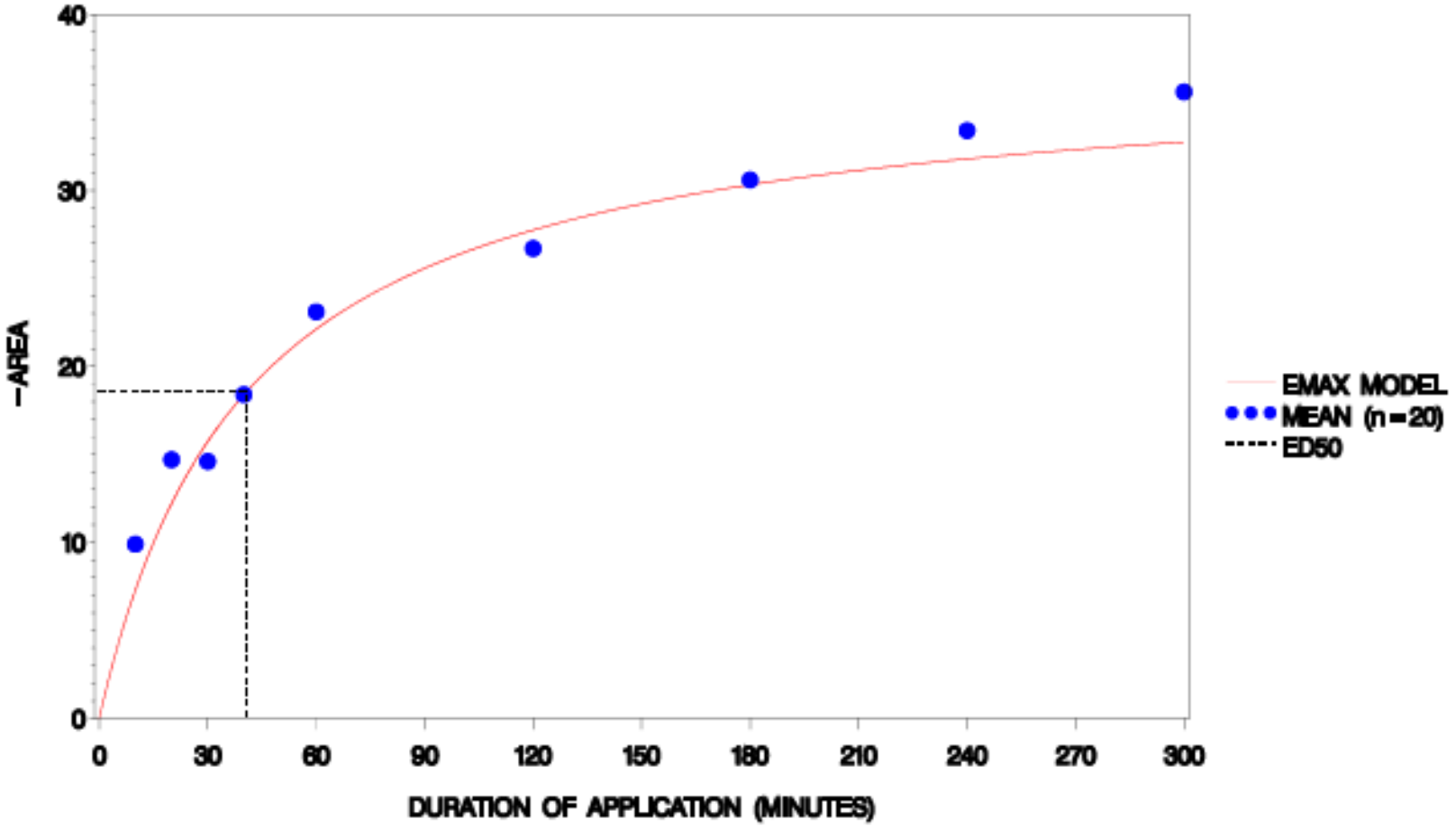
ED_{50} Study: Statistical Analysis

- Use population modeling approach to develop simple E_{max} model
- $E = (D*E_{max})/(ED_{50} + D) = AUEC_{0.5\text{-}24hr}$
  - E is the response (area) at D, the duration of application, and ED_{50} is the duration at which half-maximal response occurs
- PPharm software has been used by FDA (no longer available)
- E_{max} model is often not best fit
- Data becomes increasingly noisy (above baseline) as potency drops

ED_{50} Example: (Super Potent Cream)

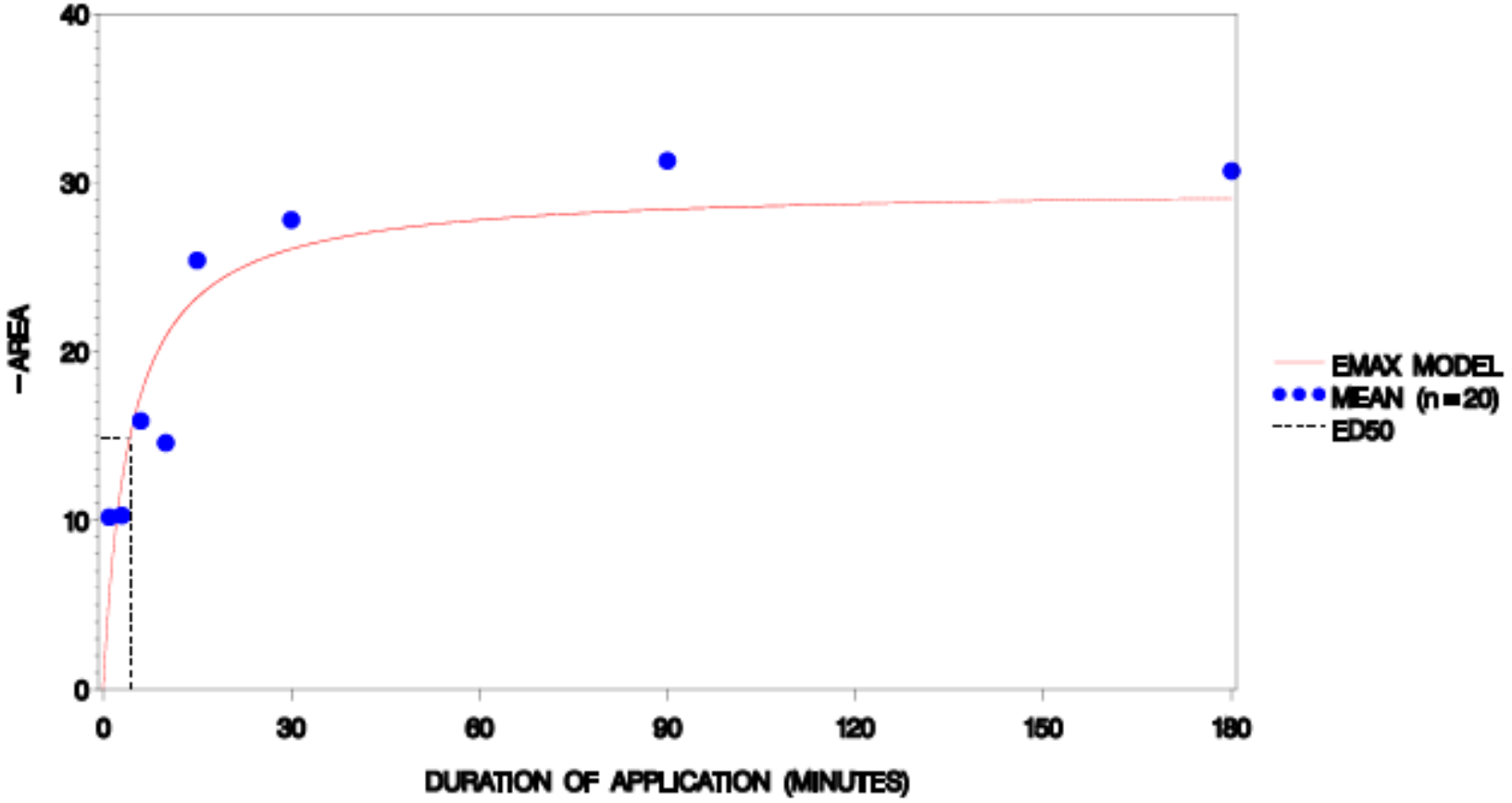


ED_{50} Example: Upper Mid-Strength Cream



NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

ED_{50} Example: Lower Mid-Strength Lotion



ED_{50} Study: Design Challenges

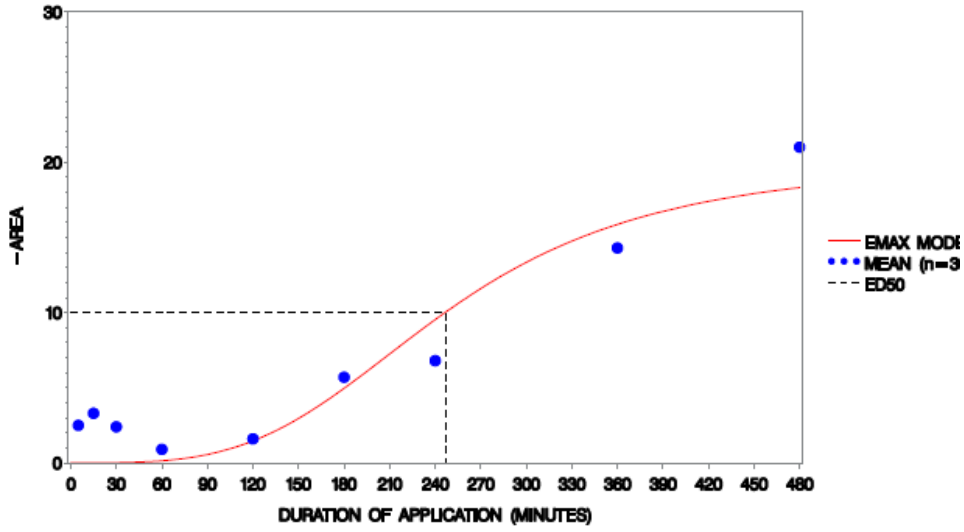
- "Published" potency can be misleading
- Very long or very low ED_{50} durations are unworkable
- Low potency products may require site occlusion
- Data can be very noisy
- Have to use all subjects, whereas pivotal studies only require analysis of the "detectors"
  - Know qualitative blanching response from individual screening data, but don't know quantitative blanching response (population)

ED_{50} Study: Analysis Challenges

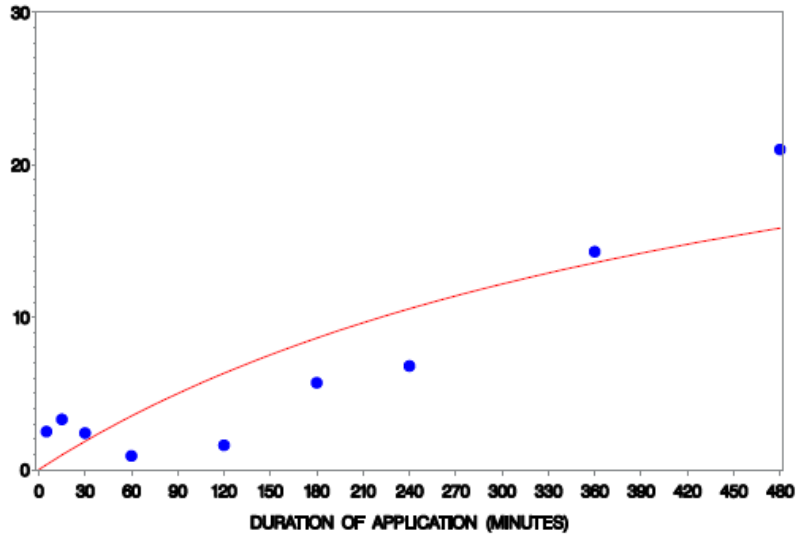
- Simple E_{max} model is not always best fit
- Some products show a sigmoidal E_{max} response
- Very short ED_{50} can drop below the first time of application (below the LLOQ!)
  - Short ED_{50} (≤ 1 min can be impractical for pivotal study)
  - Short ED_{50} can lead to high variability in pivotal study because of steep dose response
- Low E_{max}
  - Should be > 10
- Biphasic dose response
- Truncation of vasoconstrictor response at 24 hr post removal
- Data can be very noisy
- Reference lot variability

E_{max} Model Fit: Simple or Sigmoid?

Sigmoid

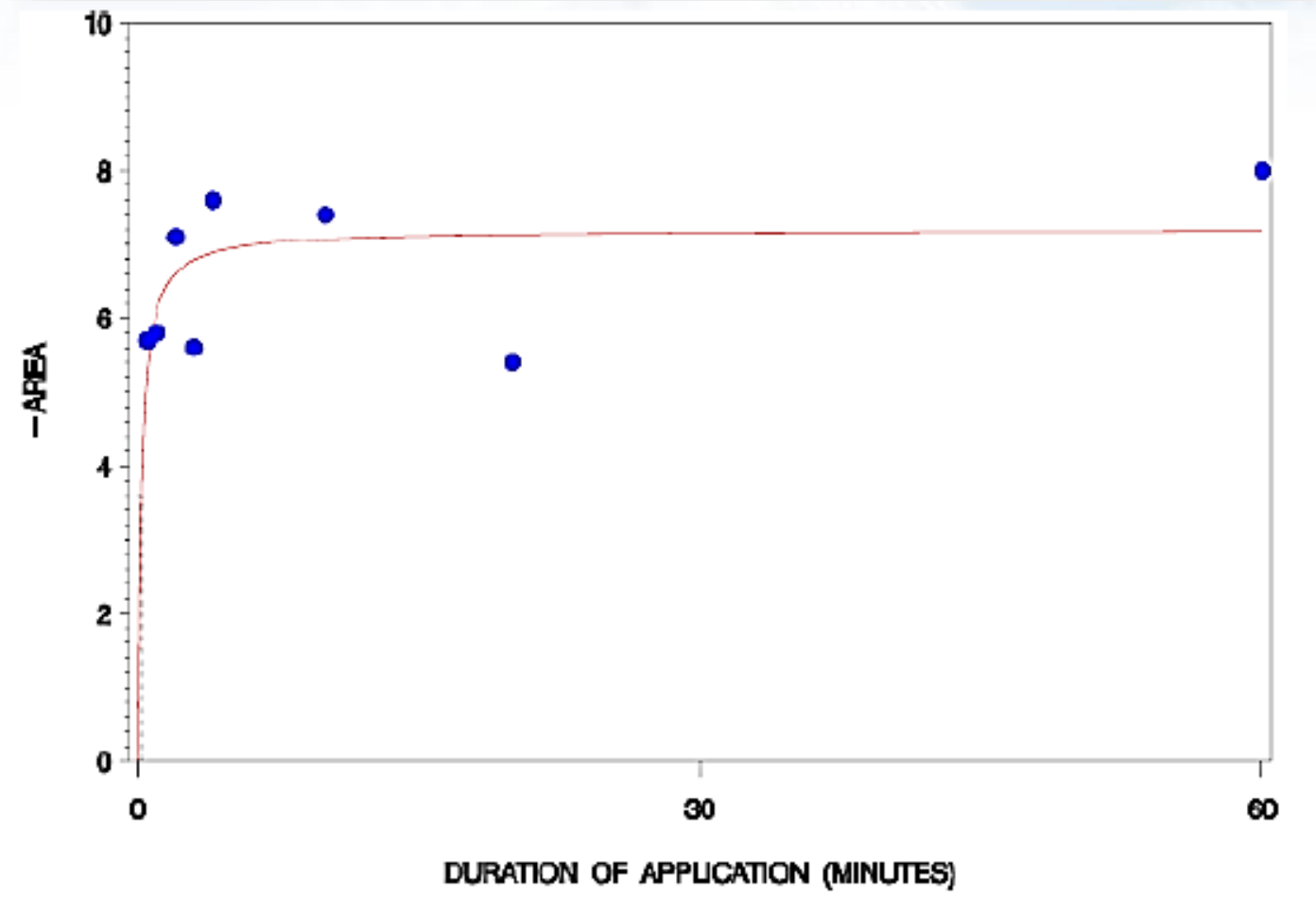


Simple



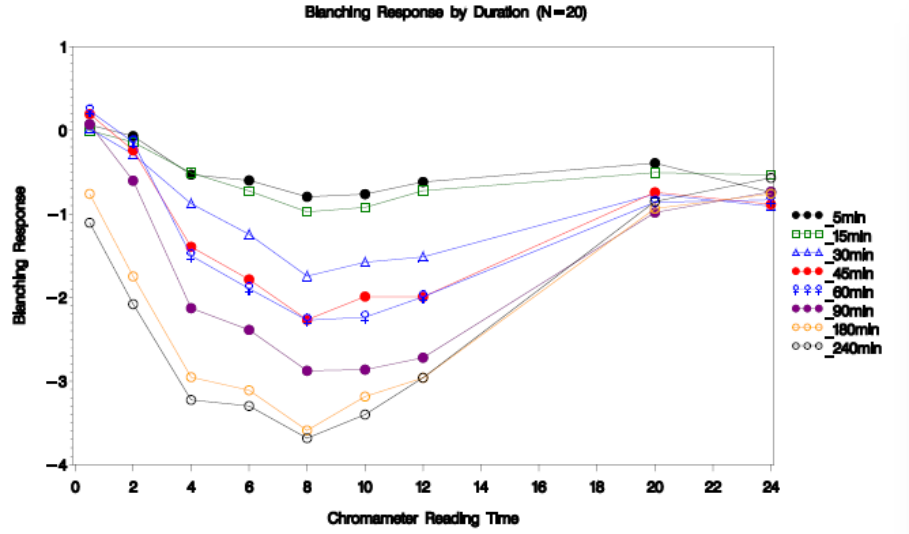
ED_{50} Analysis: Challenges

Low E_{max}

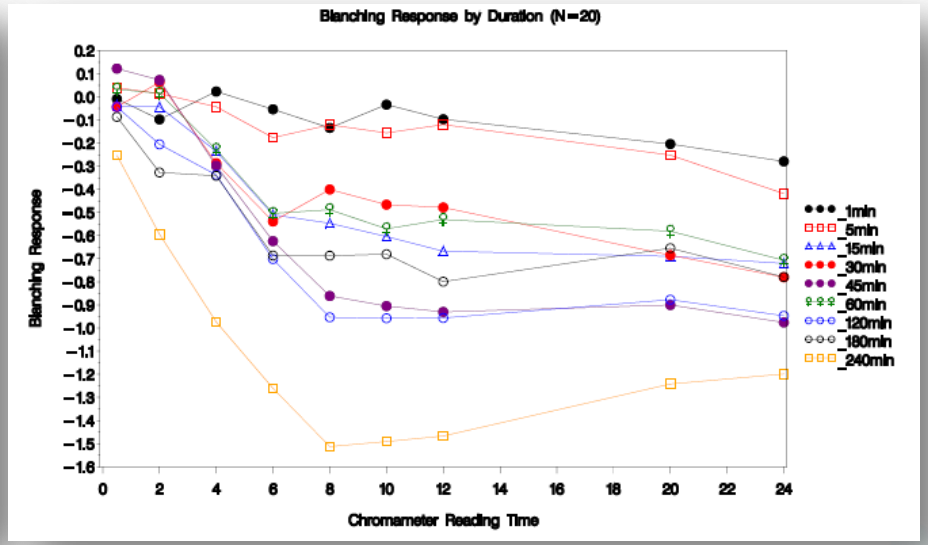


ED_{50} Analysis: Challenges

Truncation of vasoconstrictor response



Class 3 (Upper Mid-Strength) Cream

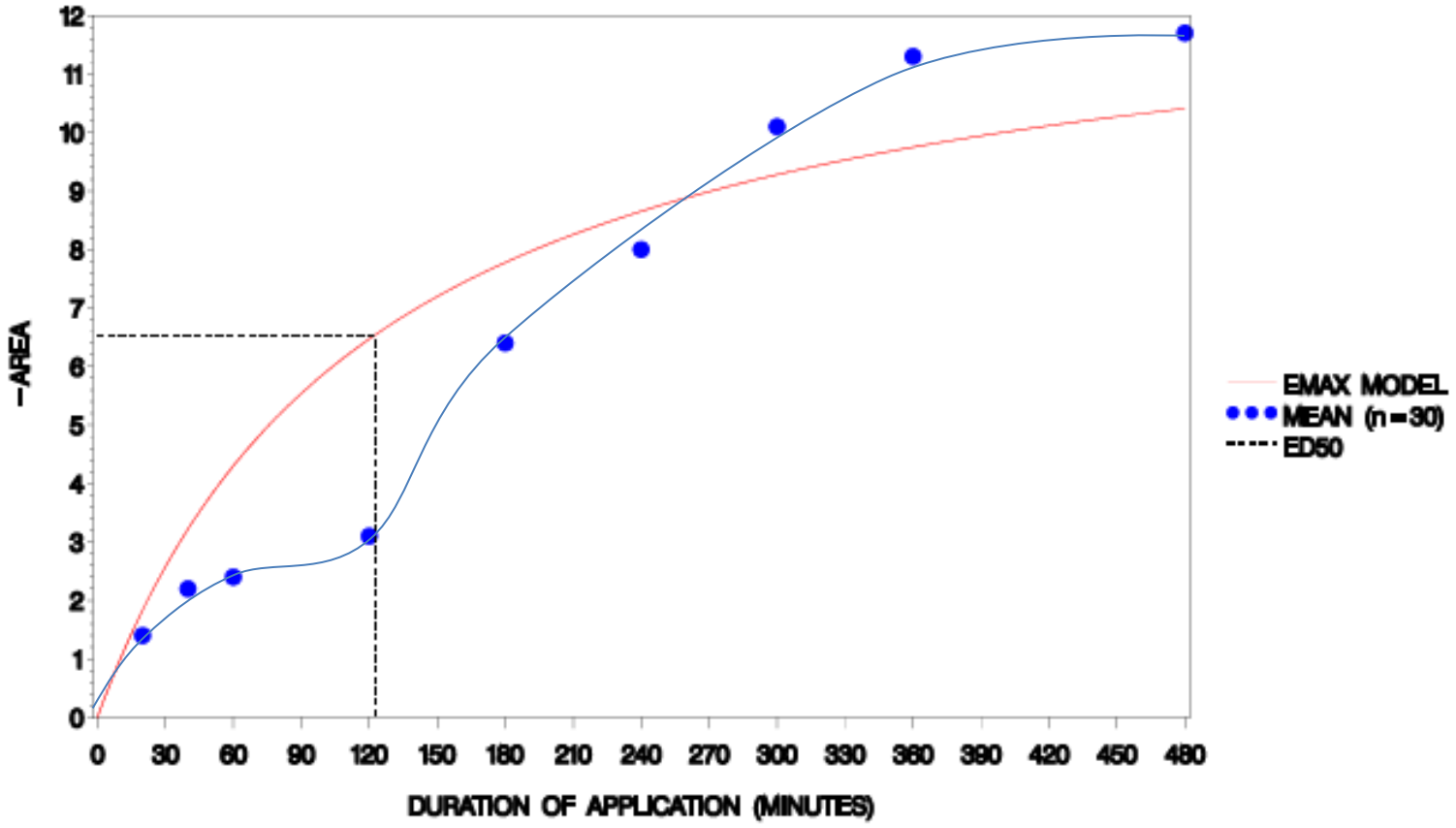


Class 6 (Mild) Gel

NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

ED_{50} Analysis: Challenges

Bi-phasic dose response for an ointment



Bioequivalence Study: Objective

- Objective is to demonstrate bioequivalence of the test to reference product
- Formulation screening study has similar objective with one or more test lots

Bioequivalence Study: Sample Size

- FDA Guidance says 40-60 qualifiers
  - What is the basis for this?
- # of qualifiers depends on CV% and T/R (may be < 40 or > 60)
- Estimate sample size using Fieller's (Locke's) method
  - N-Query software application for Equivalence: "Crossover design TOST for ratio of means (using original scale)"
- Need estimates of both within-subject and between-subject variability because Fieller's equation requires both between-subject variance and covariance estimates
- Fieller's method accounts for reference variability and treats reference mean as a variable and not as a constant when converting T- R to T/R
- Consider group sequential design with Pocock adjustment

Sample Size Estimations for 80% Power

T/R = 95%		
ISCV (%)	BSCV (%)	N
25	60	36
	100	42
30	60	48
	100	56
32	60	54
	100	60
35	60	64
	100	70
40	60	80
	100	88

Bioequivalence Study: Design

- Apply reference product for 3 different durations (D_1, ED_{50} and D_2)
  - $D_1 = ½ \times ED_{50} = ⅓ \times E_{max}$
  - $ED_{50} = ½ \times E_{max}$
  - $D_2 = 2 \times ED_{50} = ⅔ \times E_{max}$
- Apply test product for ED_{50} duration only
- Evaluate area of blanching over 24 hours ($AUEC_{0.5\text{-}24hr}$) after removal for each duration
- Use D_2/D_1 area ratio to qualify subjects for testing bioequivalence of test to reference

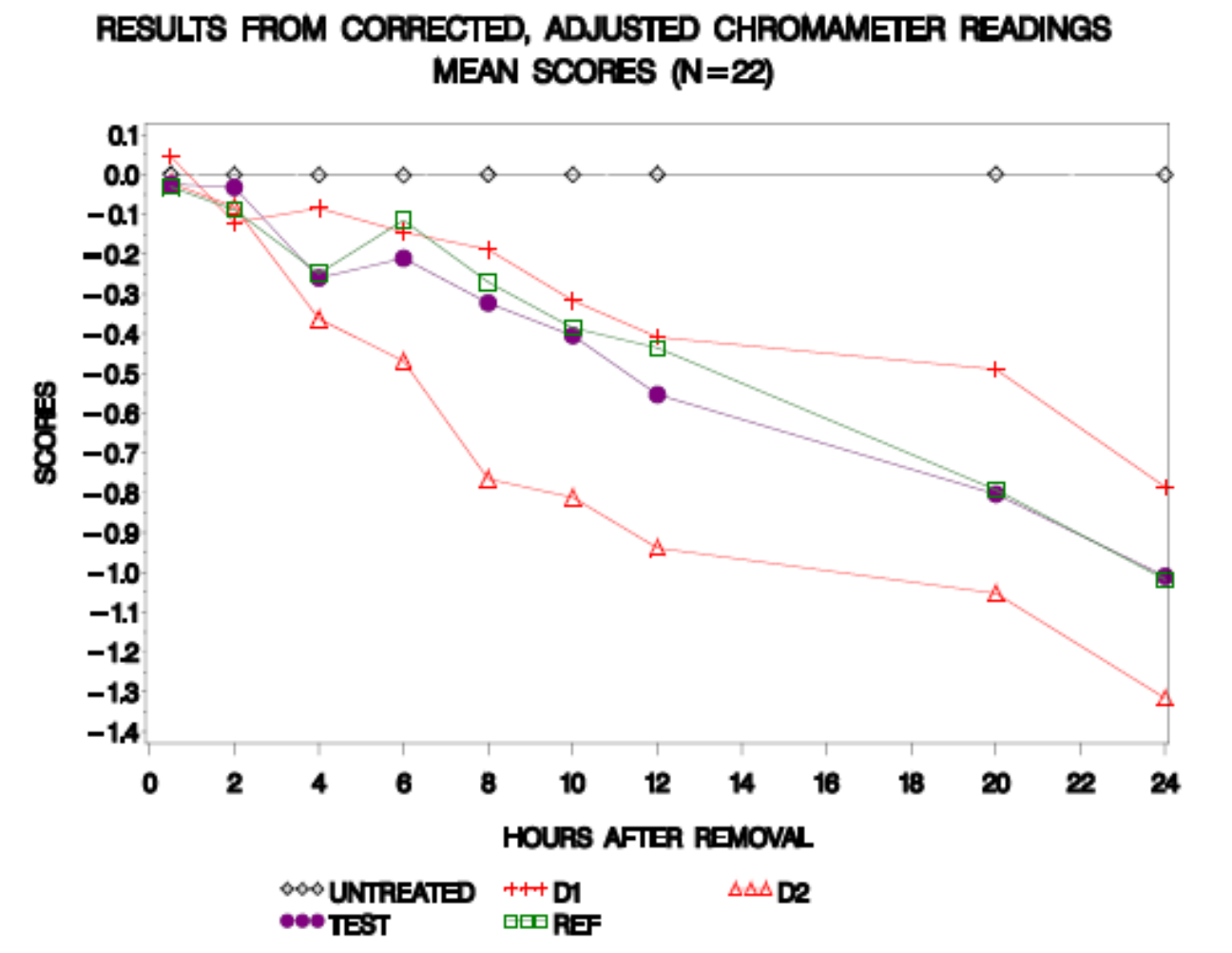
BE study: Calculation of $AUEC_{0.5\text{-}24hr}$

- The post-dose Chromameter a-scale readings at each treated site are first corrected for the average of duplicate pre-dose (baseline) reading and then corrected for the average baseline-adjusted reading for the untreated sites (on the same arm) at the corresponding post-dose reading time
- These "corrected" baseline-adjusted Chromameter values are used to calculate the area under the effect (response/time curve) ($AUEC_{0.5\text{-}24hr}$) for each site by the linear trapezoidal method
- Individual AUECs for each treatment (triplicate sites for each ED_{50} test and reference and single site for each D_1 and D_2 on each ventral forearm (left/right)) are pooled and averaged
- The mean AUEC value (n = 6 per ED_{50} treatment and n = 2 for each D_1 and D_2) is used as the response variable for each subject

Bioequivalence Study: Statistical Analysis

- Only subjects whose D_2/D_1 area ratio ≥ 1.25 included in analysis
- These subjects are considered detectors or qualifiers
- Compare Test $AUEC_{0.5\text{-}24hr}$ with RLD $AUEC_{0.5\text{-}24hr}$
- Locke's 90% CI (80-125% criterion)
  - Pocock adjustment if use two-group sequential design (94.12% CI at each stage)
- Ideally want to see Test and Reference AUEC values between those of D_1 and D_2

BE Example: Mean Blanching Profiles



BE Example: Interim Results

Number Enrolled	Number and % Qualified (N)	D_1 Mean $AUEC_{0.5\text{-}24}$	D_2 Mean $AUEC_{0.5\text{-}24}$	Test Mean $AUEC_{0.5\text{-}24}$	Ref. Mean $AUEC_{0.5\text{-}24}$	Test-to-Reference Ratio (%)	90% Conf. Interval* Lower (%)	Upper (%)	Intra-subject CV relative to LSMean(ref) (%)	Total N needed for 80% Power with T/R Ratio of: 100% *	95-106.3%*
58	22 (38%)	8.20	18.62	12.08	11.19	107.94	88.28	131.93	37.8	62	80

*Use observed BSCV and ISCV relative to reference of 102% and 37.8%, respectively

Study fails at interim analysis (~ 50% of protocol-specified 40 qualifiers)

Advised client to continue study because of acceptable T/R and study was predicted to pass assuming same T/R , BSCV and ISCV, and 40 qualifiers, despite predicted low study power of 43%

BE Example: Final Results

Number Enrolled	Number and % Qualified (N)	D_1 Mean $AUEC_{0.5\text{-}24}$	D_2 Mean $AUEC_{0.5\text{-}24}$	Test Mean $AUEC_{0.5\text{-}24}$	Ref. Mean $AUEC_{0.5\text{-}24}$	Test-to-Reference Ratio (%)	90% Conf. Interval* Lower (%)	Upper (%)	Intra-subject CV relative to LSMean(ref) (%)	Total N needed for 80% Power with T/R Ratio of: 100% *	95-106.3%*
118	45 (38%)	8.93	20.28	11.57	11.26	102.83	89.10	119.72	41.0	70	90

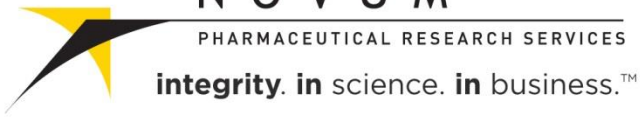
*Use observed BSCV and ISCV relative to reference of 95.8% and 41.0%, respectively

Study passes with 45 qualifiers (post-hoc power ~ 52%)

Summary and Conclusions

- Most important to control within-subject variability (ISCV) via consistent Chromameter technique
- Even with good Chromameter technique , some products' data do not fit well to a simple E_{max} model or they have inherent high ISCV that requires sample sizes > 60 (upper limit in FDA Guidance)
- Works best with medium to high potency formulations and simpler formulations such as gels and ointments
- Problematic for lower potency products and newer formulations (e.g., sprays, tapes, foam)
- Works best with fair-skinned subjects who show good blanching response
- Current alternative method to a pharmacodynamic study is not attractive (clinical trial, microdialysis)
- Need to make best use of current requirements

Thank You


NOVUM
PHARMACEUTICAL RESEARCH SERVICES
integrity. in science. in business.™

Let us meet again..

We welcome you all to our future conferences of

OMICS International

7th World Congress on

Bioavailability & Bioequivalence: BA/BE Studies Summit

On

August 29 - 31, 2016 at **Atlanta, USA**

http://bioavailability-bioequivalence.pharmaceuticalconferences.com/