The Product Datasheet- Formulation and Antibody Performance

The Antibody Society Webcast series – Antibody Validation #4

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What to look for when shopping around?

- Be convinced that your choice will meet your expectations.
- Performance must be consistent from purchase to purchase.
- Product ideally will still exist and remains available 10-20 years later!
- The Product Datasheet provides the clues:

Fixed parameters:

- Catalog number and batch/clone number
- Names and symbols of protein
- GeneID and/or SwissProt accession
- Host species and isotype
- Antigen & Epitope
- Purification method
- Formulation (buffer components)
- Amount (mg or ml)

Claims of performance:

- Application claims
- Titre in ELISA
- Positive controls (tissues; cell types; cell lines)
- Successful usage claims in literature
- Data confirming molecular integrity
- Data confirming no cross-reactivity to closely related proteins
- Negative controls
- Data confirming successful use in applications

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What are the pitfalls to watch for?

Fixed parameters:

Shared Symbols. e.g.: OCT2 (SLC22A2, GeneID 6582 and POU2F2, GeneID 5452)

Renamed clones. e.g.: re-cloned hybridoma

Batch-to-batch inconsistency. e.g.: is next purchase from different animal or different purification

Aliquots from the same batch may differ in performance due to storage/handling history

Are you buying volumes or quantities?

Has the antibody been purified, and how? (e.g. NH_3SO_4-cut; Protein A; or antigen affinity)

Do you need a domain-specific or epitope-specific antibody?

In what formulation is the antibody offered (buffer components)?

What are the pitfalls to watch for?

Performance parameters:

Are the claimed applications supported by data (from literature or datasheet?)

Do the data presented comply with the science (example: is detection in correct cell compartment or tissue)

Are data presented fit-for-purpose? (wrong: FC on cell line; ICC staining entire cells; IHC with blurred stains)

Each application is demonstrated under different conditions (tested on peptide, protein, cell type, etc)

Is the antibody compared with another gold-standard one?

Has the antibody been tested on both expressing and non-expressing cells/tissues?

Has the antibody been tested on closely related proteins (from the same protein family)**?**

Are comparisons all done at the same time and at the same dilution/antibody concentration?

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Summary

- Vendor may provide evidence, but scientist must verify.
- Choose antibody shown fit for the assay type you need it for.
- Ensure the antibody works in the cell types you need it for.
- Selectivity (correct negatives) trumps specificity (correct positives)

Read more on the subject:

Voskuil (2017) The challenges with the validation of research antibodies. F1000Res 6:161.

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What antibody have I found?

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Next Webcast in Antibody Validation: a 9-part series

1.	Andreas Pluckthun	:	The different antibody formats
2.	Glenn Begley	:	Antibodies and the reproducibility crisis in biological science
	Cecilia Williams	:	The Erß story – is your antibody like this?
3.	Jan Voskuil	:	Beware the supplier OEM
	Andy Chalmers	:	Finding antibodies in the Antibody Databases
4.	Anita Bardowski	:	Which antibody are you looking for? The RRID
	Jan Voskuil	:	Points to note on the supplier datasheets
5.	**Giovanna Roncador:**	**:**	**Correct positive and negative controls in validation**
6.	Aldrin Gomes	:	Standard technology: "even" Western blots are non-trivial
	Jim Trimmer	:	IHC issues in brain sciences
7.	Travis Hardcastle	:	Cell KO technology
	Alejandra Solache	:	Validating Antibodies with KO technology
8.	Mike Taussig	:	Validating antibodies using array technologies
	Fridjhof Lund-Johansen	:	Mass spectroscopy for mass validation
9.	Andrew Bradbury	:	Why publish sequences?
	Andreas Pluckthun	:	What are the coming alternatives ?

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Validation of Commercial tool Antibodies

What antibody have I found?

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https://www.antibodysociety.org/

Validation of Commercial Tool Antibodies

An Antibody Society Webcast series

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