

LYME DISEASE: MORE THAN A TITER

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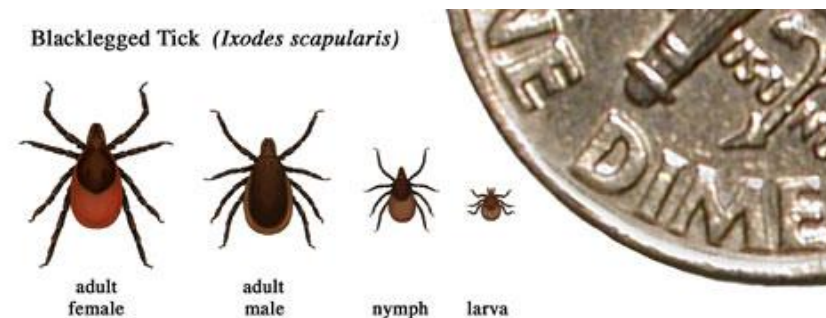
OBJECTIVES

- Discuss the etiology and pathophysiology of Lyme Disease
- Discuss serologic testing and titer utilization associated with Lyme Disease
- Apply evidence-based practice guidelines to a clinical case



WHAT IS LYME DISEASE

- Caused by the bacterium *Borrelia burgdorferi*
- Vector-borne disease
 - *Ixodes scapularis*
- Incidence 39.5 per 100,000
- Stages
 - Early localized
 - Disseminated





2001

2001



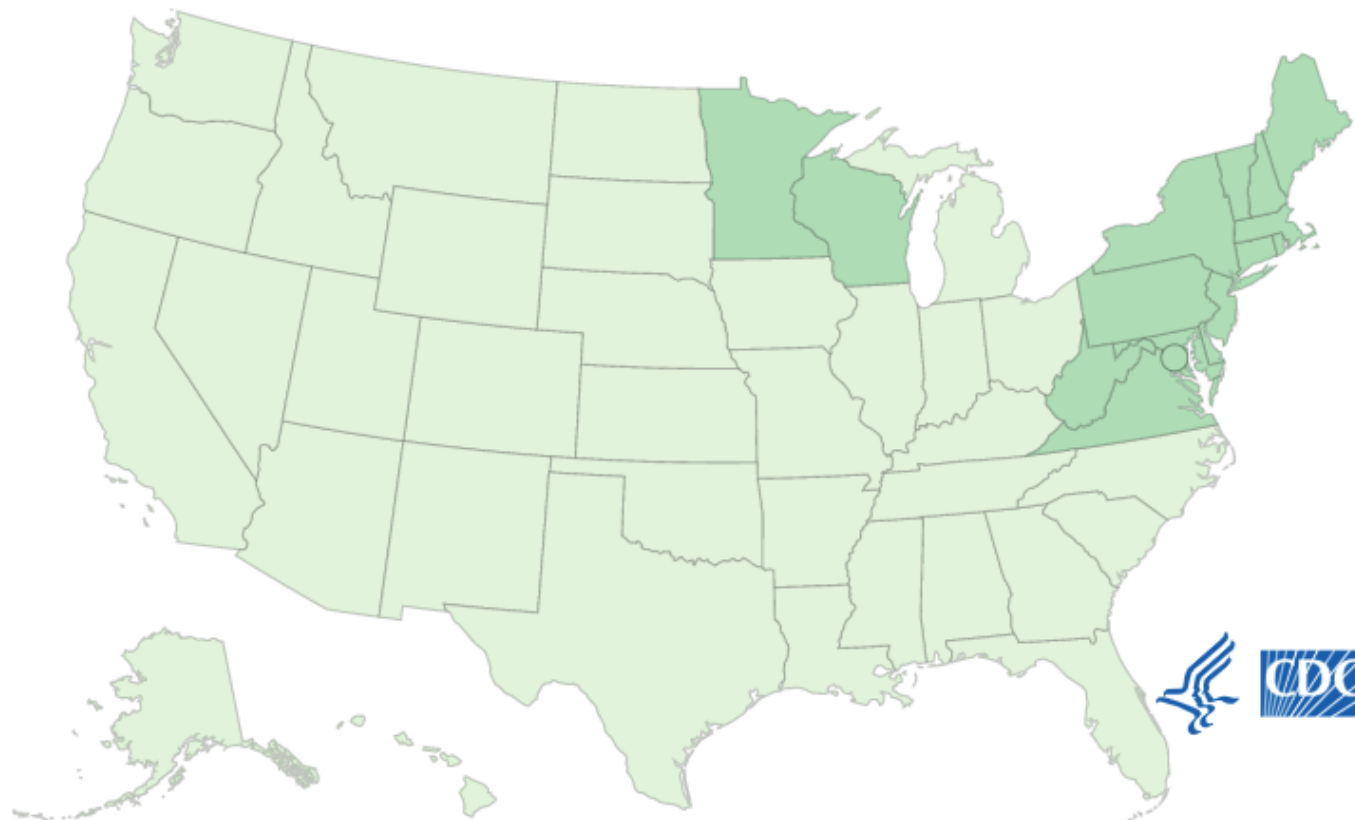
2020



Pause



Map of Lyme disease incidence* categories – United States 2019



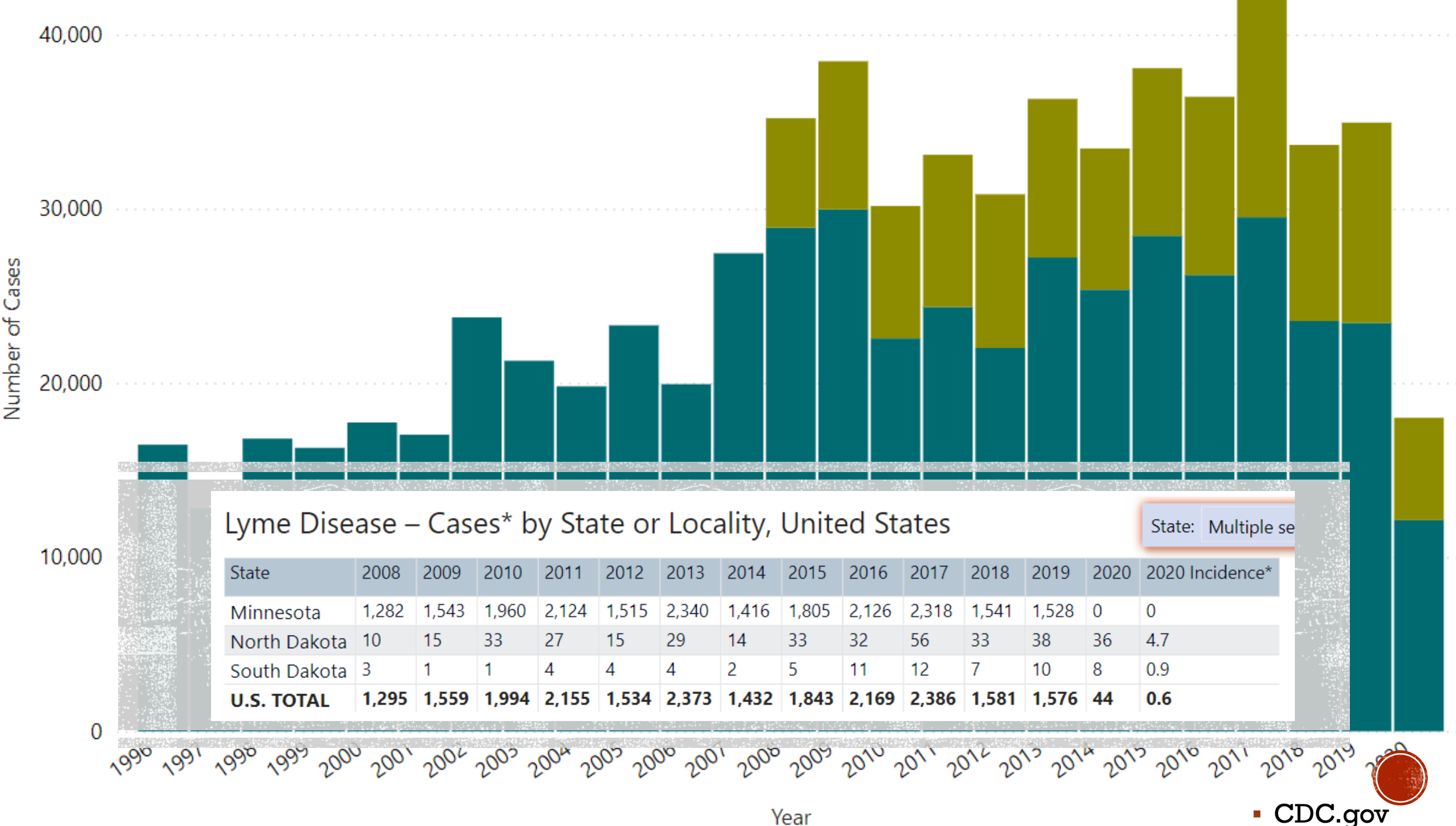
Legend

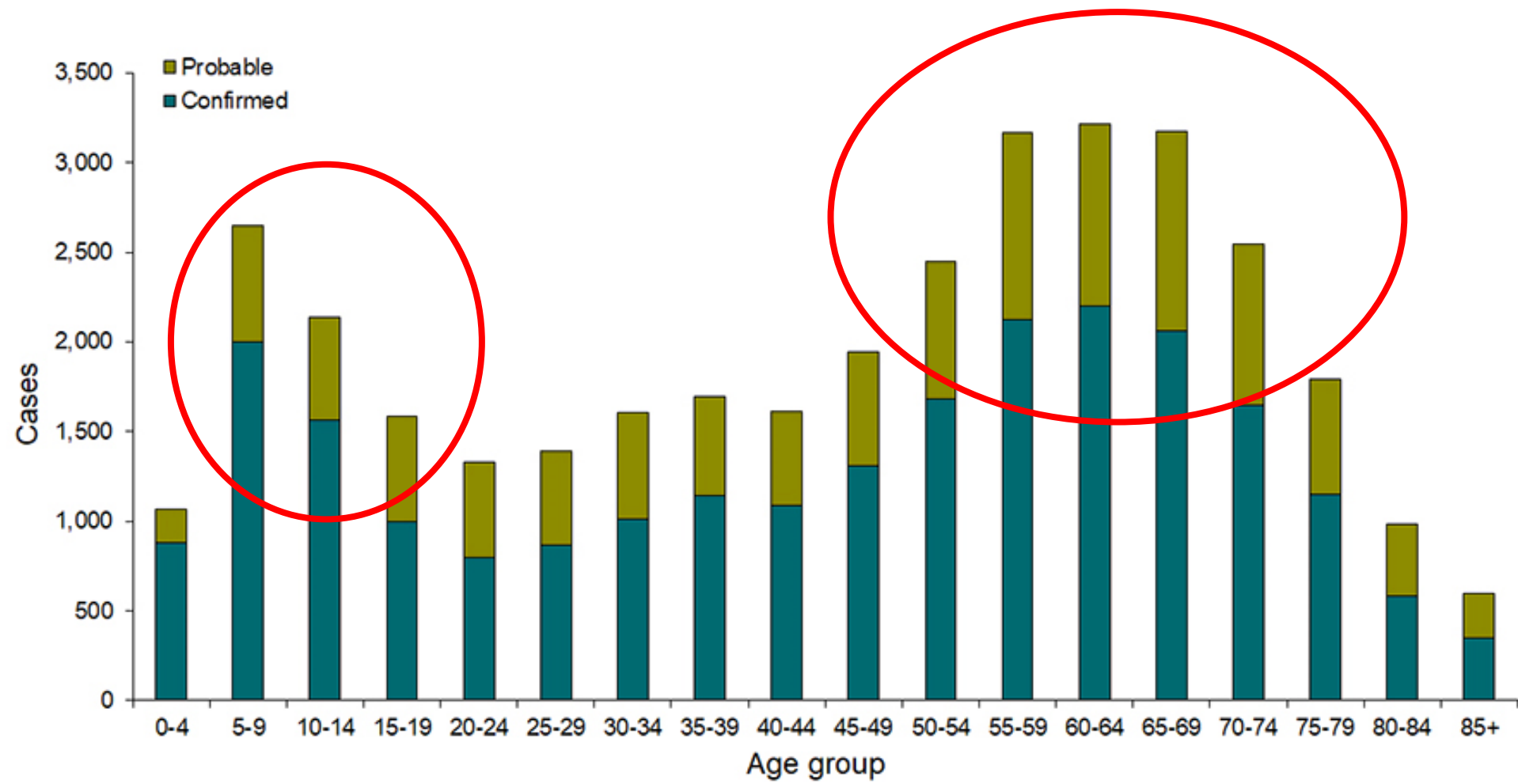
Incidence Category

● Low
incidence

● High
incidence

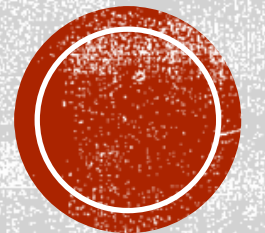








EARLY LOCALIZED SIGNS AND SYMPTOMS (3 TO 30 DAYS AFTER TICK BITE)



TREATMENT:

ERYTHEMA MIGRANS RASH

Age Category	Drug	Dosage	Maximum	Duration, Days*
Adults	Doxycycline OR	100 mg, twice per day orally	N/A	10-14
	Amoxicillin OR	500 mg, three times per day orally	N/A	14
	Cefuroxime	500 mg, twice per day orally	N/A	14
Children	Doxycycline OR	4.4 mg/kg per day orally, divided into 2 doses	100 mg per dose	10-14
	Amoxicillin OR	50 mg/kg per day orally, divided into 3 doses	500 mg per dose	14
	Cefuroxime	30 mg/kg per day orally, divided into 2 doses	500 mg per dose	14



EARLY DISSEMINATED SIGNS AND SYMPTOMS (DAYS TO MONTHS AFTER TICK BITE)

■ Non-Specific

- Patients also develop a musculoskeletal, flu-like syndrome, consisting of malaise, fatigue, chills, fever, headache, stiff neck, myalgias, and arthralgias (particularly in the knees) that may last for weeks to months (late stages).

■ Neurologic

- Inflammation of the brain and spinal cord
- Facial palsy
- Nerve pain
- Shooting pains, numbness, or tingling in the hands or feet



LYME ARTHRITIS

- Spirochete enters joint tissue and causes inflammation
- Swelling of the knee joint most common
- 4-week course of oral antibiotics
- risk of developing permanent joint damage



Table 1. Oral antibiotic regimens for Lyme arthritis.*

Age Category	Drug	Dosage	Maximum	Duration (days)	References
Adults	Doxycycline	100 mg, twice per day orally	N/A	28	1
	Amoxicillin	500 mg, three times per day orally	N/A	28	1
	Cefuroxime	500 mg, twice per day orally	N/A	28	1
Children ≥8 years old	Doxycycline	4.4 mg/kg per day orally, divided into 2 doses	100 mg per dose	28	1,4
	Amoxicillin	50 mg/kg per day orally, divided into 3 doses	500 mg per dose	28	1,4
	Cefuroxime	30 mg/kg per day orally, divided into 2 doses	500 mg per dose	28	4
Children <8 years old	Amoxicillin	50 mg/kg per day orally, divided into 3 doses	500 mg per dose	28	4
	Cefuroxime	30 mg/kg per day orally, divided into 2 doses	500 mg per dose	28	4

* For patients with an initial episode of Lyme arthritis, a full course of oral antibiotics are recommended. For patients with improving but persistent symptoms after an initial course of oral antibiotics, a second course of the same oral antibiotic or observation alone can be considered.

Table 2. Parenteral antibiotic regimens for Lyme arthritis.†

Age Category	Drug	Dosage	Maximum	Duration (days)	References
Adults	Ceftriaxone*	2 grams intravenously, once a day	N/A	14 – 28	2,3
Children	Ceftriaxone*	50-75 mg/kg intravenously, once a day	2 grams per day	14 – 28	2,3

†Intravenous ceftriaxone is the preferred regimen for the second course of antibiotics for patients without any response after the initial course of antibiotics.



NEUROLOGIC LYME DISEASE

- **Cranial nerve involvement:** When the cranial nerves are affected, facial palsy (droop) can occur on one or both sides of the face.
- **Peripheral nerve involvement:** When the peripheral nerves are affected, patients can develop radiculoneuropathy which can cause numbness, tingling, “shooting” pain, or weakness in the arms or legs.
- **Central nervous system involvement:** When the central nervous system is affected, Lyme meningitis can cause fever, headache, sensitivity to light, and stiff neck.



Antibiotic Treatment of Neurologic Lyme Disease

These regimens may need to be adjusted depending on a person's age, medical history, underlying health conditions, pregnancy status, or allergies. Consult an infectious disease specialist regarding individual patient treatment decisions.

Table 1. Facial palsy

Age Category	Drug	Dosage	Maximum	Duration (Days)	References
Adults	Doxycycline	100 mg, twice per day orally	N/A	14-21	2,3
Children (any age)	Doxycycline	4.4 mg/kg per day orally, divided into 2 doses	100 mg per dose	14-21	2,3

Table 2. Lyme meningitis or radiculoneuritis

Age Category	Drug	Dosage	Maximum	Duration (Days)	References
Adults	Doxycycline OR	200 mg per day orally, divided into 1 or 2 doses	N/A	14-21	3,5
	Ceftriaxone*	2 grams intravenously, once a day	N/A	14-21	5
Children (any age)	Doxycycline OR	4.4 mg/kg per day orally, divided into 1 or 2 doses	100 mg per dose	14-21	1,3
	Ceftriaxone*	50-75 mg/kg intravenously once a day	2 g per day	14-21	4

* Oral therapy can be substituted when the patient is stabilized or discharged to complete the course.



LATE DISSEMINATED SIGNS AND SYMPTOMS (DAYS TO MONTHS AFTER TICK BITE)

- Myocardial Abnormalities (carditis)
 - 0.3-10% of Lyme's disease cases
 - Heart palpitations or an irregular heartbeat
 - Spirochetes enter the tissues of the heart
 - Episodes of dizziness or shortness of breath
 - Treated with oral or intravenous (IV) antibiotics
 - Can be fatal
 - Between 1985 and 2019, 11 cases of fatal Lyme carditis



DIAGNOSIS OF LYME DISEASE

- **Endemic areas with EM rash**
 - should be started on treatment without blood tests
- **Laboratory tests are important for establishing the diagnosis**
 - antibody detection tests most common



FDA CLEARED OR APPROVED TESTING

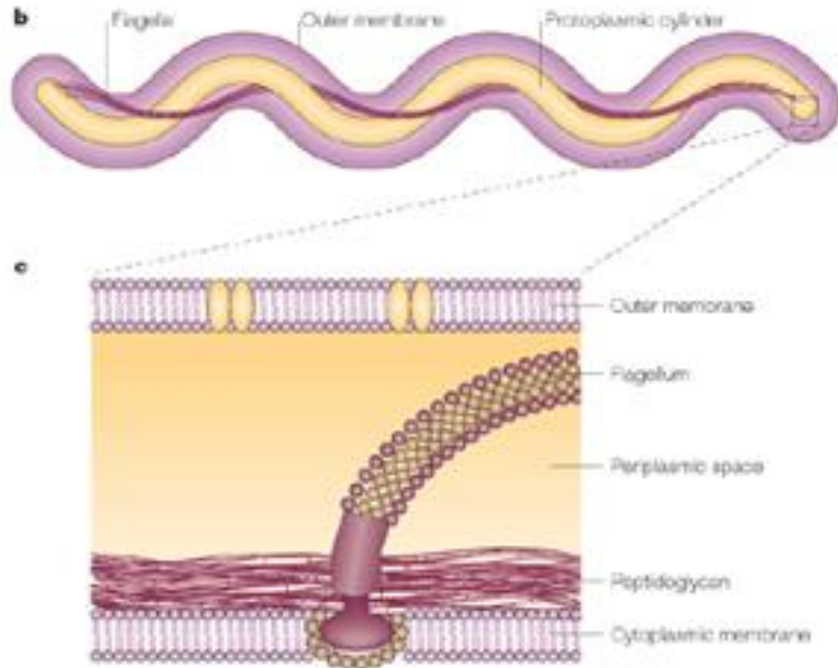
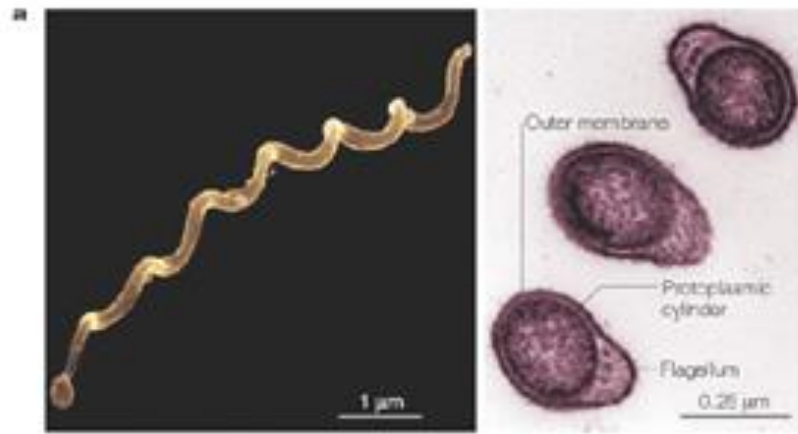
- CDC recommends a Two-step testing procedure
 1. Enzyme immunoassay (EIA) or Enzyme-linked immunosorbent assay (ELISA)
 2. Western immunoblot test is performed to confirm
- Guidelines from the Infectious Diseases Society of America, American Academy of Neurology, and American College of Rheumatology (IDSA/AAN/ACR)
 1. Performing acute-phase serum antibody testing in patients with 1 or more skin lesions
 2. Repeat testing can be performed on a convalescent-phase serum sample collected at least 2–3 weeks afterward
- ❖ Routine use of sequential serologic testing in individual patients with early Lyme disease should be **discouraged**
- ❖ Acute and convalescent-phase serologic testing has **no role** in Lyme disease



SEROLOGIC TESTING

- Test results do not DIRECTLY rule in or rule out Lyme disease
- Most frequently used test is the enzyme immunoassay (EIA) or enzyme-linked immunosorbent assay (ELISA)
 - Less common is the immunofluorescent assay (IFA)
- Principal limitation
 - high frequency of both false-negative results and false-positive results
- EM Rash mimics
- Confirmation with Western Blot recommended



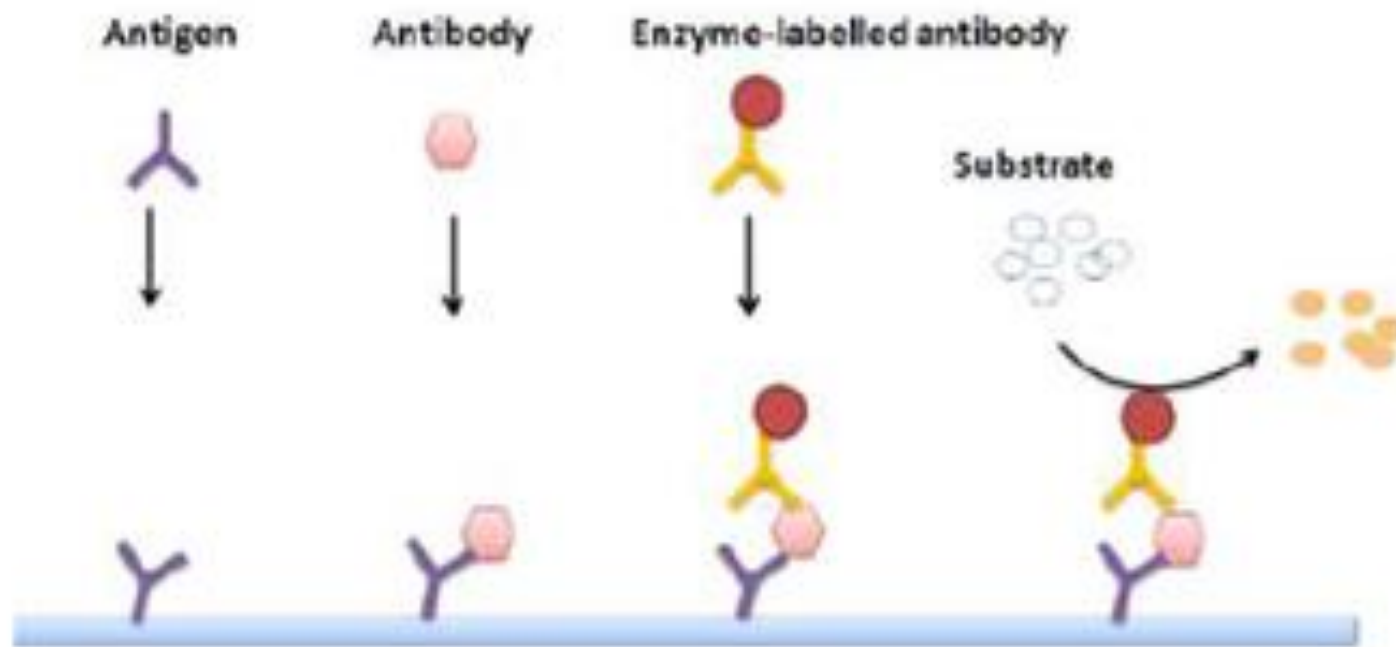


Nature Reviews | Microbiology

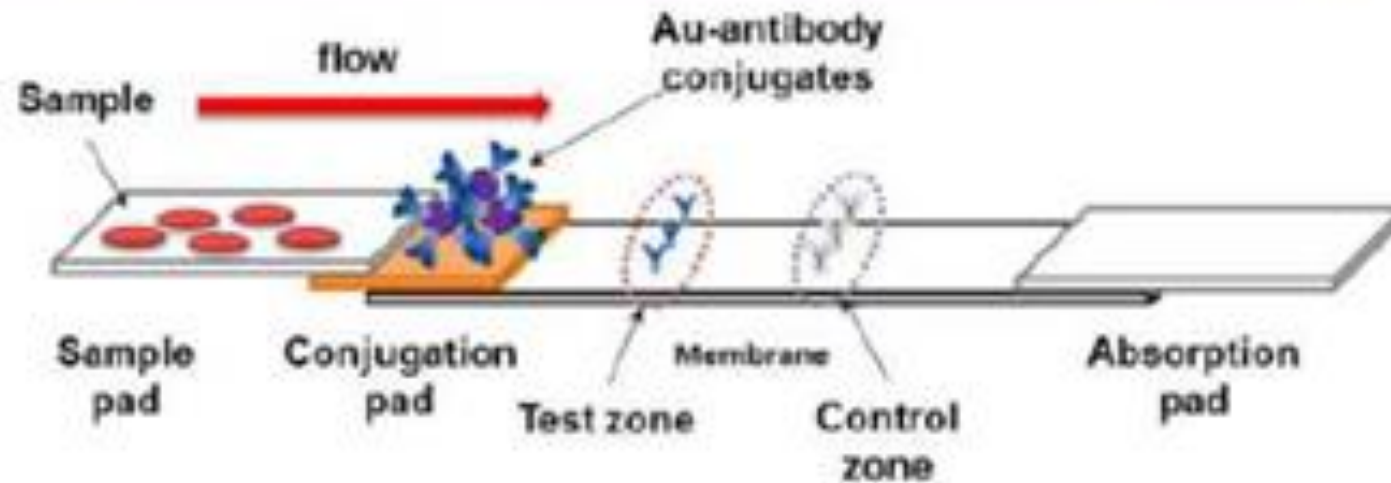
Rosa P, Tilly K, Stewart P. 2005. The burgeoning molecular genetics of the Lyme disease spirochaete. *Nature Reviews Microbiology* 3:129-143. The burgeoning molecular genetics of the Lyme disease spirochaete.

- **Flagellin (FlaB)** – illicit a strong IgG and IgM response, but it cross reacts with other bacteria, and antigens in neural tissue, synovium,, and myocardial tissue.
- **Flagellar outer sheath protein (FlaA)**
- **OspC** – expressed during tick feeding, may have a role in initial pathogenesis
- **Decorin binding protein A** – binds to host-collagen-associated proteoglycan decorin
- **OspA and OspB** – expressed later on during infection
- **Vmp-like sequence expressed (VisE) protein**





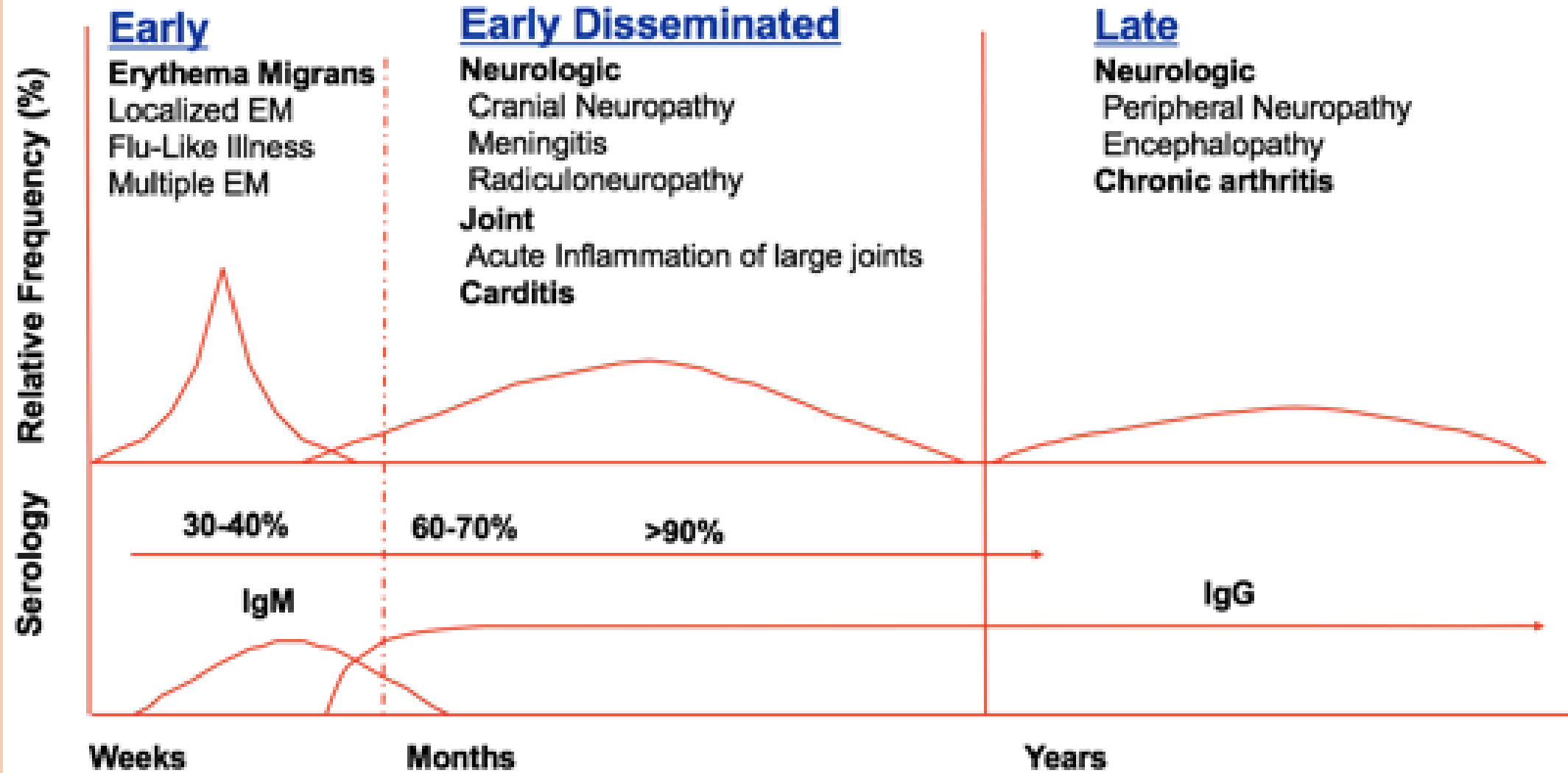
ELISA and CMIA



Lateral flow assay- you put the sample and it flows down the device. The conjugation pad acts like the antibody that is present on the wells of a normal ELISA and the Test zone works like the added antibody in an ELISA

This is basically an OTC pregnancy test





(Adapted from: [Bahr, D.W. 1998. Natural History of Lyme Disease. In: Bahr, D.W., and J. Evans \(Eds\), Lyme Disease. pp 35- 48.](#))

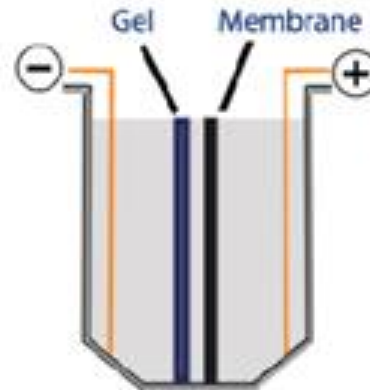


WESTERN BLOT TESTING

Put protein into the page, and then use electricity to separate them by weight



1. Load and separate protein samples on SDS-PAGE gel.



2. Electrophoretically transfer fractionated proteins onto PVDF or nitrocellulose membrane.

Once separated, the gel gets put next to a membrane and said proteins get transferred into the membrane.

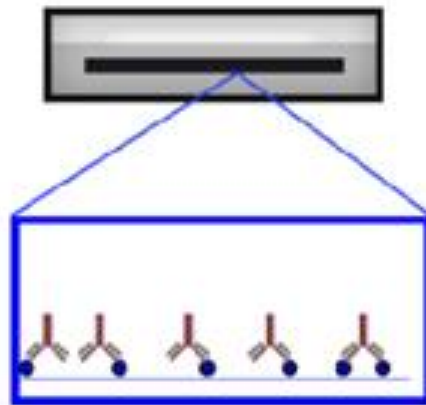
Container



3. Block the membrane with neutral protein (BSA or milk).

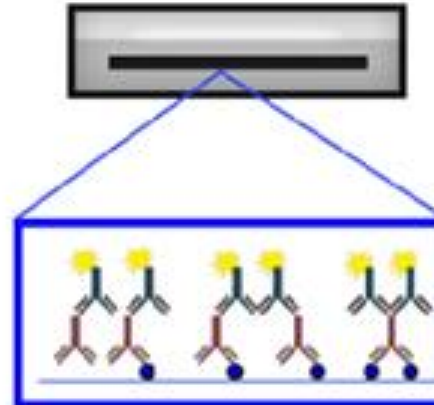
You can think of this as being a very convoluted ELISA with an additional step.

This is helpful because we can use the whole cell protein components from the organism (in this case, *Borrelia burgdorferi*) and allows you to see which antibody reacts to said target proteins.



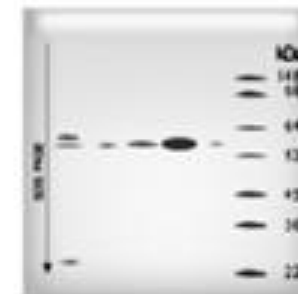
4. Incubate the membrane with primary antibody specific to target protein. Wash.

Once in the membrane, then we can use antibodies against a protein of interest.



5. Incubate the membrane with HRP-labeled secondary antibody specific to primary antibody. Wash.

Then we used another antibody to mark said protein of

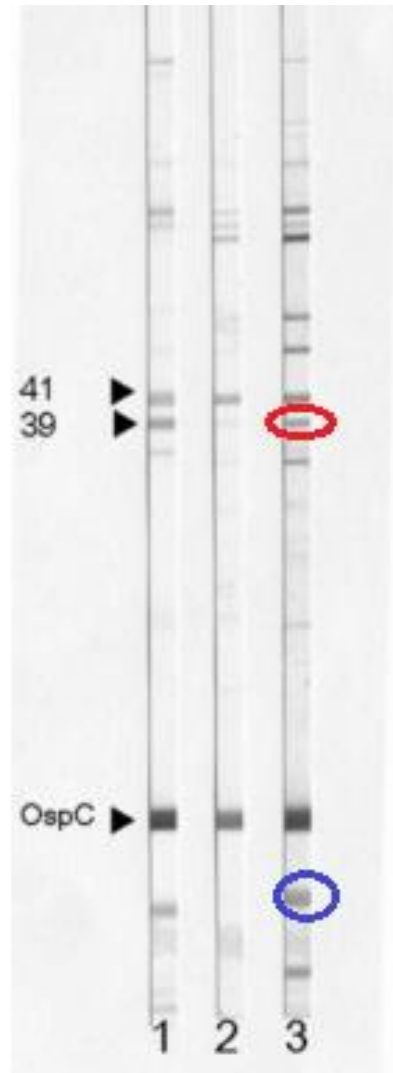


6. Incubate the blot with chemiluminescent HRP substrate and expose to film.

WESTERN BLOT TESTING

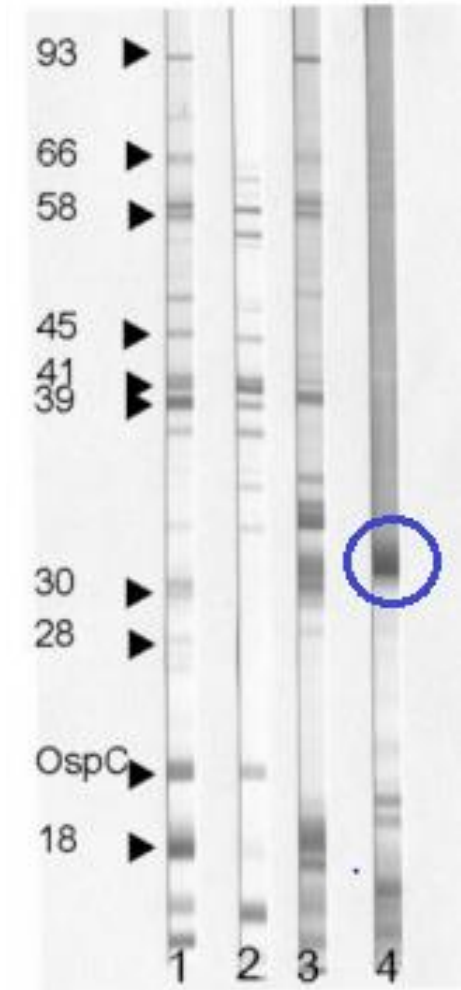
separate proteins
based on weight

*Expected

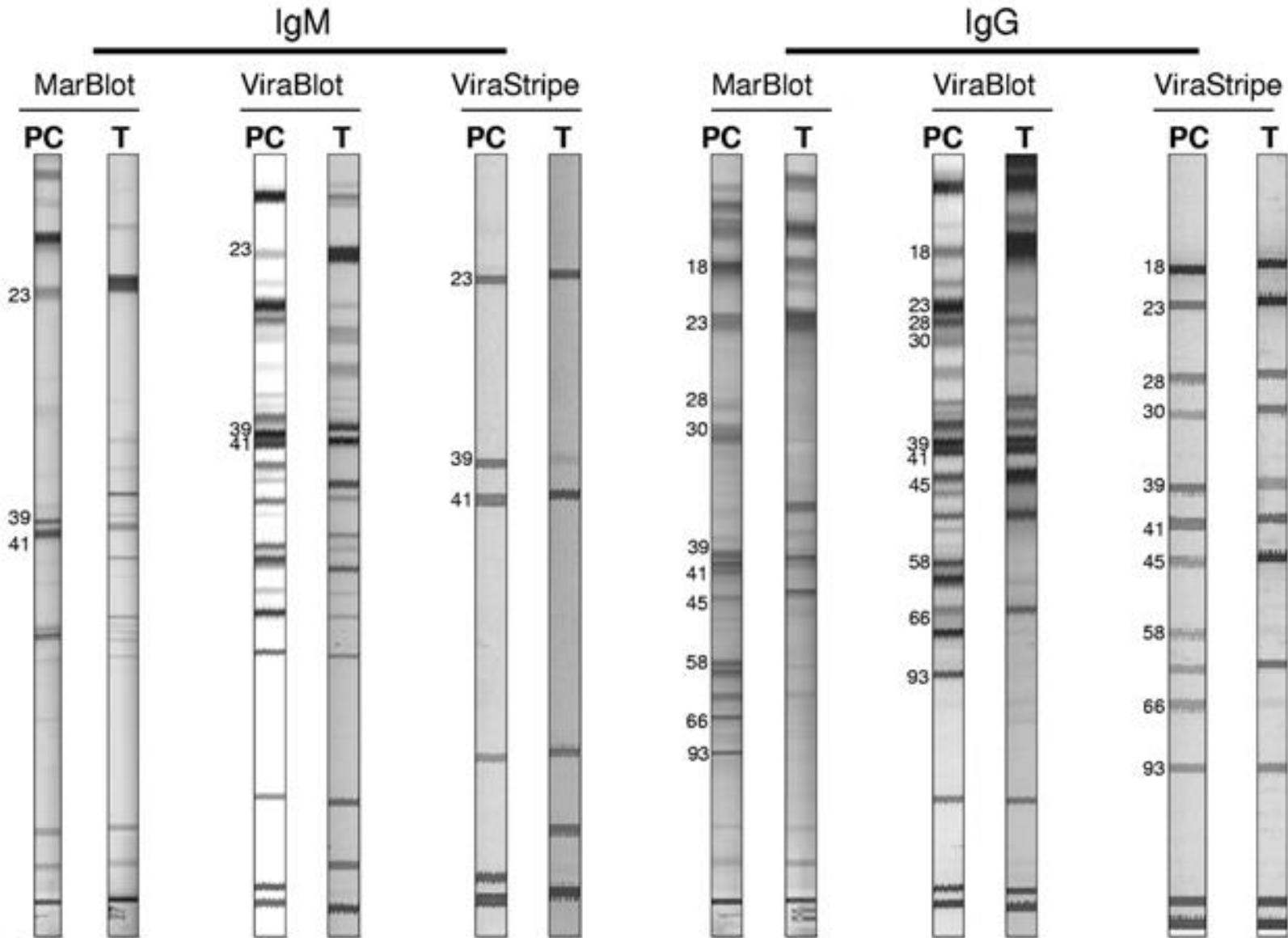


antibody binds into a
different antigen

*unexpected
*cross-reacting



HOW TO INTERPRET WESTERN BLOT



bands OpsC
41 or 39kDa

93, 66, 58, 45,
39, 30, 28, 21,
OpsC, or 18kDa



C6 PEPTIDE TESTING

In Control Subjects

Sample Size and Reference	Specificity (%)		
	Standard 2-tier	2-EIA	C6 EIA Alone
N = 1300; Ref [10]	99.5 (98.9–99.8)	99.5 (98.9–99.8) <i>P</i> = 1.0	98.4 (97.5–99.0) <i>P</i> = .01
N = 2208; Ref [9]	99.5 (99.1–99.7)	99.5 (99.1–99.8) <i>P</i> = 1.0	98.9 (98.4–99.3) <i>P</i> = .05
N = 100; Ref [36]	100 (95.6–100)	100 (95.6–100) <i>P</i> = 1.0	100 (95.6–100) <i>P</i> = 1.0
N = 347; Ref [27]	98.3 (96.2–99.3)	98.3 (96.2–99.3) <i>P</i> = 1.0	96.5 (94.0–98.1) <i>P</i> = .2

The numbers in parentheses represent the 95% confidence intervals surrounding the specificity values.

The *P* values pertain to the comparison with results obtained by standard two-tiered testing (whole-cell sonicate enzyme immunoassay (EIA) followed by immunoglobulin M and immunoglobulin G Western blots). The values provided for the 2-EIA protocol refer to results obtained using a Food and Drug Administration (FDA)-cleared, commercially available whole-cell sonicate EIA followed by an FDA-cleared, commercially available C6 EIA.

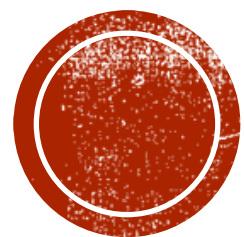
- **Next gen EIAs**
- measures IgG
- peptide from the sixth invariant region (C6)
- variable major protein-like sequence-expressed (VlsE) lipoprotein
- may be more sensitive in patients with EM
- may be effective in differentiating from Lyme disease
 - southern tick-associated rash illness (STARI)
 - confirming infection in patients who may have been infected in Europe



LABORATORY DEVELOPED TESTS FOR LYME DISEASE

- Clinical validity and safety have not been cleared or approved by the FDA
- Examples include:
 - Capture assays for antigens in urine
 - Immunofluorescence staining, or cell sorting of cell wall-deficient or cystic forms of *Borrelia burgdorferi*
 - Culture for *Borrelia burgdorferi*
 - Lymphocyte transformation tests
 - Quantitative CD57 lymphocyte assays
 - "Reverse Western blots"
 - IgM or IgG blot assays without a previous enzyme immunoassay
 - PCR testing





CASE STUDIES



CASE #1

- 3-year-old boy seen in urgent care for nausea, vomiting, painful joints, body aches, and fever.
- History
 - visiting his grand parents in Northern Minnesota
 - He played outside daily for hours



CASE #1

- Mom had removed a “baby tick” 7 days earlier
- He had a “rash” at the bite site



Day 1



Day 3



Day 5



Day 7

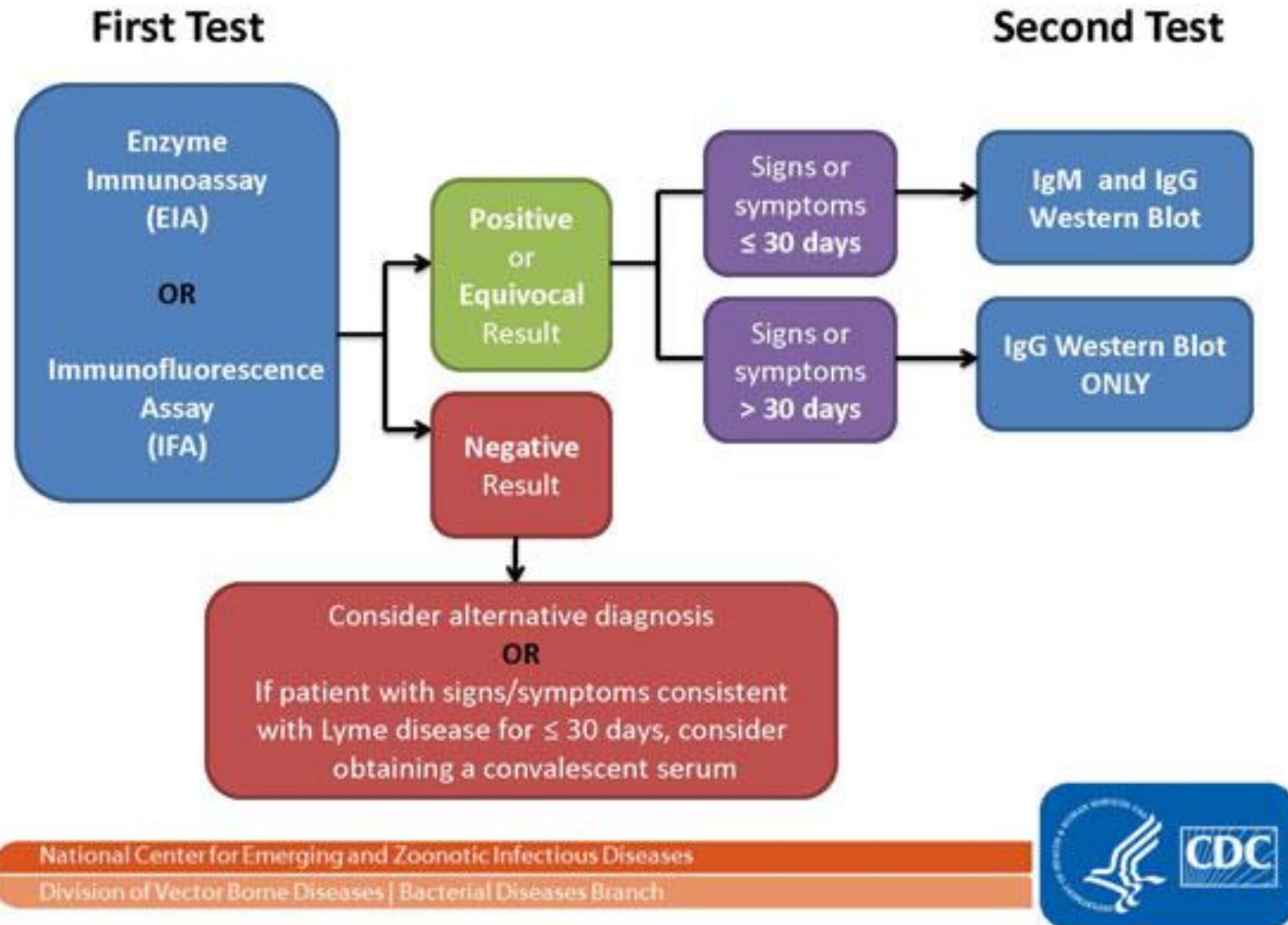


CASE #1

- Provider at urgent care wanted to start antibiotic treatment
 - 14-day regimen of antibiotics
- Wanted to order serologies for a Lyme Titer



Two-Tiered Testing for Lyme Disease



1 week of
antibiotics



CASE #2

- 47-year-old male presented to the ER for multiple episodes of “fainting” over the last couple of weeks.
- Normal, healthy appearing
- All routine lab work is normal

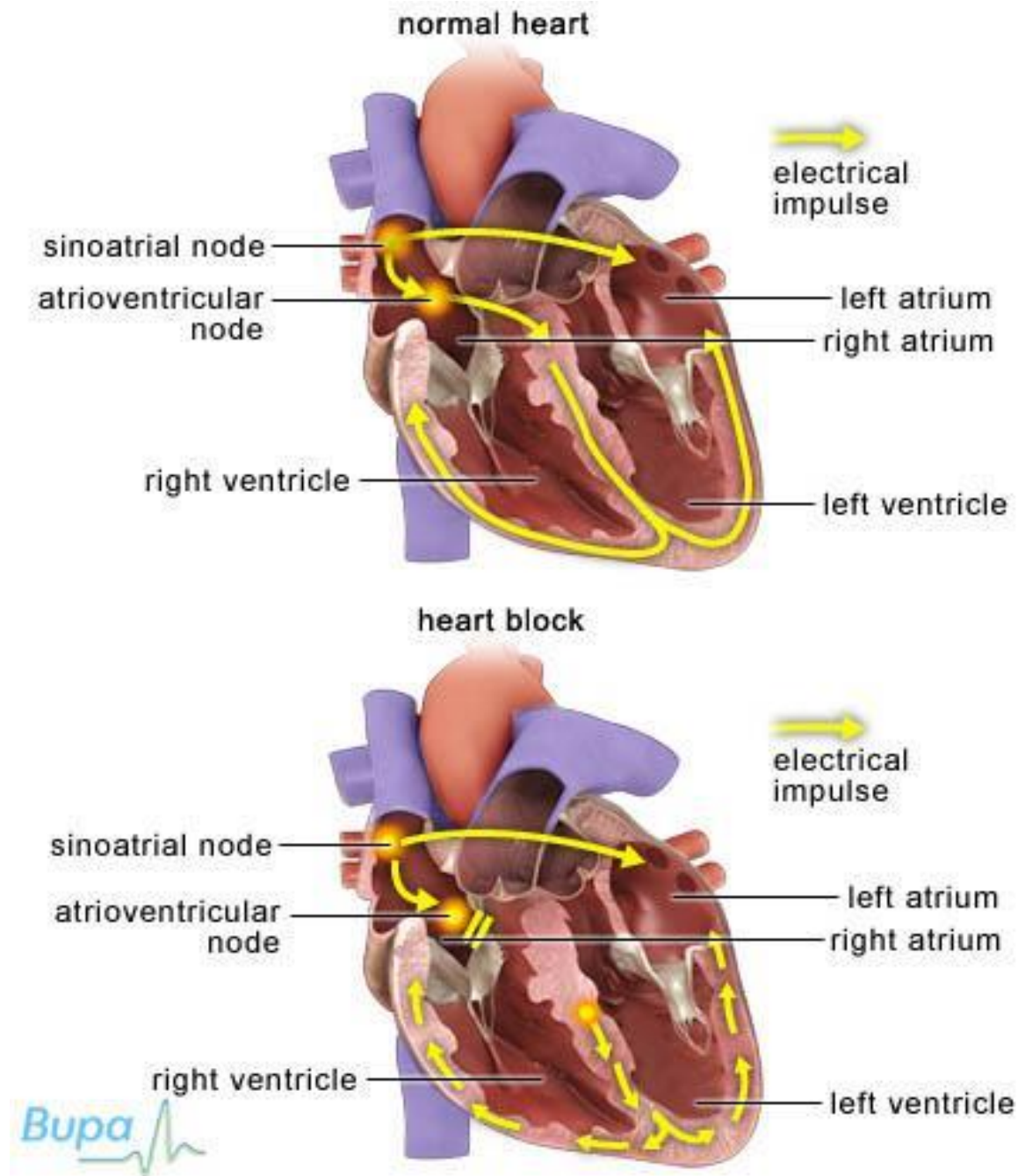


CASE #2

- Works as a Hunting and Camping Trail Guide in the Appalachian Mountains
- He has another “fainting” episode in the ER and physician realizes they are seizures.
 - Begins neurologic and cardiac work up
 - Has complete AV heart block on his EKG



LYME'S CARDITIS



- AV Block
 - 1st degree
 - 2nd degree
 - Mobitz 1 (Wenckebach)
 - Mobitz 2
 - 3rd degree
- Reduced EF
 - As low as 35%
- Cardiomegaly

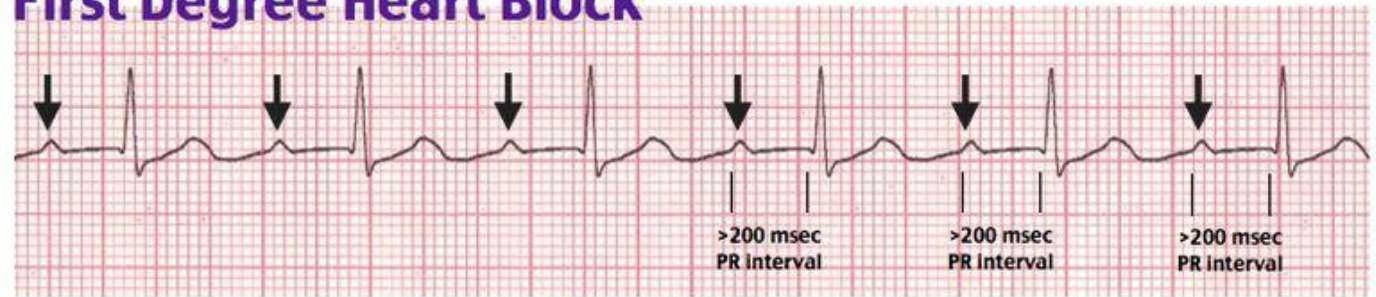


AV BLOCK

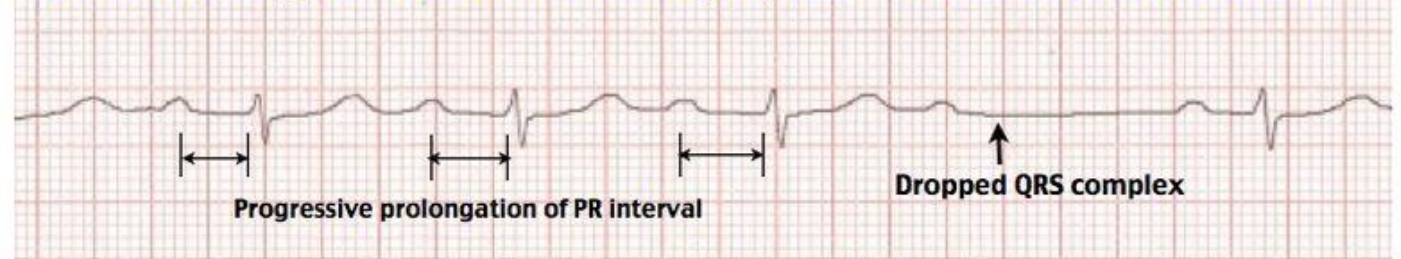
Normal sinus rhythm



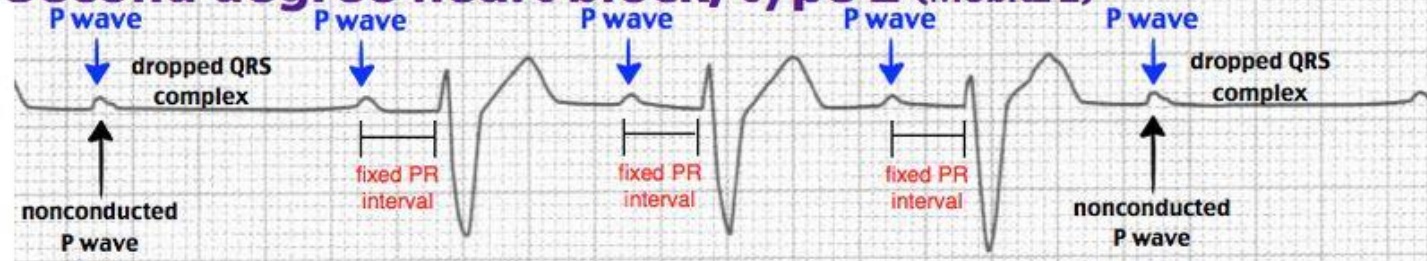
First Degree Heart Block



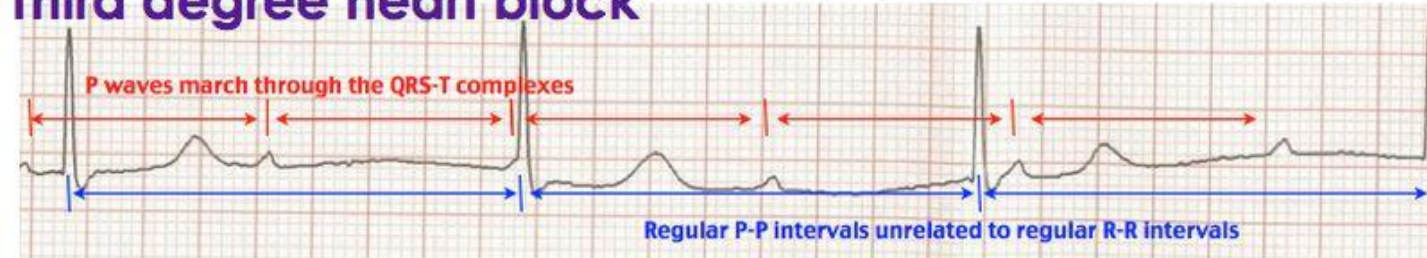
Second degree heart block, type I (Wenckebach/Mobitz I)



Second degree heart block, type 2 (Mobitz 2)



Third degree heart block




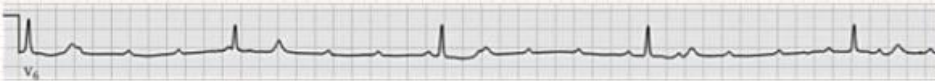


Second-degree type II

Heart Blocks



AV INTERVENTION

Degree	Findings	Prognosis/Rx
1 st degree	<p>Every P wave is followed by QRS complex, but PR interval is prolonged.</p> 	Usually benign, except under highly unusual circumstances.
Type I 2 nd degree (Wenckebach)	<p>PR interval progressively lengthens until a P wave is blocked.</p> 	<p>Usually benign, but may progress to 3rd degree block.</p> <p>Pacemaker needed if etiology is not reversible and patient is symptomatic.</p>
Type II 2 nd degree	<p>PR interval is constant with some P waves intermittently blocked.</p> 	Pacemaker is indicated if etiology is not quickly reversible.
3 rd degree	<p>Complete dissociation of P wave and QRS complex.</p> 	<p>Pacemaker is necessary.</p> <p>Urgency of pacemaker placement depends upon symptoms, width of QRS complex and the ventricular escape rate.</p>



CASE #2

- He had been sick with flu-like symptoms a few months back that had resolved
- Had a tick, but no rash that he noticed
- Patient received a temporary pacemaker and antibiotics



SUSPICIOUS INDEX IN LYME CARDITIS (SILC) SCORE

Variable	Value
Age < 50 years	1
Male	1
Outdoor activity/endemic area	1
Constitutional symptoms*	2
Tick bite	3
Erythema migrans rash	4

* fever, malaise, arthralgia, and dyspnea

The total score indicates low (0-2), intermediate (3-6), or high (7-12) suspicion of Lyme carditis.



TREATMENT

TABLE 2. Antibiotic treatment for Lyme carditis.^{1,4}

Presentation	Line of treatment	Antibiotic	Duration of therapy
1st degree AVB*	1st	Doxycycline 100 mg PO BID	14–21 days
	2nd	Amoxicillin 500 mg PO TID	
	3rd	Cefuroxime 500 mg PO TID	
	4th	Erythromycin 250 mg PO QID	
2nd or 3rd degree AVB	1st	Ceftriaxone 2 g IV q24h	10–14 days, [†] followed by oral antibiotics [‡]
	2nd	Cefotaxime 2 g IV q8h	
	3rd	Penicillin G 5 million U IV q6h	

*AVB: atrioventricular block

[†]Until 1:1 atrioventricular conduction is restored

[‡]Combined total course of IV and oral antibiotics should be 14–21 days



FOLLOW-UP

Case #1

- Constitutional symptoms resolved within days of starting antibiotics
- Joint/nerve pain resolved in about 6 weeks
- No further follow-up



FOLLOW-UP

Case #2

- Resolution of AV block
 - Complete: about 7-10 days
 - Lesser/Minor: within 6 weeks
- AV Conduction restored stepwise fashion
 - Resolution = temporary ventricular pacemaker (TVP)
- Regular EKG checks at 4-6 weeks initially, with taper



PROGNOSIS

- Some develop chronic conditions
 - Musculoskeletal, cognitive disturbances, fatigue, difficulty sleeping
- Post-treatment Lyme Disease Syndrome (PTLDS)

Case #1

- Short term is good
- Long term is unknown

Case #2

- Good with appropriate therapy
- Few reports of permanent AV block
- Although rare, cases of fatal Lyme disease secondary to carditis have been reported
 - 11 cases 1985-2019



**WHY THIS TOPIC IF
NORTH DAKOTA HAS LOW
PREVALENCE?**

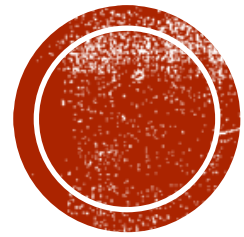


Then



Now





QUESTIONS?

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