



## Qualification of Potency Standards

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**biogen idec**

# Talk Overview

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- Phase-based approach for potency standard qualification
  - Early stage
  - Late stage
  - Commercial
- Case study
- Preventing drift – to be covered by Noel Rieder, 9/22 session

# Considerations for Potency Standards (PS)

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## The Standard used in the Potency Assay

- May **BE** the in house reference standard
- May **be derived from** the in house reference standard
  - **Potency Working Standard (WS)**
    - Dilution of a reference standard to a fixed concentration
- May **BE** an International Reference Standard (IS)
  - Limited quantities
  - Used during early development
  - Used during qualification exercises
  - Used for once-a-year potency verification

# Qualification of Reference Standards for Potency Testing

- **Phased-based approach**



- Additional qualification of the Reference Standard is performed for use as an assay standard in potency testing
- Assures that activity measurements are consistent throughout development and stability assessments
- The qualification process evolves as molecule development progresses
  - Increased product knowledge
  - Increased analytical capability

**BASIC** → **ELABORATE & CONTROLLED**

# Research Reference Standard (RRS)

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- Initial characterization by early research assays
- Potency confirmed by animal models and assigned as 100%
  - No pre-established acceptance criteria
- Stored at -70C
- Used for cell line selection, process and formulation development, & method development
- Limited quantities

# Qualification of Interim Reference Standard

(Interim = until we can make a GMP reference standard)

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- Prototype DS is used for Interim Reference Standard (IRS)
  - Post production cell line selection and process development lockdown
  - Typically IND-enabling Tox material
  - IRS qualified using future release (qualified) and some characterization assays

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## Further Qualification of IRS for Potency Testing

- *In vitro* bioactivity assay is used
  - ✓ **Using a RRS**
    - ❑ One successful run; one analyst
    - ❑ Potency assigned as 100% if activity is w/in 75-133% of RRS
      - 75-133% is typical early bioactivity release limit
    - ❑ Differences will be investigated

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### ✓ Using an IS

- Multiple runs; 2-3 analysts
- More than one day
- Average of all runs assigned as potency value
- 95% CI within 80 -125% or predefined number of runs (~20)
- Normalized to the activity of the IS

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- **IRS is stored at -70C and placed on stability**
- **Used to release Phase I clinical material**

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- Reference Standard prepared from GMP DS batch 1 or 2
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## Further Qualification of GMP RS for Potency Testing

### ✓ **Using an in house IRS**

- Potency compared against IRS assigned as 100% if activity is within 80 – 125%
  - ❑ **One successful run; 1 analyst**
  - ❑ **95% CI around the mean must be w/in 80 – 125%**

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## Further Qualification of GMP RS for Potency Testing

### ✓ **Using an IS**

- Potency compared against IS, units to be assigned
  - ❑ **Multiple successful runs; 2 analysts; multiple days**
  - ❑ **95% CI around the mean must be w/in 80 – 125%**
  - ❑ **Mixed model statistical evaluation used to determine 95% CI**
  - ❑ **If meets; stop here – use mean RP of all runs**
    - $\%RP \times IS \text{ Potency (IU/mL)} = \text{Potency of candidate RS (IU/mL)}$
  - ❑ **If does not meet; add more runs until criteria is achieved**

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## Further Qualification of GMP RS for Potency Testing

### ✓ **Using an International Standard**

- Potency compared against IS, units to be assigned
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    - $\%RP \times IS \text{ Potency (IU/mL)} = \text{Potency of candidate RS (IU/mL)}$
  - ❑ **If does not meet; add more runs until criteria is achieved**
- **Confirmation run performed using new RS as assay standard and IS as a sample**

# Late Stage & Commercial Reference Standards

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- Reference Std prepared from commercial process at scale
- Qualified using lot release assays & expanded characterization to assess CQAs
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## Further Qualification for Potency Testing

- A Potency Working Standard (WS) is prepared from the RS and used to release commercial batches & monitor stability
- Potency compared against previous GMP RS or IS
- Replication far more than previous
- Last RS before commercial (where there is no IS)
  - Arbitrary units may be assigned to establish a “gold” standard
  - Assures future RS consistency

# Case Study: Calibration of Potency WS

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- Potency WS prepared by diluting RS into formulation buffer
  - Qualification follows a pre-approved protocol
    - The amount of testing is based on assay variance
    - Candidate working standard is tested until the half width of the 95% CI meets predetermined accuracy acceptance criteria.
- Assay controls used to demonstrate appropriate assay performance and to capture a potential assay shift
- Potency WS qualification protocol describes procedure for requalification and expiry extension
  - Potency of WS must stay within 95 – 105% of original assigned value

# Case Study: Calibration of Potency WS

Testing Site	Analyst	Vial #	Potency (IU/mL)	Log <sub>e</sub> Potency
A	1	1	9.43	2.244
		2	9.56	2.258
		3	9.75	2.277
	2	4	10.07	2.310
		5	10.05	2.308
		6	10.23	2.325
	3	7	9.90	2.293
		8	9.88	2.291
		9	9.92	2.295
B	4	10	10.57	2.358
		11	10.58	2.359
		12	10.58	2.359
	5	13	10.71	2.371
		14	10.44	2.345
		15	10.18	2.320
	6	16	10.13	2.315
		17	10.25	2.327
		18	10.15	2.318
Grand Mean Log <sub>e</sub> Potency				2.315
Standard Deviation				0.036
Standard Error				0.008
t-distribution t-value				2.110
Halfwidth of the 95% CI				0.018
Count (n)				18
<b>Assigned Potency Value (IU/mL)</b>				<b>10.1</b>

- Candidate WS was tested at 2 sites by 6 different analysts; several days
- Relative potency was determined vs IS
- $\%RP \times IS \text{ Potency (IU/mL)} = \text{Potency candidate WS (IU/mL)}$
- Testing is stopped once half width of 95%CI <0.03
- Log-transformed Mean Potency is Assigned to Candidate WS

# Control Performance During Potency WS Qualification

<u>Control Values During new WS Qualification</u>	
Analyst	Control Potency (IU/mL)
1	69.13
	70.98
2	63.79
	66.30
3	66.19
	61.29
4	66.79
	68.81
5	63.17
	64.46
6	61.98
	60.59
<b>Mean</b>	<b>65.29</b>
Std Dev	3.30
N	12.00

<u>Historical Control Values</u>		
	Site 1	Site 2
<b>Mean</b>	<b>65.9</b>	<b>65.6</b>
Std	3.94	2.96
N	212	118

- Control – different DS batch
- Potency of control during WS qualification vs historical
  - Comparable potency
  - Comparable assay variance
- Confirmation run passed

# Conclusions

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- Accurate assignment of RS potency is critical for prevention of potency drift
- The use of a RS for potency determination requires additional qualification
- Qualification exercises increase in complexity as drug candidate development progresses

# Acknowledgments

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Evangelos Bakopanos

## Reference for fixed halfwidth confidence interval:

Chang YI and Martinsek AT, Fixed Size Confidence Regions for Parameters of a Logistic Regression Model, *The Annals of Statistics*, 20, 4, (1992), 1953-1969.