

Systematic Elements of Metrology, and how they work.

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What is Metrology?

- the stuff needed so data can support informed decision making
 - in a good world, decisions are informed with data
 - which are the results of measurements!
- systematic approach that tells us how to compare results, and how well we can trust them



Systematics

- Confidence and comparability rests on three legs:
 - Traceability
 - Measurement Uncertainty
 - Method Validation



Traceability

- tying results to a common reference
 - usually realized with calibration
 - enables comparison of results amongst those using the common reference
 - across space and time
 - the *System Internationale* (SI) is a nice set of references
- gives a “scale”
- biological measurement results are often traceable to the *control*
 - enrichment of a molecular signal



Measurement Uncertainty

- reasonable expectation of dispersion around my result
 - given *my* measurement system
 - a combination of all sources of variability or limitations in knowledge



Method Validation

- demonstration by provision of objective evidence...
 - that what I'm measuring is what I intend to be measuring
 - that it's fit-for-purpose
 - prove I'm not just reporting artifacts
- *Analytical Validation*, which is distinct from *Clinical Validation*



Confidence

- Sort these bits out, and I can be confident that my measurement results...
 - can be reproduced
 - are comparable
 - will take on a range of likely values, with a known likelihood
 - unbiased
- Now, I can make decisions.



Comparing results

- results are only useful when compared
 - to other results
 - e.g., to observe a trend
 - to limits
 - e.g., a threshold for action
 - different results in different places or measured at different times...
 - “comparability over space-and-time”



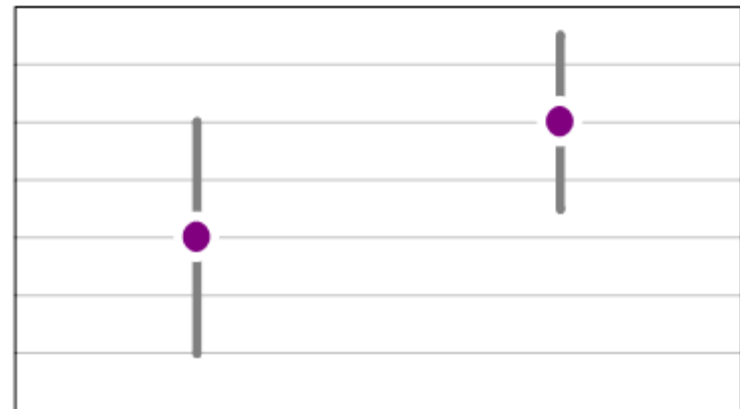
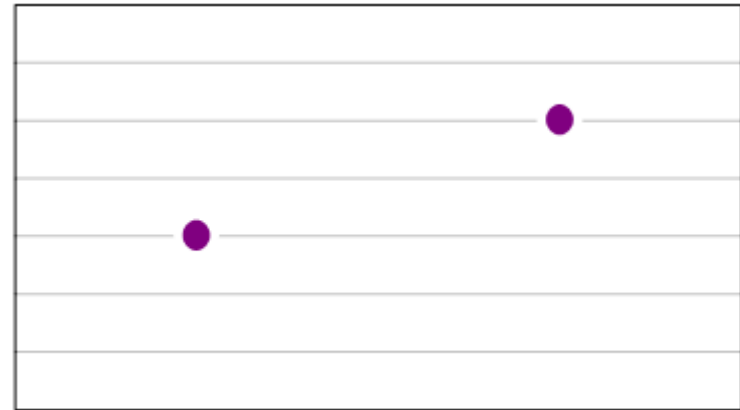
Comparability of results

- sole goal of traceability
- results linked to a common reference ***can be compared***
 - they're on a common scale
- scope of reference defines scope of comparability...
 - a global network
 - like the SI
 - or local standards
 - or ***shared references***



Measurement Uncertainty

- Are these results the same?
- how well do you know the result?
 - essential part of being able to compare!
- are these results good enough?
 - fit-for-purpose



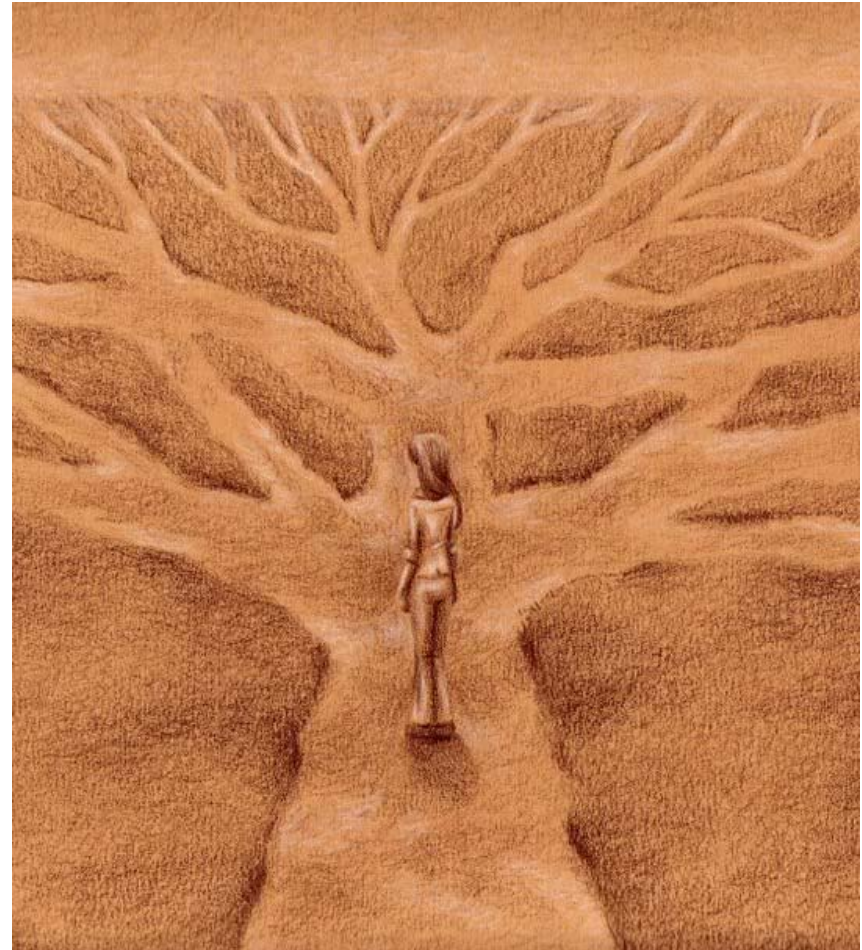
Traceability in chemistry...

- is different.
 - identity
 - what am I measuring, anyway?
 - interference
 - do I get the same response for analyte in my calibration material and in its matrix?
 - morphology
 - is the analysis the same everywhere in my sample?



Traceability in biological measurements...

- is even more different.
 - *usually traceable to control*
 - challenging to control conditions of complex measurements
 - often the property I'm measuring *is defined by the measurement*
 - difficult to control the scale
 - sometimes I'm measuring an identity
 - traceability ill-posed



Method Validation...

- *Method Validation* is a systematic way to...
 - establish scope
 - present a clear “measurement model”
 - evaluates bias
 - provide objective evidence that...
 - I’m measuring what I say I am
 - the result is robust
 - and fix it if it’s stable



Method validation

- “checks the model”
 - tests completeness
 - tests assumptions
 - helps establish an uncertainty budget
- identifies relevant parameters to keep under control
- tests scope

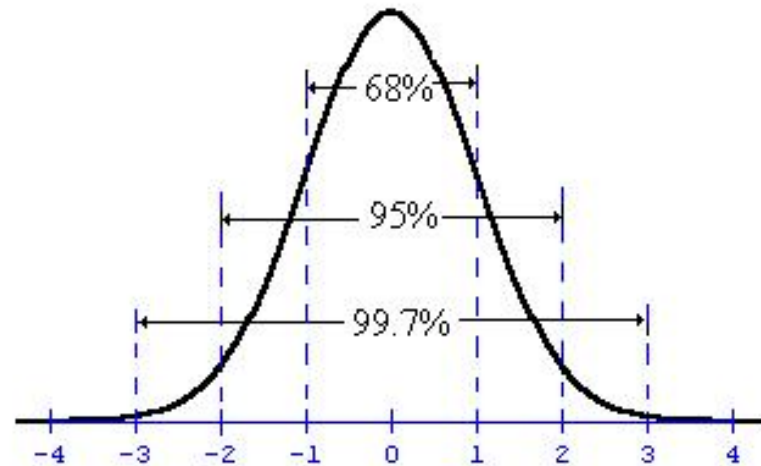


All of this,
taken together,
establishes
“reproducible
results”

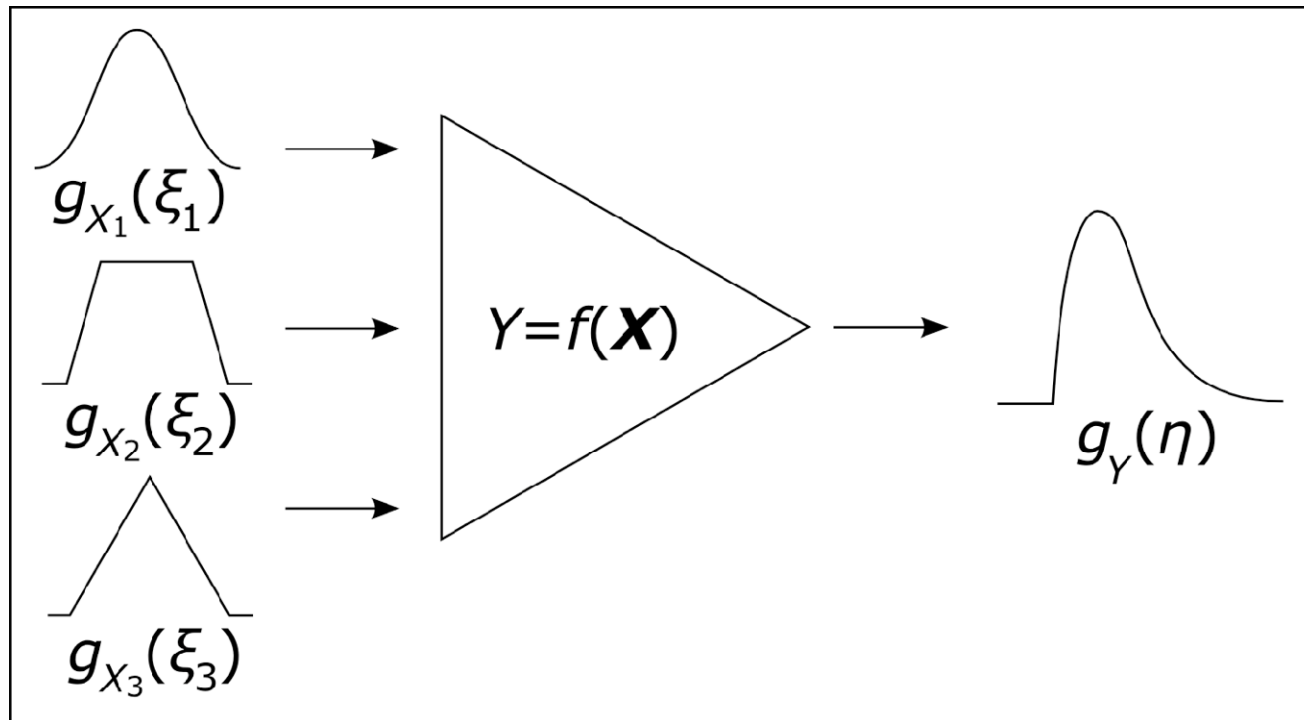


Measured Data are Samples from Distributions

- The *real* thing we're measuring has some distribution
 - we sample it
 - from our samples we infer the underlying distribution
 - our sampling is fraught with extra dispersion and biases
 - a lot of what we try to do is to separate out the underlying stuff that gives rise to the distribution of our measured data
 - because we want to know the distribution of the property of the *real* thing we're measuring



Basics of Measurement Uncertainty Calculation



PDF's of the influence quantities X_1, X_2, X_3

equation of the measurand

measurand Y

Ishikawa Diagram

Ishikawa diagram - Wikipedia, the free encyclopedia

Ishikawa diagram
From Wikipedia, the free encyclopedia

Ishikawa diagrams (also called **fishbone diagrams** or **cause-and-effect diagrams**) are diagrams that show the causes of a certain event. Common uses of the Ishikawa diagram are product design and quality defect prevention, to identify potential factors causing an overall effect. Each cause or reason for imperfection is a source of variation. Causes are usually grouped into major categories to identify these sources of variation. The categories typically include:

- **People:** Anyone involved with the process
- **Methods:** How the process is performed and the specific requirements for doing it, such as policies, procedures, rules, regulations, and laws
- **Machines:** Any equipment, computers, tools etc. required to accomplish the job
- **Materials:** Raw materials, parts, parts, paper, etc. used to produce the final product
- **Measurements:** Data generated from the process that are used to evaluate its quality
- **Environment:** The conditions, such as location, time, temperature, and culture in which the process operates

Contents [hide]

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Overview [edit]

Ishikawa diagrams were proposed by Kaoru Ishikawa^[1] in the 1960s, who pioneered quality management processes in the Kawasaki shipyards, and in the process became one of the founding fathers of modern management. It was first used in the 1950s, and is considered one of the seven basic tools of quality control.^[2] It is known as a fishbone diagram because of its shape, similar to the side view of a fish skeleton.

Motor's Motors famously used an Ishikawa diagram in the development of the Miata sports car, where the required result was "limbs (not) or "I know what I like as One". The main causes included such aspects as "touch" and "braking" with the lesser causes including highly granular factors such as "50/50 weight distribution" and "able to rest elbow on top of driver's door". Every factor identified in the diagram was included in the final design.

Causes [edit]

Causes in the diagram are often categorized, such as to the 4 Ms, described below. Cause-and-effect diagrams can reveal key relationships among various variables, and the possible causes provide additional insight into process behavior. Causes can be derived from brainstorming sessions. These groups can then be labeled as categories of the fishbone. They will typically be one of the traditional categories mentioned above but may be something unique to the application in a specific case. Causes can be traced back to root causes with the 5 Why's technique.

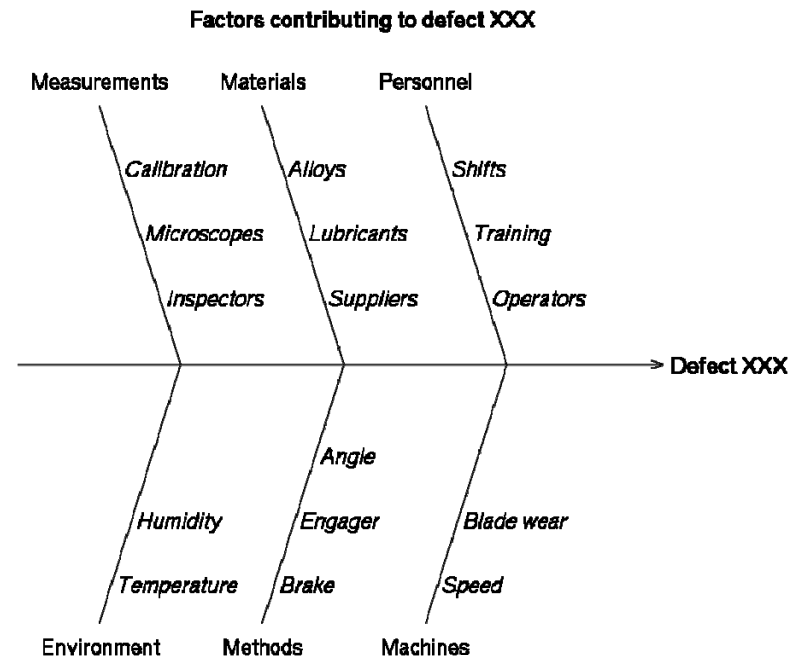
Typical categories are:

The 6 Ms (used in manufacturing) [edit]

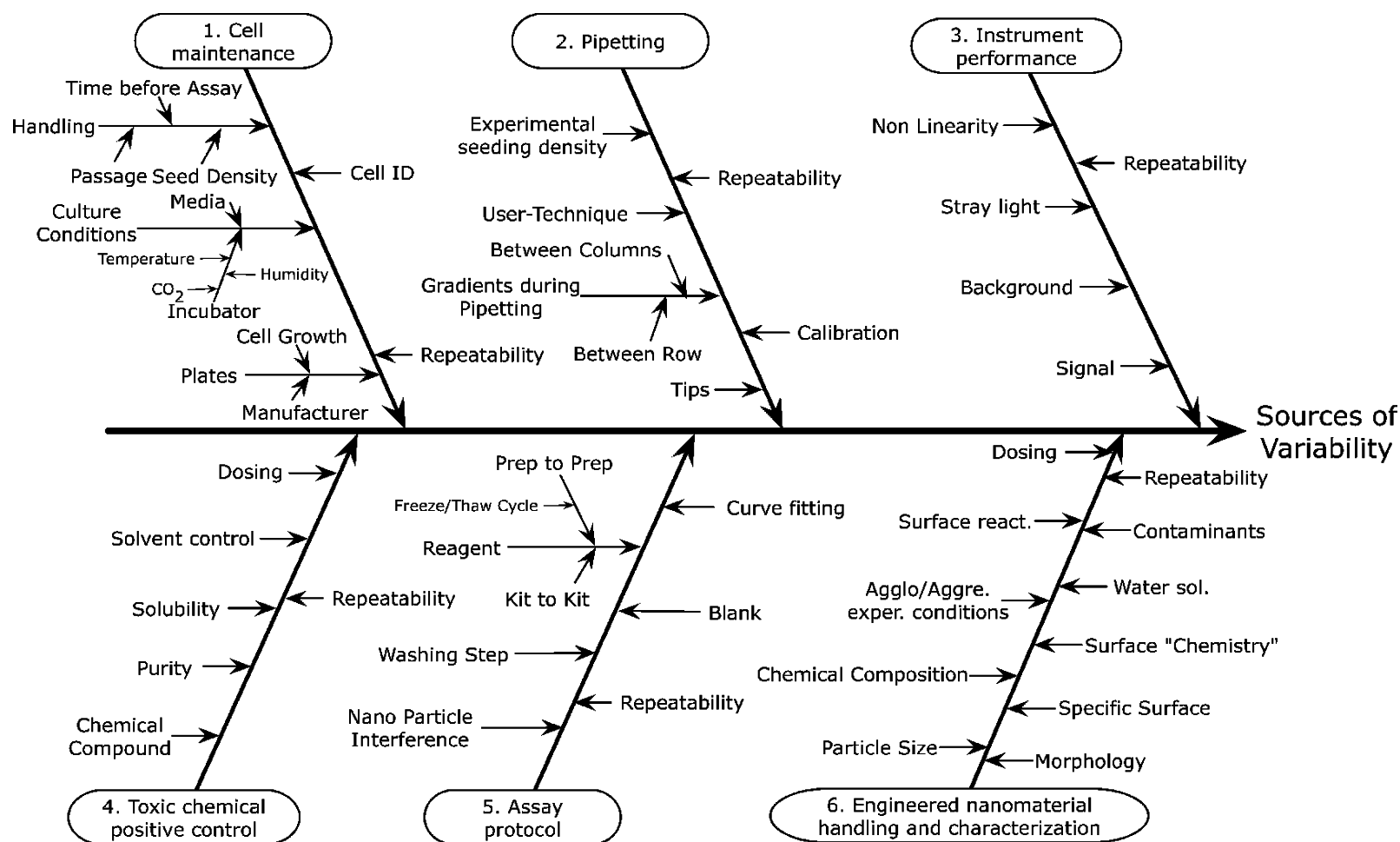
- **Machine** (technology)
- **Method** (process/inspection)
- **Material** (raw, consumable etc.)
- **Man Power** (physical work) Mind Power (brain work) Keiretsu, Suggestions
- **Money**
- **Milieu** (External Environment or surroundings)

The 8 Ps (used in service industry) [edit]

- **Product**—Service



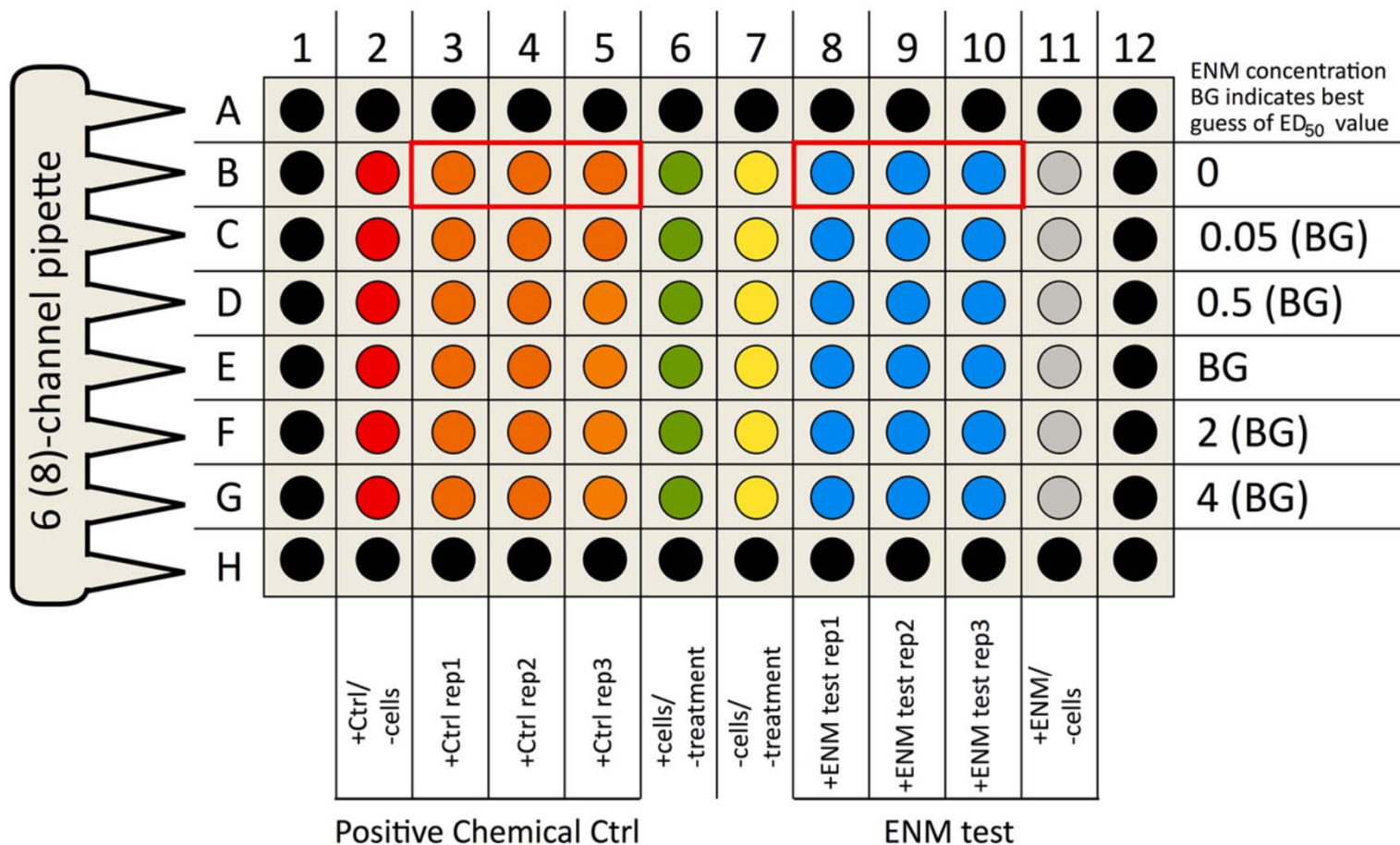
How does this look for a cell-based assay?



Use of Cause-and-Effect Analysis to Design a High-Quality Nanocytotoxicology Assay

Matthias Rösslein,^{†,‡} John T. Elliott,^{*,§,‡} Marc Salit,^{||} Elijah J. Petersen,[§] Cordula Hirsch,[†] Harald F. Krug,[‡] and Peter Wick[†]

How do we control in a cell-based assay?



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Potency, Strength are *method defined...*

viewed only as recommendations, unless specific regulatory or statutory requirements are cited. The use of the word *should* in FDA's guidances means that something is suggested or recommended, but not required.

II. BACKGROUND

A. What is Potency Testing?

Potency is defined as “the specific ability or capacity of the product, as indicated by appropriate laboratory tests or by adequately controlled clinical data obtained through the administration of the product in the manner intended, to effect a given result.” (21 CFR 600.3(s)). Strength⁶ is defined as “[t]he potency, that is, the therapeutic activity of the drug product as indicated by appropriate laboratory tests or by adequately developed and controlled clinical data. . . .” (21 CFR 210.3(b)(16)). Regulations require that “[t]ests for potency shall consist of either in vitro or in vi vo tests, or both, which have been specifically designed for each product so as to indicate its potency in a manner adequate to satisfy the interpretation of potency given by the definition in § 600.3(s) of this chapter.” (21 CFR 610.10).

Potency tests, along with a number of other tests, are performed as part of product conformance testing,⁷ comparability studies (Ref. 3), and stability testing (Ref. 4). These tests are used to measure product attributes associated with product quality and manufacturing controls, and are performed to assure identity, purity, strength (potency), and stability of products used during all phases of clinical study. Similarly, potency

What do I need to do for metrology of cell therapeutics?

- what results do I need to be able to compare?
- what are the main sources of variability of my measurement process?
 - what can I control?
 - what can I calibrate (and fix under control)?
- what are the main sources of variability in my test sample?
 - how can I control those, so my test sample is what I think it is?
 - what controls can I use to calibrate/normalize these sources?
 - media, passage number, incubation conditions...

Next thing you know, someone will suggest a “*reference cell line*”

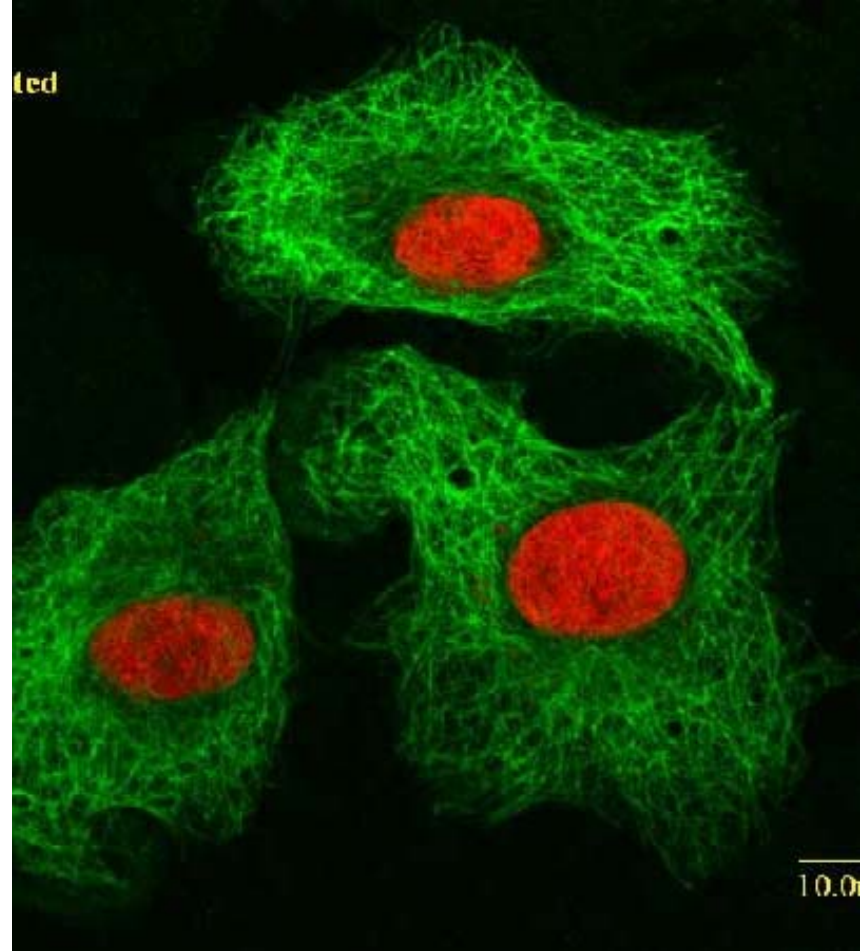
- What’s that reference cell line gonna do?
 - act to calibrate the measurement system?
 - give it a scale to report results on:
 - “1.375 more potent than the control.”
 - “58% as strong as the control.”
 - act to establish measurement uncertainty?
 - “when I run the reference, I see this dispersion.”
 - is the underlying distribution of the reference always the same?
 - can I deconvolve it from my measurement system?
 - act to validate?
 - these results are valid because I got the “*right answer*” on the reference cell line

Nothing to this.

but I need some things.

- I need an assay I can use to compare a test cell line against a reference cell line
 - for the property of interest
 - I need the reference cell line to behave identically to the test cell line in my assay, today and tomorrow
 - I need the reference cell line to be the same in my lab and another lab, always
- I need to understand the dispersion and bias of my measurement system
 - so I can compare meaningfully the sample and reference
 - so I can establish the scope of what will work
 - measurement properties AND cell properties effect this
 - different cell phenotype might have different response...

Measuring Biological Stuff



Craft

- Metrology is more akin to a craft than a technology
 - two different skilled metrologists might take different approaches to the same problem
 - *but they should both come to largely equivalent solutions!*
 - matter of style
 - **must be defensible**

