Updates in Autoimmune Neurology

Phenotype-Specific Testing and Avoiding Misdiagnosis

Tammy Smith, MD PhD

Assistant Professor, Department of Neurology, Division of Neuroimmunology, University of Utah
GRECC Researcher at Salt Lake City VA Medical Center
Clinical Consultant, ARUP Laboratories

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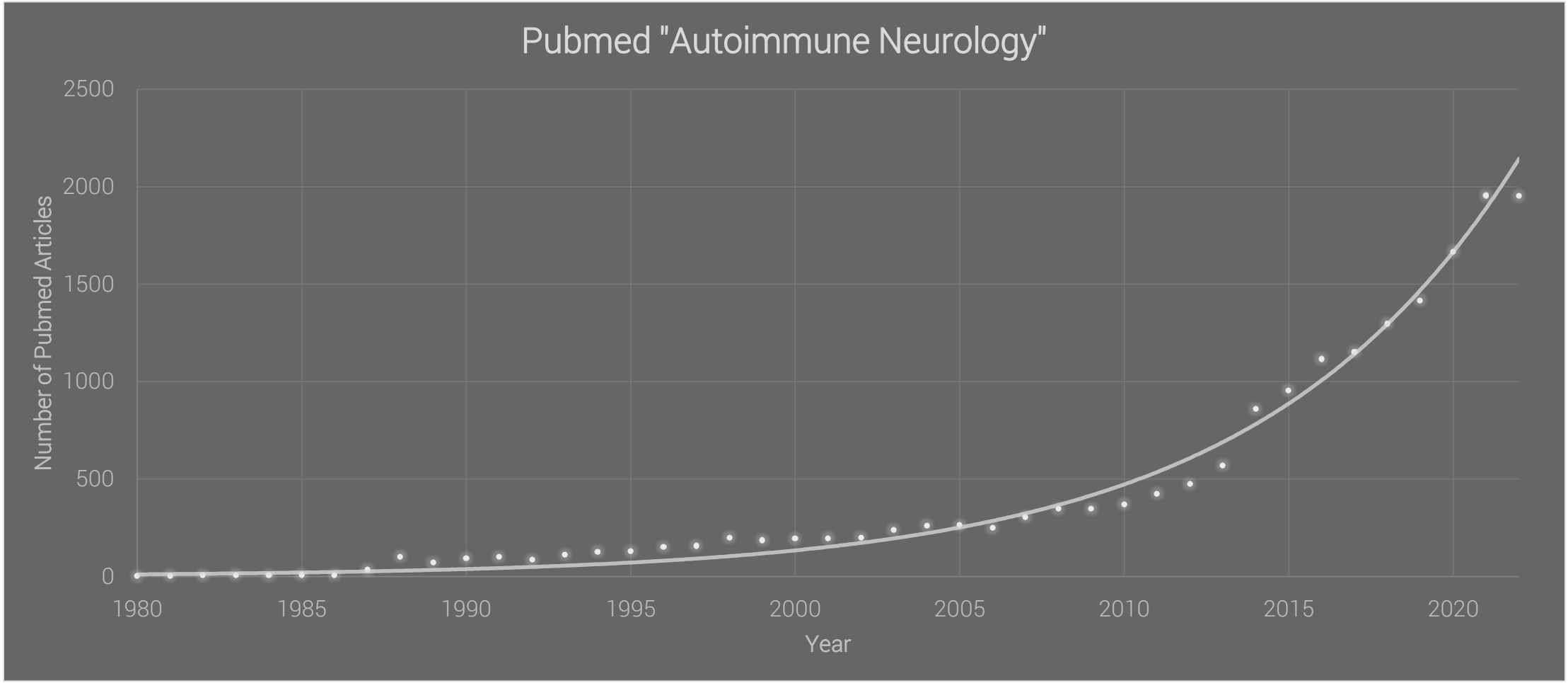
Immunity is beneficial only to the extent that it is effectively dealing with a threat more dangerous than itself.

The Field of Autoimmune Neurology is Growing Rapidly



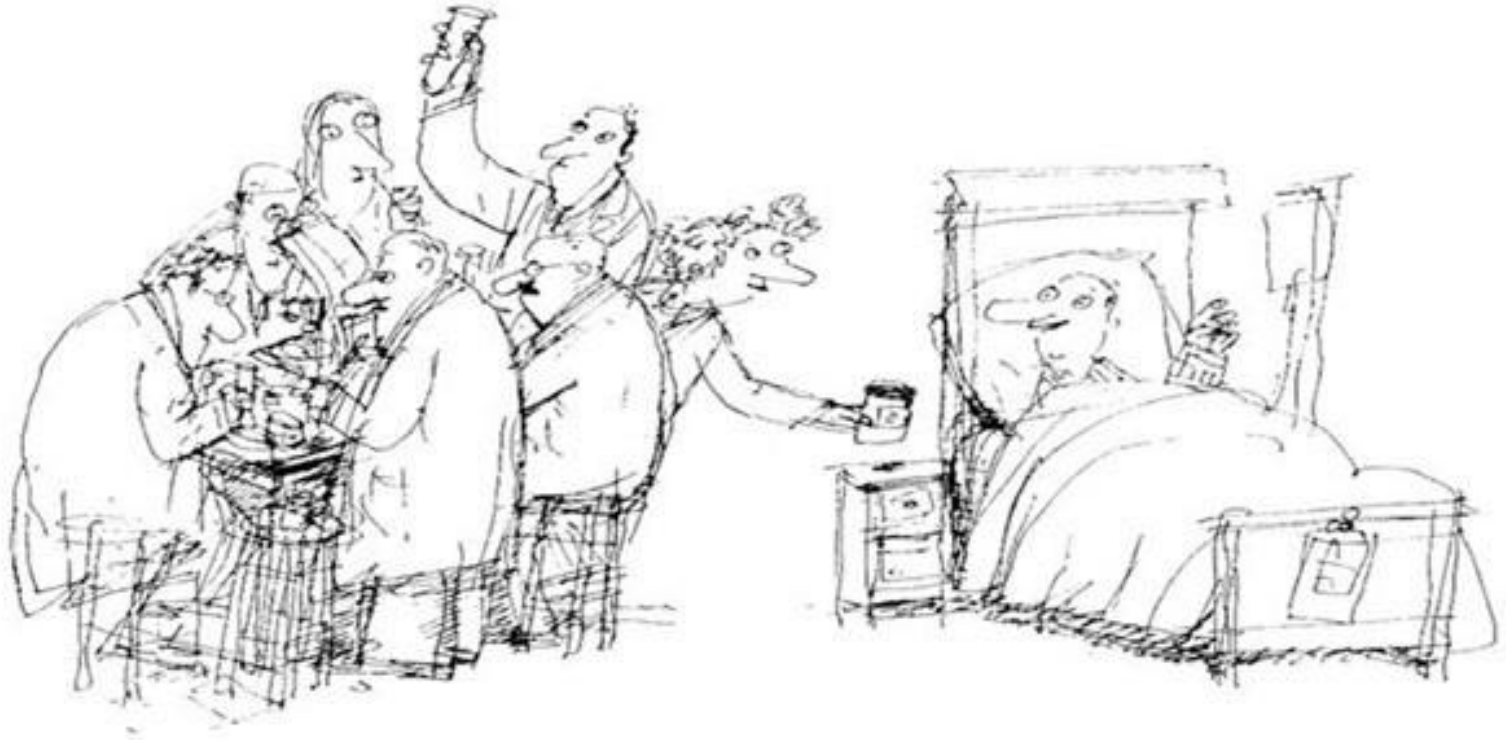
Learning Objectives

- Describe how the localization of antineural antibody targets impacts testing strategies and informs treatment decisions
- Identify clinical characteristics which support testing for antineural antibodies
- Recognize how phenotype-specific panels can be utilized to increase the chance of obtaining clinically meaningful results
- Describe the most common pitfalls which lead to misdiagnosis of immune-mediated neurologic disorders

Anti-neural Antibody Targets

Localization Informs Testing Strategies and Treatment Decisions

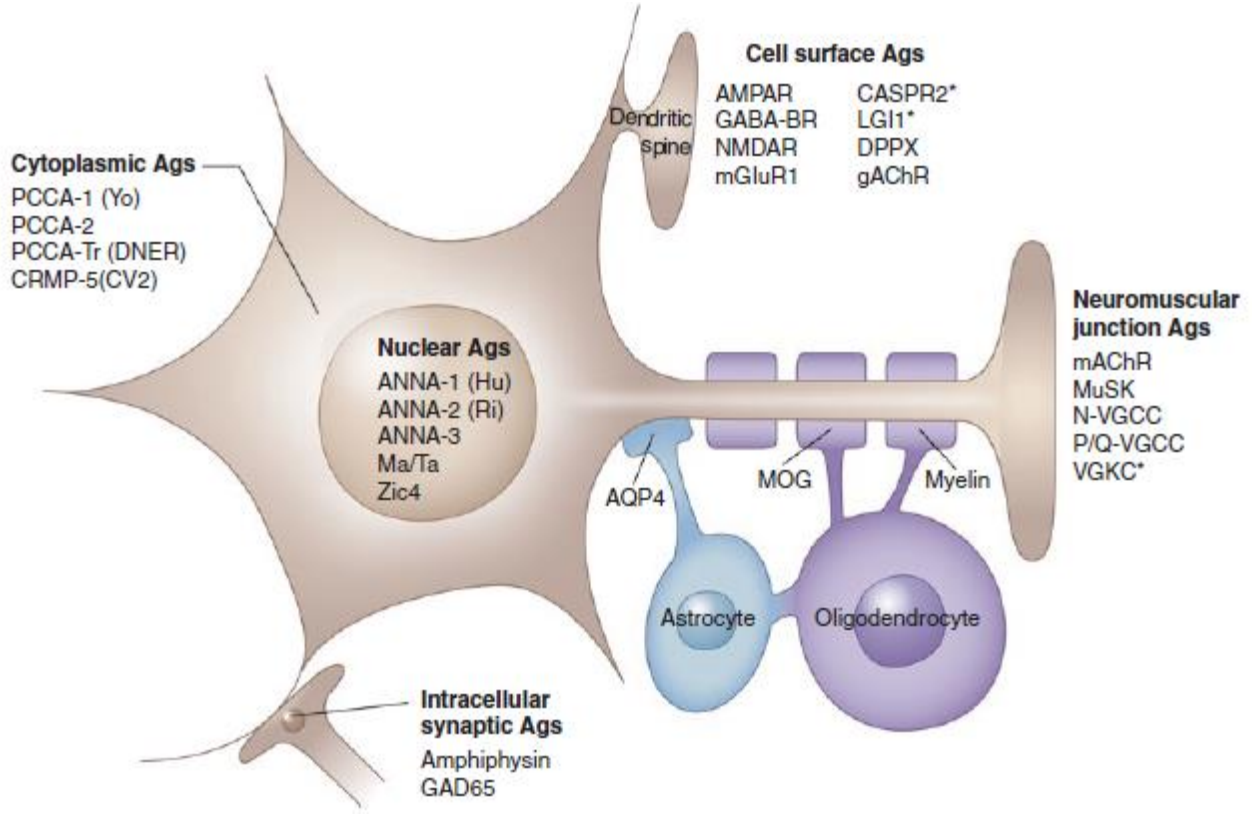
Bedside to Bench to Bedside Medicine



SOURCE: NATIONAL INSTITUTES OF HEALTH (NIH) – BENCH TO BEDSIDE PROGRAM

Anti-Neural Antibodies Are Historically Classified Based on Pathologic Localization

- Intracellular
  - » Nuclear
  - » Cytoplasmic
  - » Enzymes
  - » Transcription factors
  - » RNA binding proteins
- Plasma Membrane/Secreted
  - » Neurotransmitter receptors
  - » Ion channels
  - » Ion channel complex components
  - » Water channels



From: Haven and Peterson, Neuroimmunology ISBN: 978-3-030-61883-4

Case #1

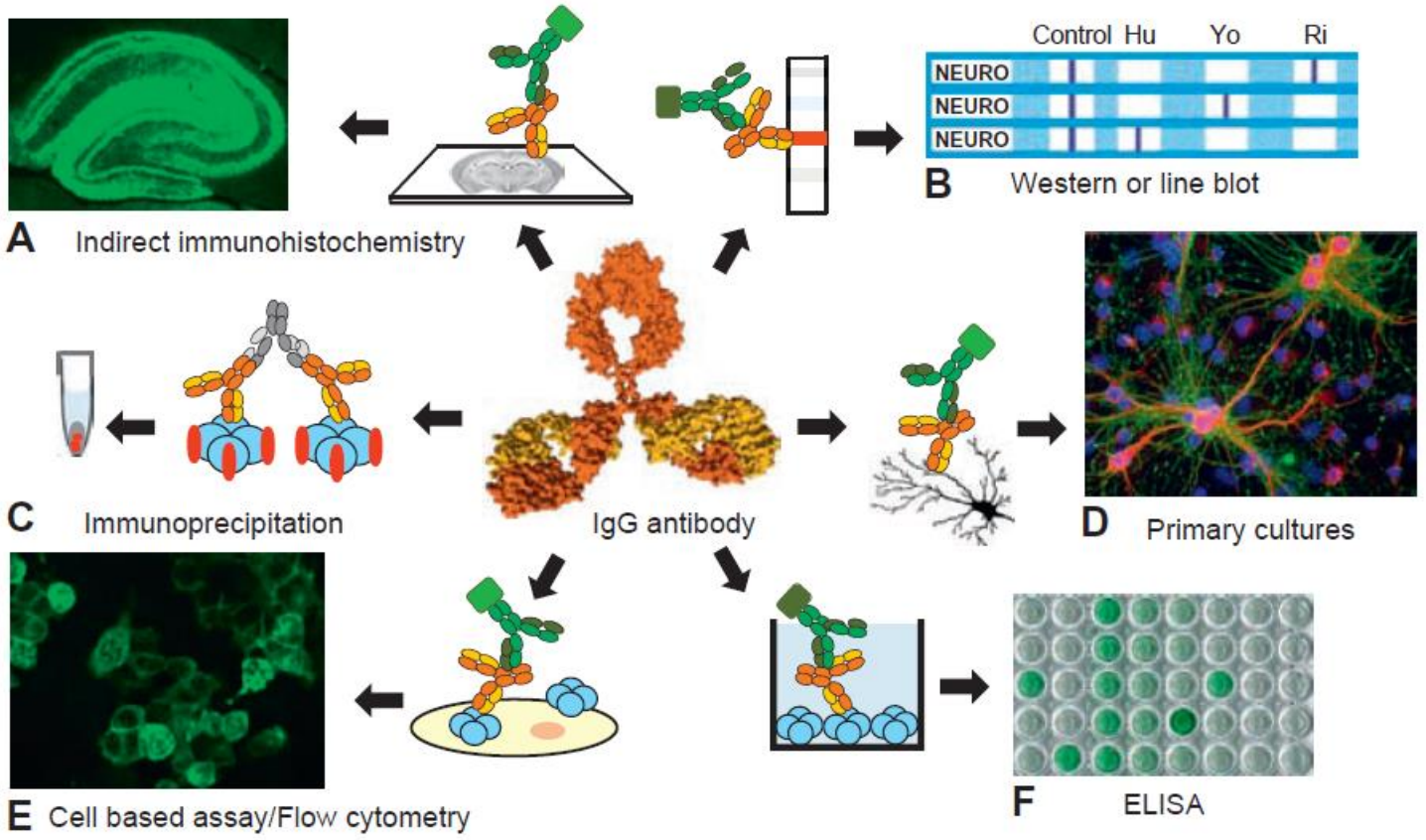
61yo previously healthy female

- Was at the grocery store when she first noticed poor coordination in her LEFT foot
- Progressed over 3 months to the point that she was wheelchair-bound and needed help with all ADLs
- Developed dysarthria

Testing

- Initial MRI brain, CT C/A/P normal
- Serologic tests for syphyilis, HIV, vitamin B12 normal
- Basic CSF studies normal
- Paraneoplastic antibody panel from serum positive for PCA-1 (anti-Yo) by both IFA and line blot

Detection of Neural Autoantibodies in the Clinical Laboratory



Waters et al. Handbook of Clinical Neurology, Vol. 133, Chapter 9, pgs.147-163

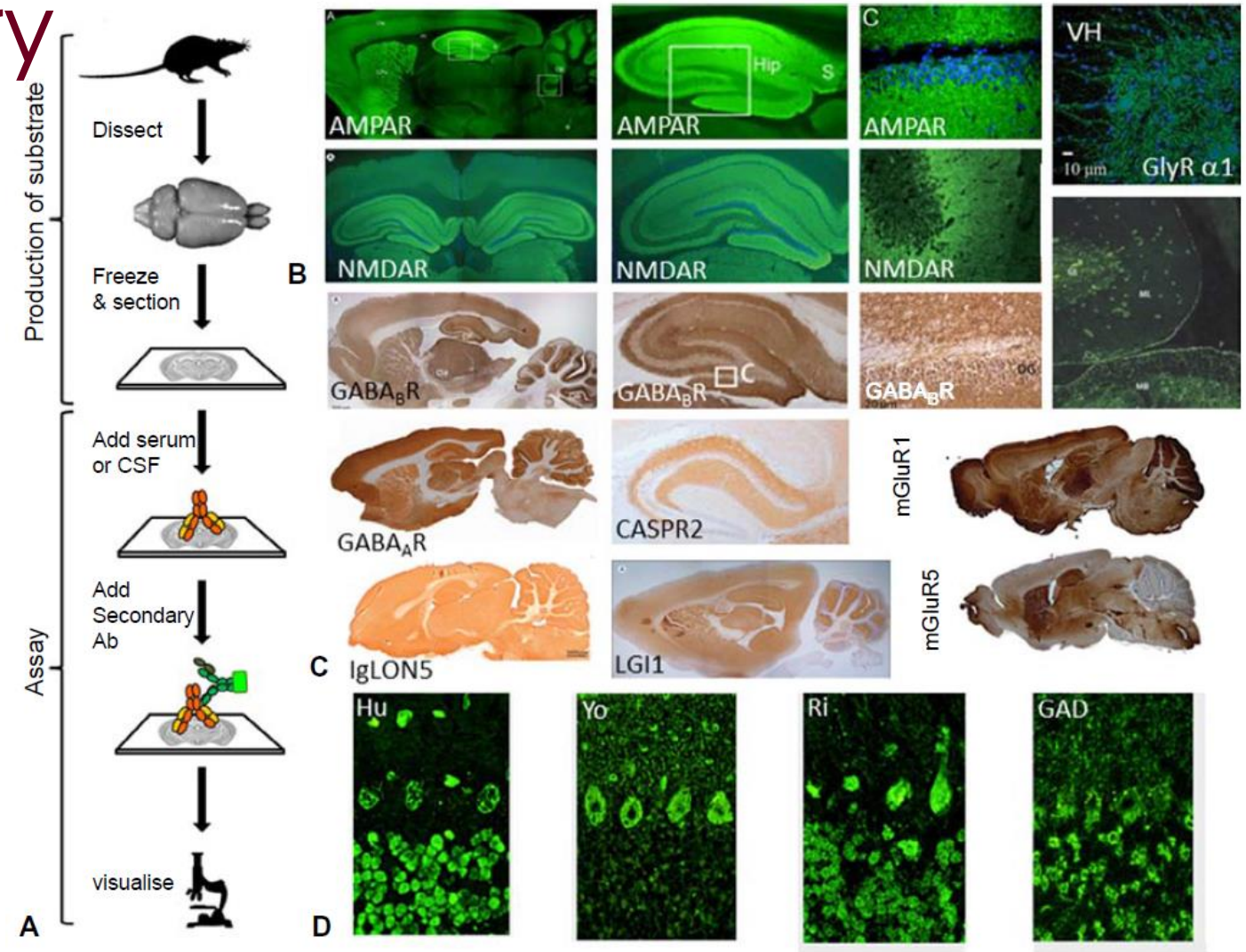
Tissue-Based Indirect Immunofluorescence or Immunohistochemistry

Advantages

Antigens are in their native setting
Simultaneously screen for many antibodies
Can discover new autoantibodies

Disadvantages

Requires significant training
Staining pattern may not be unique
Difficult to identify multiple coexisting antibodies
Rare antibodies pose issues with validation and maintaining competency
Subjective
Time consuming
Lacks standardization
Requires a second method to confirm specific autoantibody



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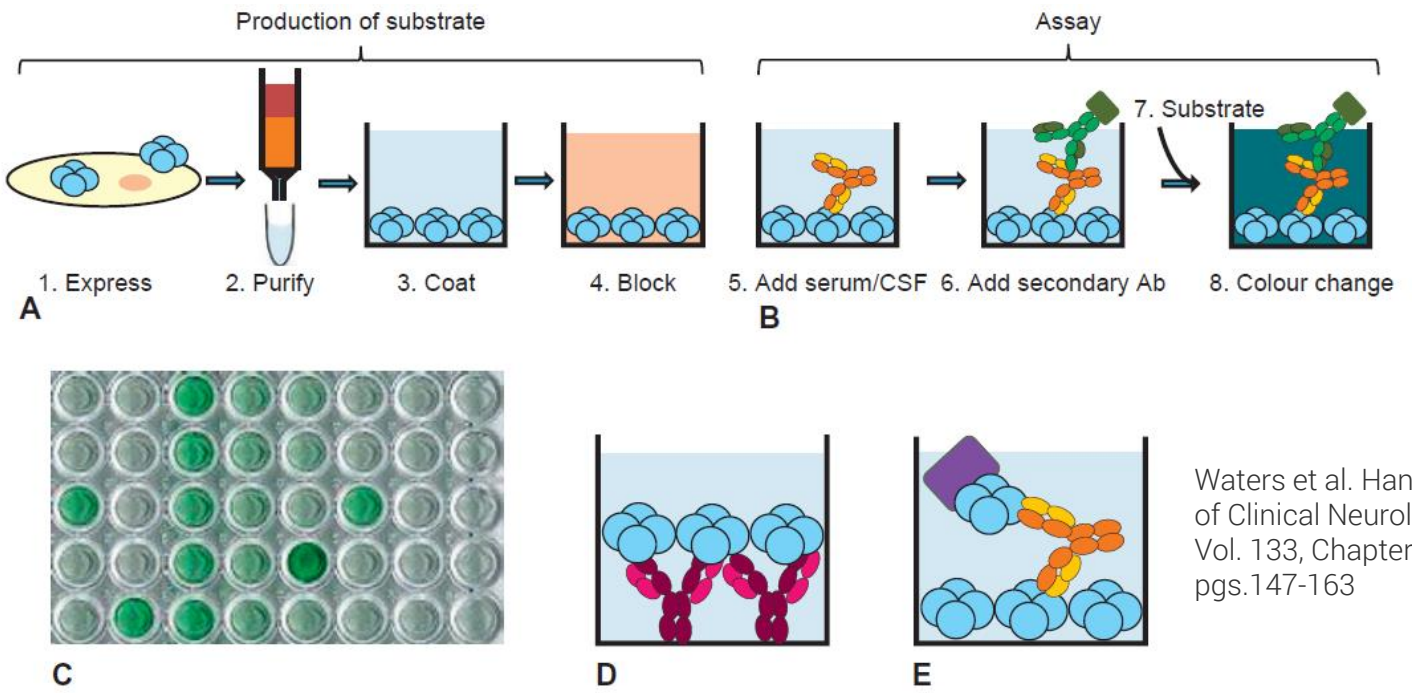
ELISAs



Waters et al. Handbook of Clinical Neurology, Vol. 133, Chapter 9, pgs.147-163

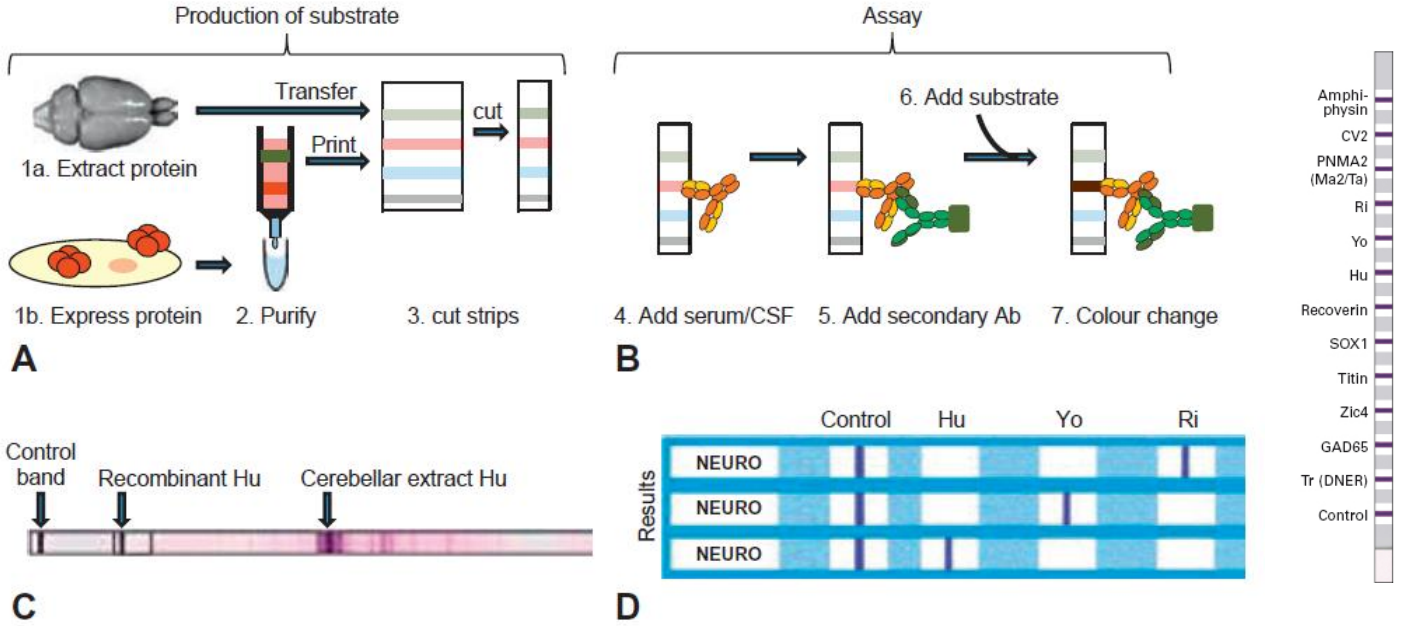
Advantages

- Less subjective than IFA
- Higher throughput, can be automated

Disadvantages

- Antigens may not be in their native form (false negatives and false positives)
- False positives due to nonspecific binding (plate, heterophile antibodies, etc.) or detection of IgM and IgA antibodies (panel E)

Western Blot or Line Blot Testing



Adapted from Waters et al. Handbook of Clinical Neurology, Vol. 133, Chapter 9, pgs.147-163 and www.euroimmun.com

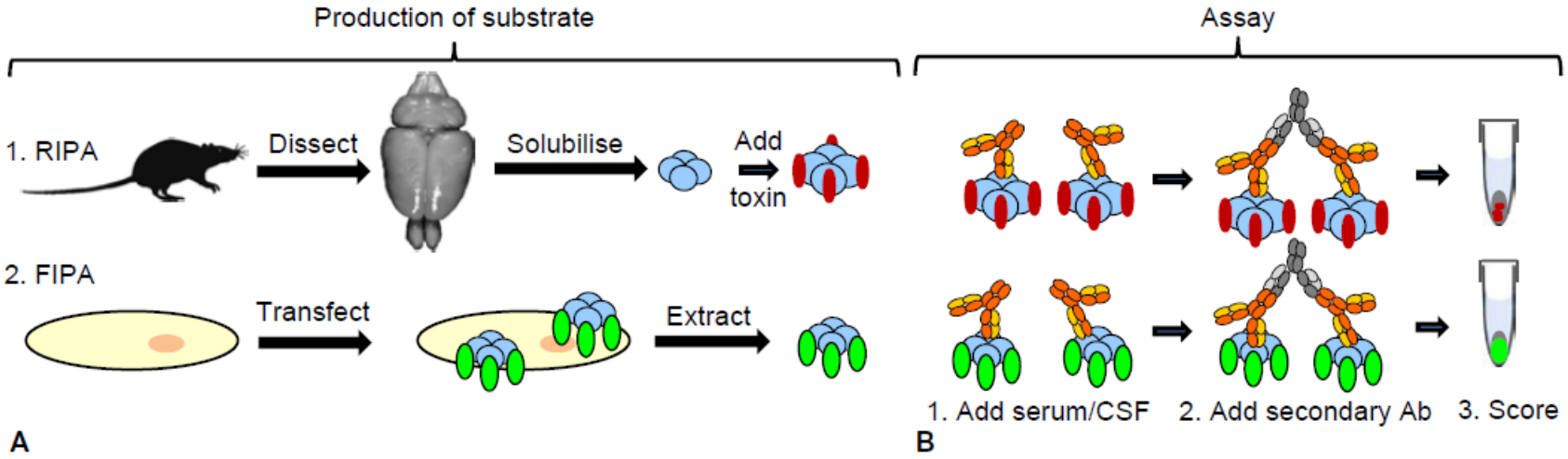
Advantages

- Can screen for and identify multiple antibodies at the same time
- Less subjective than IFA
- Higher throughput, can be automated

Disadvantages

- Antigens are not in their native form (false negatives and false positives)
- Can be difficult to obtain rare positive samples for validation and as controls (manufacturer controls often contain a single antibody)
- Clinical relevance of WB or IB positive but IFA negative results is questionable

Radio- or Fluorescent Immunoprecipitation Assays



Waters et al. Handbook of Clinical Neurology, Vol. 133, Chapter 9, pgs.147-163

Advantages

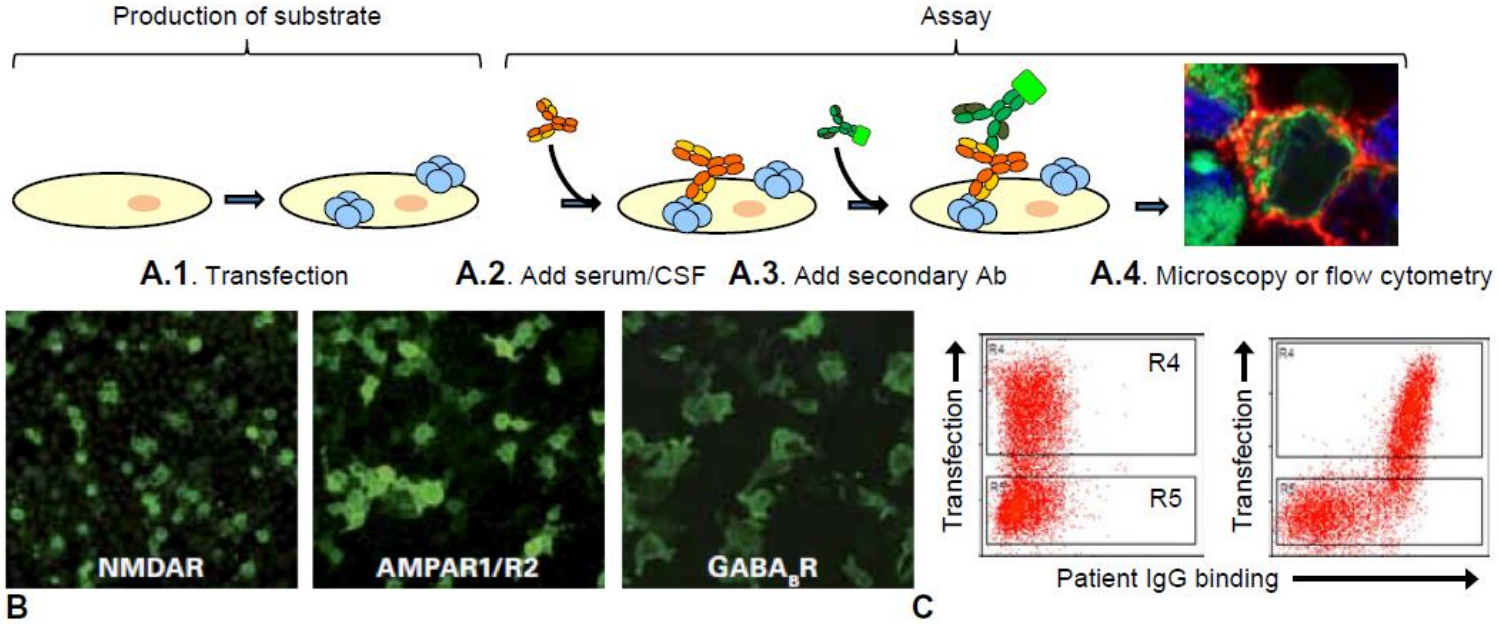
- Antigens are in their native form
- Increased sensitivity compared to IFA, WB, LB, ELISA
- Less subjective than IFA

*Preferred method for detecting antibodies to synaptic receptors

Disadvantages

- Radioactivity
- May identify multiple autoantibodies due to immunoprecipitation of a protein complex, which may have to be confirmed using an additional assay (eg. VGKC complex ➔ LGI1 and CASPR2)

Cell-Based Assays (CBAs)



Adapted from Waters et al. Handbook of Clinical Neurology, Vol. 133, Chapter 9, pgs.147-163 and www.euroimmun.com

Advantages

- Antigens are not purified
- Less subjective than IFA
- Requires less training for proficiency
- Very sensitive and specific method for detecting antibodies against many of the cell surface targets

Disadvantages

- Can only be used to detect antibodies against the transfected antigen
- Can't identify new autoantibodies

*Preferred method for detecting antibodies to cell surface receptors

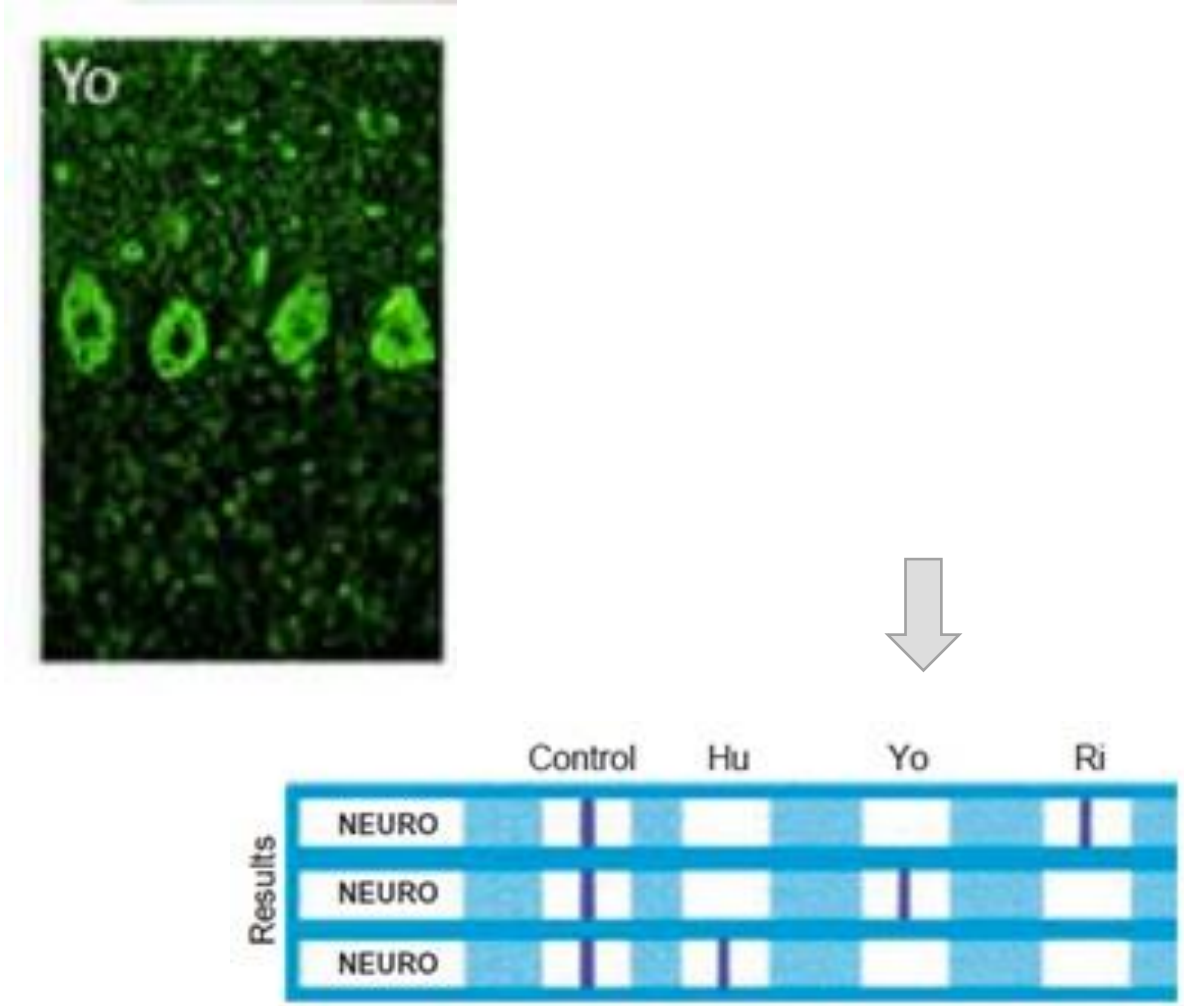
Autoantibodies and Methods For Their Detection in the Clinical Laboratory

Category	Specific antibody	Detection methods[a]					
		TBA	WB/LIA	RIA	ELISA	CBA/IFA	CBA/FACS
Intracellular antigens	AGNA-1 (Sox-1)	x	x	–	x	–	–
	Amphiphysin	x	x	–	x	–	–
	ANNA-1 (Hu)	x	x	–	x	–	–
	ANNA-2 (Ri)	x	x	–	x	–	–
	ANNA-3	x	–	–	–	–	–
	CRMP-5 (CV2) (CV2)	x	x	–	x	x	–
	GAD65	x	x	x	x	–	–
	Ma/Ta	x	x	–	–	–	–
	PCCA-1 (Yo)	x	x	–	x	–	–
	PCCA-2	x	–	–	–	–	–
	PCCA-Tr (DNER)	x	x	–	x	–	–
	Recoverin	x	x	–	x	–	–
	Titin	–	–	–	x	–	–
	Zic4	x	x	–	–	–	–
Neural cell-surface antigens	AMPAR	x	–	–	–	x	–
	AQP4	x	x	–	x	x	x
	CASPR2	x	–	–	–	x	–
	DPPX	x	–	x	–	x	–
	gACHR	–	–	x	–	–	–
	$GABA_BR$	x	–	–	–	x	–
	LGI1	x	–	–	–	x	–
	mGluR1	x	–	–	–	x	–
	MOG	x	–	–	–	x	x
	Myelin	x	–	–	–	x	–
	NMDAR	x	–	–	x	x	–
Neuromuscular junction antigens	mACHRBIN	–	–	x	–	–	–
	MuSK	–	–	x	–	–	–
	N-VGCC	–	–	x	–	–	–
	PQ-VGCC	–	–	x	–	–	–
	STR	x	–	–	x	–	–
	VGKC	–	–	x	–	–	–

From: Haven and Peterson, Neuroimmunology ISBN: 978-3-030-61883-4

Haven and Peterson. Neuroimmunology, Chapter 2, pgs.15-27

Case #1



Testing

- Initial MRI brain, CT C/A/P normal
- Serologic tests for syphyilis, HIV, vitamin B12 normal
- Basic CSF studies normal
- Paraneoplastic antibody panel from serum positive for PCA-1 (anti-Yo) by both IFA and line blot

When To Test for Antineural Antibodies

Clinical Context Provides Important Clues

Could This Be Autoimmune?

- Acute to subacute onset of symptoms, rapid progression
- Inflammatory cerebrospinal fluid
- Personal or family history of autoimmunity
- History of cancer, or elevated cancer risk factors, or ICI exposure
- Neuroimaging suggestive of inflammation

APE² score

New onset, rapidly progressive mental status changes or new onset sz	1		Facial Dyskinesias	2
Neuropsychiatric changes; agitation, aggressiveness, emotional lability	1		Seizure refractory to >2 ASM	2
Autonomic dysfunction	1		Inflammatory CSF	2
Viral prodrome	2		Brain MRI suggestive of encephalitis	2
Fasciobrachial dystonic seizures	3		Systemic cancer diagnosed within 5y of symptom onset	2

Maximum score: 18

Scores ≥4

doi.org/10.1093/jalm/jfab106

Case #2

19yo previously healthy female

- Brought to the hospital after family noticed 2 weeks of severe insomnia, and 10 days of increasing confusion, agitation, and paranoia
- While undergoing initial evaluation, she had a witnessed GTC; subsequently noted to have frequent orofacial dyskinesias

Testing

- CSF protein 63, 9 WBC (90% lymphocytes), positive OCBs, HSV PCR negative
- MRI brain unremarkable
- APE² score = 6
- NMDAR Ab in CSF positive
- Pelvic MRI showed RIGHT ovarian teratoma

Could This Be Seronegative Autoimmune?

All 4 of the following criteria must be met:

- Rapid progression (<3 months) of working memory deficits, altered mental status, or psychiatric symptoms
- Exclusion of well-defined syndromes of autoimmune encephalitis
- Absence of well-characterized autoantibodies in serum and CSF AND AT LEAST TWO of the following
  - » MRI abnormalities suggestive of autoimmune encephalitis
  - » CSF pleocytosis, CSF-specific OCBs or elevated CSF IgG index (or both)
  - » Brain biopsy showing inflammatory infiltrates and excluding other disorders (eg: tumor)
- Reasonable exclusion of alternative causes

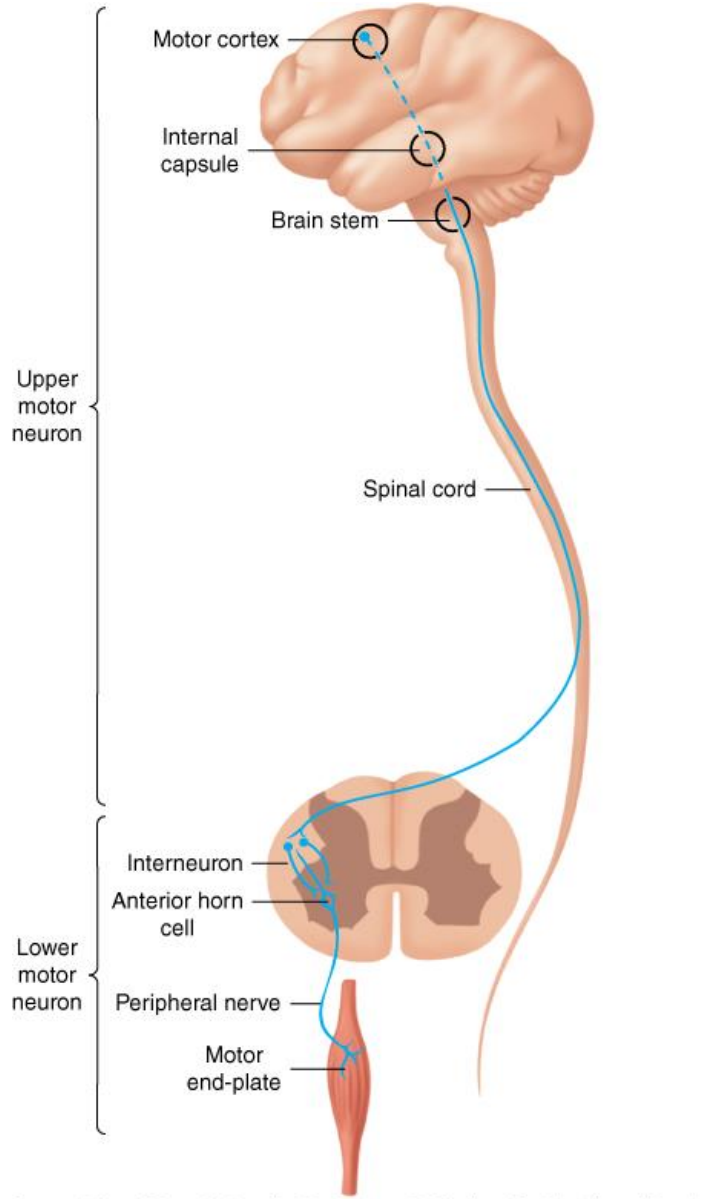
Lancet Neurol 2016; 15: 391–404

Which Antibodies to Test For?

"I don't want to miss anything!"

Anti-Neural Antibodies Can Target Any Location in the Nervous System

- Brain
- Spinal Cord
- Peripheral Nerve
- Neuromuscular Junction
- Muscle



Source: McKean S, Ross JJ, Dressler DD, Brotman DJ, Ginsberg JS: *Principles and Practice of Hospital Medicine*: www.accessmedicine.com

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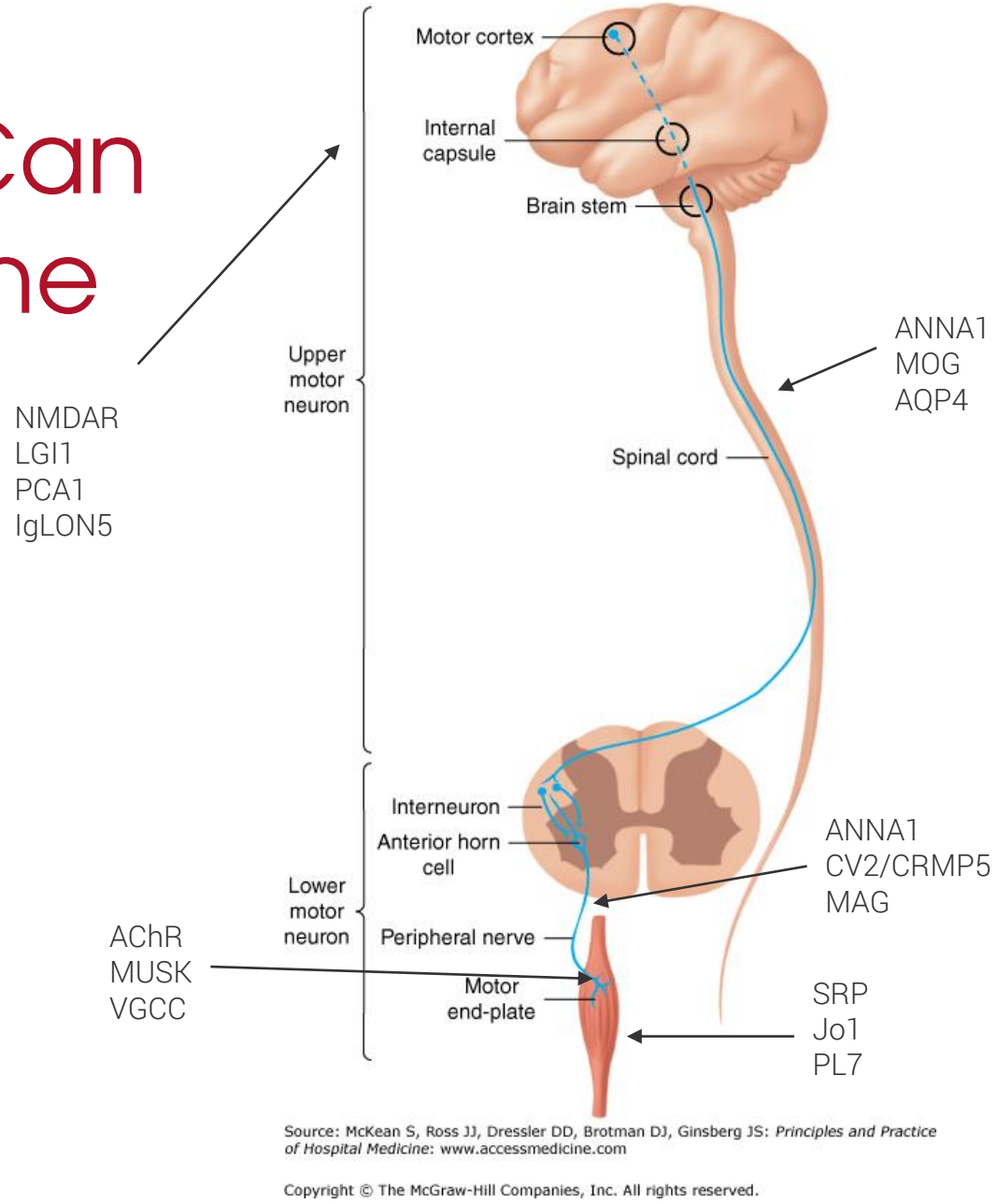

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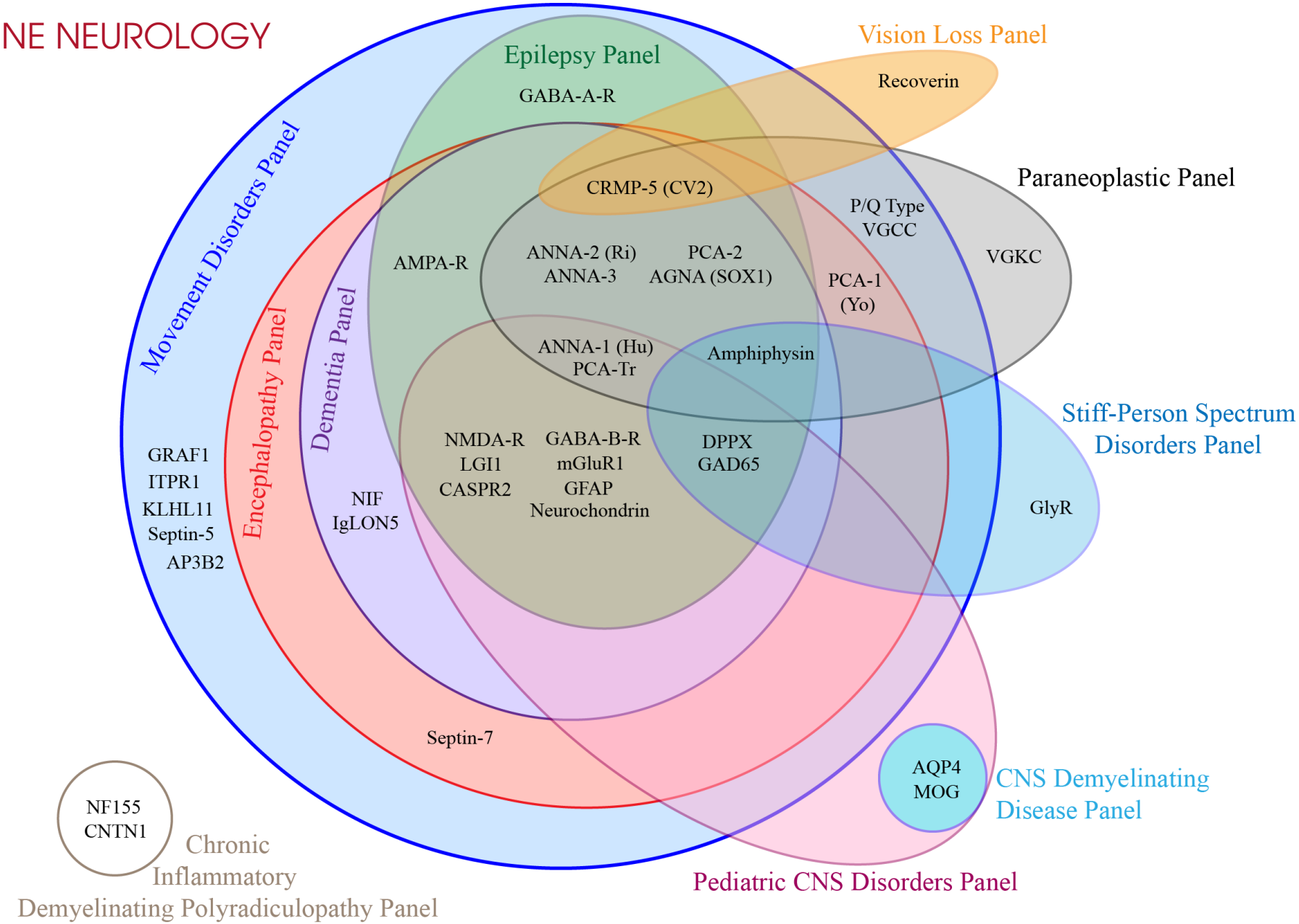
Clinical Cases

- 63M with one month of LEFT-sided movement involving face and arm, occurring 30-40 times a day, now with short term memory loss
- 36F with 2 months of RIGHT foot clumsiness and progressive stiffness, now with severe muscle spasms and stiffness of the spine, and exaggerated startle response
- 48M with several months of double vision late in the day and proximal weakness which worsens with activity and improves with rest
- 19F with 3 days of progressive bilateral vision loss (20/200 OS and count fingers OD), CSF with 8 WBC, OCB negative
- 79F with 2 years of gradually progressive short term memory loss

UPDATES IN AUTOIMMUNE NEUROLOGY



Avoiding Misdiagnosis

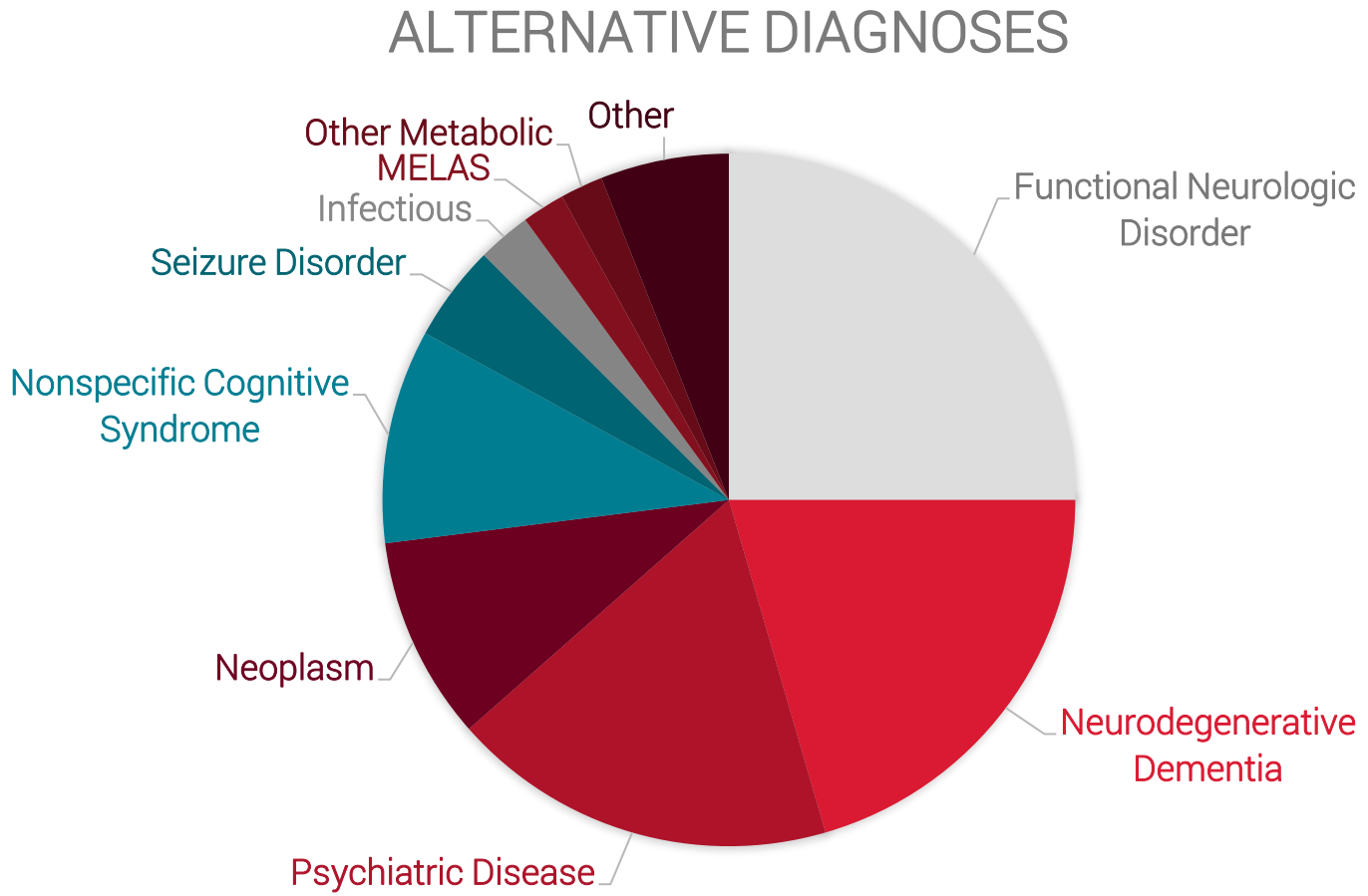
Is this an immune-mediated neurologic disease?

Missed Diagnosis or Misdiagnosis?

Misdiagnosis of Autoimmune Encephalitis

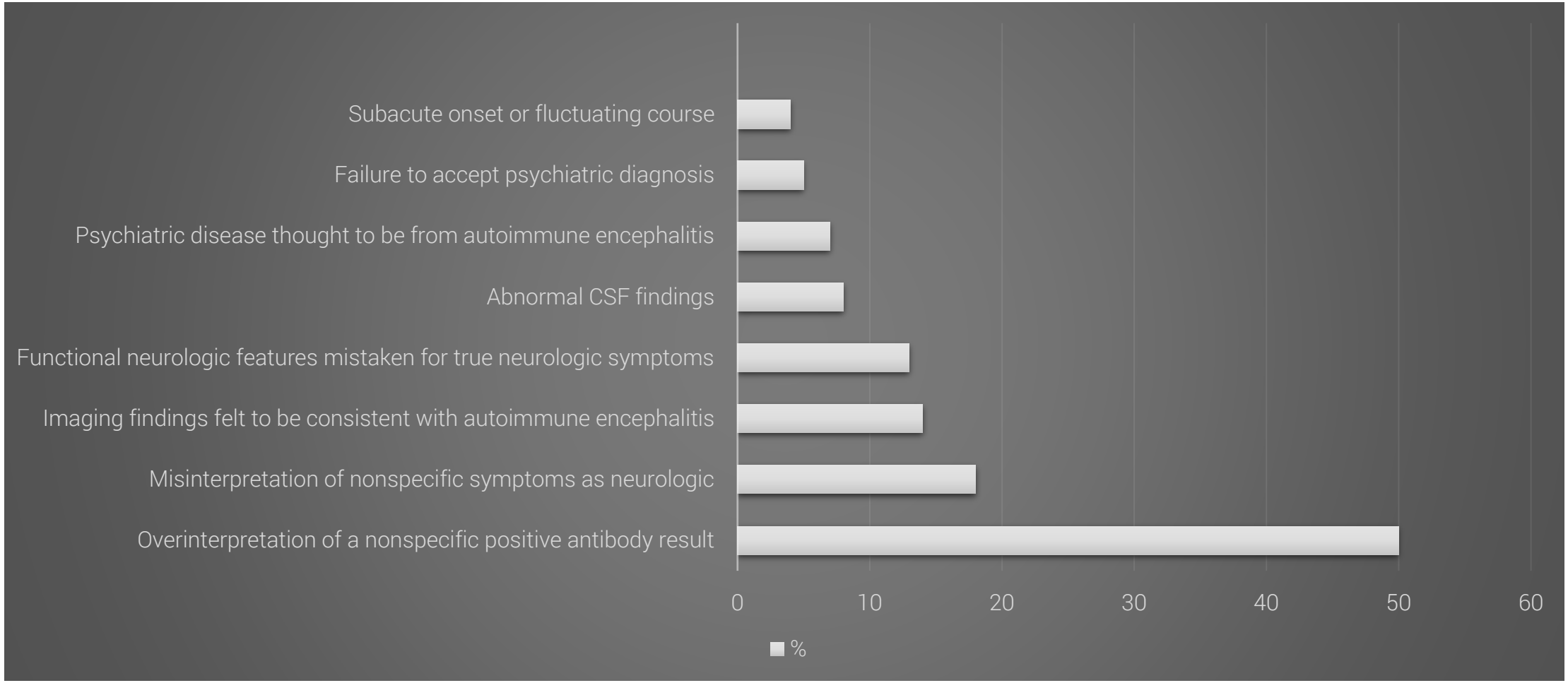
Criteria for Possible AE

- Subacute onset (rapid progression of <3 months) of short term memory loss, altered mental status, or psychiatric symptoms
- At least one of the following
  - New focal CNS findings
  - Seizures not explained by a previously known seizure disorder
  - CSF pleocytosis
  - MRI brain features of encephalitis



JAMA Neurol. 2023;80(1):30-39. doi:10.1001/jamaneurol.2022.4251

Reasons for Misdiagnosis



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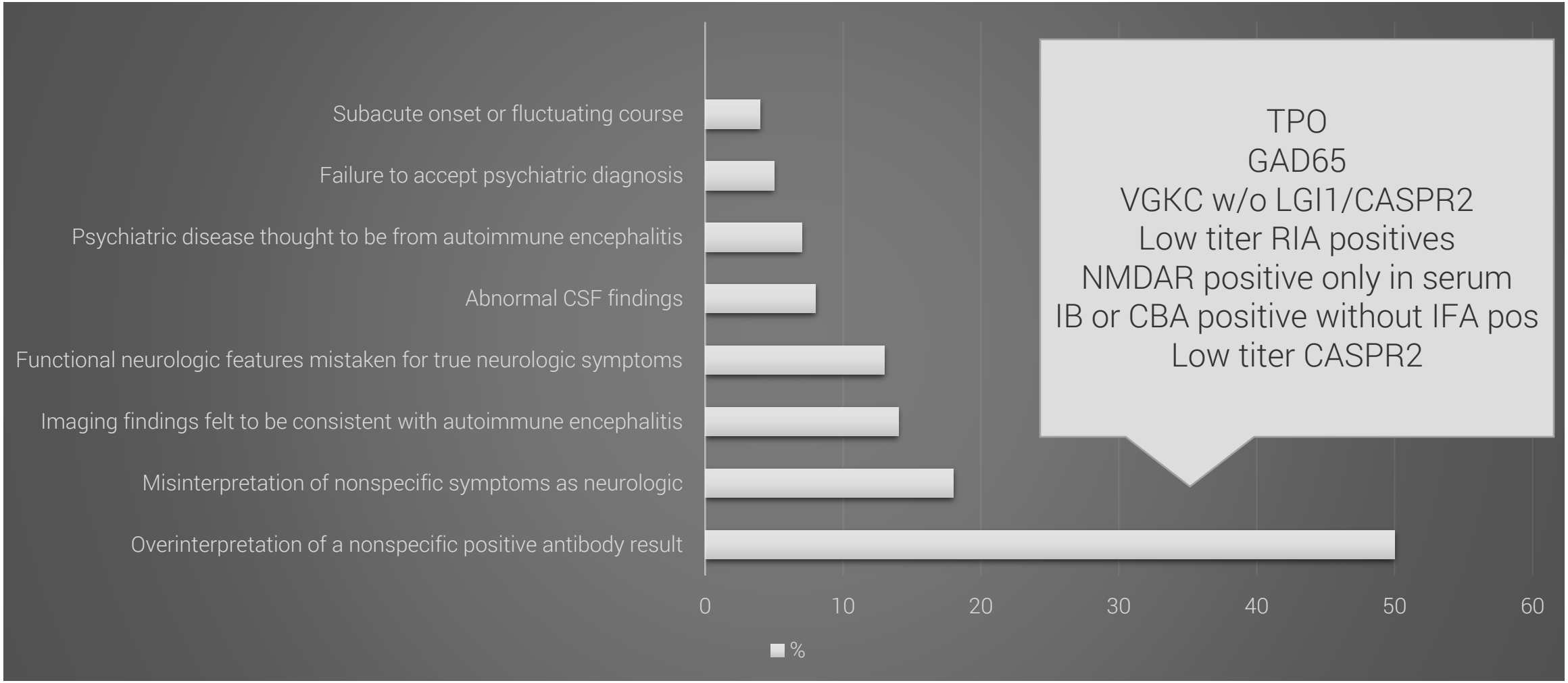

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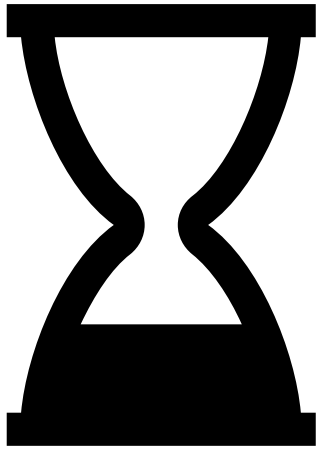
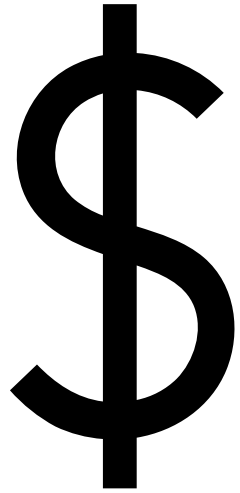
Consequence of Misdiagnosis





Review

Antineural antibody targets and clinical laboratory testing of these antibodies

When it is appropriate to test for antineural antibodies

Which antibodies to test for and how to choose

Avoiding Misdiagnosis

ARUP is a nonprofit enterprise of the University of Utah and its Department of Pathology.