Verifications of new lots and expired antibodies

Hazel Chambers-Smith

Sri Lanka Workshop on Basics of Immunohistochemistry, June 2018

Verifications

- In Australia, we are governed by NATA and the requirements of Australian standard
- ISO15189 Medical laboratories — Requirements for quality and competence

What the standard means

- The standard tells us what we must/should/shall do in order to meet the requirement.
- In the case of reagent management, the standard is clear…

5.3.2 Reagents and consumables

5.3.2.1 General

The laboratory shall have a documented procedure for the reception, storage, acceptance testing and inventory management of reagents and consumables.

5.3.2.2 Reagents and consumables — Reception and storage

Where the laboratory is not the receiving facility, it shall verify that the receiving location has adequate storage and handling capabilities to maintain purchased items in a manner that prevents damage or deterioration.

The laboratory shall store received reagents and consumables according to manufacturer's specifications.

5.3.2.3 Reagents and consumables — Acceptance testing

Each new formulation of examination kits with changes in reagents or procedure, or a new lot or shipment, shall be verified for performance before use in examinations.

Consumables that can affect the quality of examinations shall be verified for performance before use in examinations.

Verification of a new lot of an antibody

- In an ideal world, antibody lots would never change and we would have consistent, reproducible, clean staining every time.
- In the real world, batches change, there are challenges in manufacturing processes.
- This can lead to subtle changes in how an antibody can behave

What can cause changes in how an new antibody behaves?

- Changes in cell lines
- Changes in temperature during transport (eg shipped at room temperature instead of -20°C).
- Issues in manufacturing
- Beware of non-IVD products

How can we check

- Keep on ongoing log of lot numbers
  - Date of receipt
  - Date in use (ability to trace patients affected by poor reagents)
  - Date of expiry
  - Date of discard

Lot to lot tracking

- You can keep electronic or paper based lot tracking (do whatever is easiest and what people will reliably complete)
- Lot numbers can be tracked on the instrument too
- New lot checked slides ran can be filed in a yearly archived system by date order or by antibody name
- You could also just log which case was ran with the new lot (slide could be retrieved later)

Antibody	Ordering Details								Budget Information			Performance Check			
	Order Date	Supplier	Product Code	Oracle Req	Volume	Date Rec	Expiry	Lot	Cost	Qty	Total	Date	Control	Against Lot	By
	2017-04-18	VENTANA	06419216001	1105828	50 test	########	########	1626504C	$641.16	1	$641.16	########	HNPCC	N/A	MC
	2017-06-14	VENTANA	06419216001	1128168	50 test	########	########	V0000059	$641.16	1	$641.16	########	HNPCC	N/A	MC
	2017-08-02	VENTANA	06419216001	1147796	50 test	########	########	V0000163	$641.16	1	$641.16	########	HNPCC	N/A	MC
PMS2	2017-09-25	VENTANA	06419216001	1168866	50 test	########	########	V0000798	$641.16	1	$641.16	########	HNPCC	N/A	BB
	2017-12-21	VENTANA	06419216001	1202742	50 test	########	########	v0000798	$641.16	1	$641.16	########	HNPCC	N/A	HCS

g Details					Budget Information			Performance Check				In Use	Notes
Oracle Req	Volume	Date Rec	Expiry	Lot	Cost	Qty	Total	Date	Control	Against Lot	By		
1105828	50 test	########	########	1626504C	$641.16	1	$641.16	########	HNPCC	N/A	MC	2017-06-08	
1128168	50 test	########	########	V0000059	$641.16	1	$641.16	########	HNPCC	N/A	MC	2017-08-02	
1147796	50 test	########	########	V0000163	$641.16	1	$641.16	########	HNPCC	N/A	MC	2017-09-25	
1168866	50 test	########	########	V0000798	$641.16	1	$641.16	########	HNPCC	N/A	BB	2017-11-21	
1202742	50 test	########	########	v0000798	$641.16	1	$641.16	########	HNPCC	N/A	HCS	2018-01-18	

- Just remember whatever system you put in place, make is as simple as possible.
- It is the individual lab that determines how the system is set up
- Any external assessors just need to be able to see it demonstrated

Controls

- Stain a new antibody with a known positive, and known negative
- Often good to use a multi-tissue block
  - Epithelial – eg Cytokeratins,
  - Stromal tissue – eg Vimentin,
  - Endothelial – eg CD31,
  - Lymphoid tissue – T and B Cells
  - Neural tissue – eg GFAP
- May require a specialised tissue component
  - Her-2, INI-1, BRAF and MMR proteins

- Record outcomes of testing – it is ok for staining patient tissue?
- Best to use on-slide controls rather than a separate slide containing control only.

Expired antibodies

- Our standard doesn’t specify we cannot use an expired antibody.
- The standard specifies the following..

5.3.2 Reagents and consumables

5.3.2.1 General

The laboratory shall have a documented procedure for the reception, storage, acceptance testing and inventory management of reagents and consumables.

5.3.2.2 Reagents and consumables — Reception and storage

Where the laboratory is not the receiving facility, it shall verify that the receiving location has adequate storage and handling capabilities to maintain purchased items in a manner that prevents damage or deterioration.

The laboratory shall store received reagents and consumables according to manufacturer's specifications.

5.3.2.3 Reagents and consumables — Acceptance testing

Each new formulation of examination kits with changes in reagents or procedure, or a new lot or shipment, shall be verified for performance before use in examinations.

Consumables that can affect the quality of examinations shall be verified for performance before use in examinations.

How to manage expired antibodies?

- Best practice is to not use them!
- In reality that is easier said than done!
- Know that expired antibodies can behave differently to expected staining.
- Reagent log would contain information about date received, in use date and patient tissue stained
- Would be easy to identify affected cases

- Some instances there is no reason to not use an expired antibody
  - Antibody must stain in the same manner as optimised
  - No changes in staining patterns
  - No background
  - On-slide control should be enforced so that test and control conditions are the same and test can be treated as valid.

Ordering reagents

- Regular ordering can minimise accumulating too many antibodies that will expire
- We train all of our staff and review what they are going to order
- We print an inventory and do a physical check

Expired RTU's

- Seem to be reliable for the majority of antibodies eg CKIT, Ca125, INI1 etc
- Using on slide controls ensures every slide is verified using the most suitable control

Expired Concentrates/ RUO antibodies

- Much more variable results
- In my experience, -20 stored antibodies have shown aberrant staining 12 months after receipt, giving worrying false positive expression
- Example K27mutant on a brain tumour
- But some IF antibodies still show great signal 20 years past expiry…

The US has regulations on the use of expired antibodies:

- Clinical Laboratory Improvement Act of 1988 by the College of American Pathologists.
- These regulations mandate that expired reagents cannot be used in the clinical diagnostic laboratory on human tissue.

Expired antibodies

- Depending on the product usage you can:
- - Register the antibody as a prep kit (prep kit buttons are usually not free so are a hidden cost to your consumables budget)
- -Change the protocol to titration for that antibody (more inconvenient to use, increased error risk, but doesn’t cost extra)
- Hard to monitor when titration as the check becomes visual-likely to run out

In summary

- Make your logging system simple
- Using on-slide controls is critical
- Using expired antibodies is acceptable as long as results are verified
- Try to manage stock to avoid having many expired antibodies
- Understanding product usage can help tailor your order
- Training staff on ordering is helpful


The Royal
Children's
Hospital
Melbourne