THE UNIVERSITY OF KANSAS HEALTH SYSTEM

A Prescription for Hemolysis:

When You Can Read The Handwriting, And When You Can't

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Objectives

- Distinguish the clinical features of a potential case of drug-induced immune hemolytic anemia (DIIHA)
- Identify laboratory tests and values that will aid in the diagnosis of DIIHA
- Describe the theoretical mechanisms that lead to hemolysis in DIIHA
- List drugs that have been implicated in cases of DIIHA

Case Study 1

- 63 years old male
- Paraplegia and neurogenic bladder
- Rehabbing at skilled nursing facility, but reports having to self catheterize every 12 hours (urology recommended 3-4 hour)
- Presenting with altered mental status
  - Determined to be caused by toxic metabolic encephalopathy due to a complicated UTI
- Prescribed piperacillin/tazobactam (ZOSYN), 3.375g every 6 hours

Case Study 1: Day 0

- On admission, laboratory results unremarkable aside from urinalysis:
  - HGB: 13.9g/dL
  - PLT: 226K/µL
  - WBC: 7.9K/µL
  - T. Bili: 0.7mg/dL
- Urinalysis: Yellow
  - Nitrite: POS
  - Leukocytes: 3+
  - Blood: NEG
  - Bacteria: Many
  - WBC: 10 – 20/HPF
  - RBC: 0 – 2/HPF

Case Study 1: Days 1 – 6

- Patient being evaluated and treated for UTI
  - HGB trending lower
  - WBC trending higher
  - BUN trending higher
  - Day 0 blood culture NEG after 5 days

Day	0	2	4	6
HGB (g/dL)	13.9	13.0	11.6	8.8
WBC (K/µL)	7.9	10.6	14.1	23.7
BUN (mg/dL)	20	19	27	37

Case Study 1: Day 6 Type and Crossmatch, DAT

- Due to drop in HGB, a type and crossmatch (XM) is ordered

Screening Cells	Solid Phase	Gel	LISS AHG
SC1 (R1R1)	1+	2+	
SC2 (R2R2)	0	1+	
SC3 (rr)	3+		
AC		3+	1+

- DAT Results

Polyspecific	IgG Specific	Complement	Saline
2+	2+	(+)	0/0

Case Study 1: LISS Panel

	D	C	c	E	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	Lu^a	Lu^b	AHG
1	+	+	0	+	+	0	+	0	+	+	+	0	+	+	0	+	+	0	+	(+)
2	+	+	0	0	+	0	+	+	0	0	+	+	0	+	+	+	+	0	+	(+)
3	+	0	+	+	0	0	+	+	0	+	+	0	+	+	0	0	+	0	+	0
4	+	0	+	0	+	0	+	0	0	0	+	0	+	+	+	0	+	0	+	1+
5	0	+	+	0	+	0	+	0	0	+	0	0	+	0	+	0	0	0	+	1+
6	0	0	+	+	+	0	+	0	+	+	0	+	0	+	0	+	+	0	+	1+
7	0	0	+	0	+	+	+	0	+	+	0	0	+	+	0	+	0	0	+	1+
8	0	0	+	0	+	0	+	+	0	0	+	+	0	0	+	+	+	0	+	3+
9	0	0	+	0	+	+	0	+	0	+	0	0	+	+	0	+	+	0	+	3+
10	+	+	0	0	+	0	+	+	+	+	+	+	0	0	+	0	+	+	+	(+)
TC	+	0	+	0	+	0	+	0	0	+	0	0	0	0	+	+	+	0	+	1+

Case Study 1: Eluate

	D	C	c	E	e	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	M	N	S	s	Lu^a	Lu^b	AHG
1	+	+	0	0	+	0	+	0	+	0	+	0	+	+	+	0	+	0	+	(+)
2	+	+	0	0	+	+	+	+	0	+	+	0	+	+	0	+	0	0	+	(+)
3	+	0	+	+	0	0	+	0	+	0	+	0	+	+	0	+	+	0	+	0
4	+	0	+	0	+	0	+	0	0	+	0	0	0	0	+	+	+	0	+	1+
5	0	+	+	0	+	0	+	+	+	+	0	0	+	+	0	+	+	0	+	1+
6	0	0	+	+	+	0	+	+	+	+	0	+	0	0	+	+	+	0	+	1+
7	0	0	+	0	+	+	+	0	+	+	0	0	+	+	+	0	+	0	+	1+
8	0	0	+	0	+	0	+	0	+	0	+	+	0	+	0	0	+	0	+	3+
9	0	0	+	0	+	0	+	+	0	+	+	0	+	0	+	0	+	0	+	3+
10	0	0	+	0	+	0	+	0	0	+	+	0	0	+	+	+	0	0	+	(+)
11	+	+	0	0	+	0	+	0	+	+	+	+	0	+	0	0	+	0	+	1+

Case Study 1: Other Testing

- Solid phase panel was panreactive
- Patient was e antigen POS

- Send out to Community Blood Center IRL corroborated findings
  - Full phenotype ascertained if transfusion of red blood cells (RBC) needed

Case Study 1: Day 7

- Patient appears to have resolved his UTI, however his clinical picture is much worse:
  - HGB: 6.2g/dL
  - PLT: 399K/µL
  - WBC: 27.4K/µL
  - T. Bili: 11.8mg/dL
- Urinalysis: Amber/Brown
  - Nitrite: NEG
  - Leukocytes: NEG
  - Blood: 3+
  - Bacteria: None
  - WBC: 0 – 2/HPF
  - RBC: 0 – 2/HPF

Case Study 1: Day 7

- A rapid drop in HGB prompts consults for advice in transfusion from the Blood Bank and Hematology
- Workup and results suggest autoimmune hemolytic anemia
  - Total Bilirubin (0.7 → 11.8mg/dL)
  - Lactate Dehydrogenase 1377U/L

Day	0	6	7 (Morning)	7 (Evening)
HGB (g/dL)	13.9	8.8	6.2	5.0
T. Bili (mg/dL)	0.7	-	11.8	11.8
BUN (mg/dL)	20	37	47	51

Case Study 1: Day 7

- Etiology of autoimmune hemolytic anemia not clear
  - Infection, drugs, and malignancy considered
- Transfusion not recommended
  - Patient is actively hemolyzing
  - Available phenotypically matched units are least incompatible

- Piperacillin/tazobactam administration stopped

Case Study 1: Day 8

- Patient's condition continues to deteriorate
- Additional testing:
  - Cold Agglutinins Titer: < 1:32
  - Haptoglobin < 30mg/dL
- Emergent therapeutic plasma exchange initiated (17 units)

Day	7 (Evening)	8 (Morning)	TPE	8 (Evening)
HGB (g/dL)	5.0	4.1		3.8
T. Bili (mg/dL)	11.8	12.4		8.0
BUN (mg/dL)	51	60		72

Case Study 1: Day 9

- Patient becomes clinically unstable and symptomatic, requiring transfusion of two units of RBCs throughout the day
- The patient has demonstrated acute kidney injury, dialysis started

Day	7 (Morning)	8 (Evening)	9 (Morning)	9 (Evening)
HGB (g/dL)	6.2	3.8	3.6	4.2
T. Bili (mg/dL)	11.8	8.0	3.4	2.6
BUN (mg/dL)	47	72	76	68
LDH (U/L)	1377		708	
Hapto (mg/dL)			<30	

Case Study 1: Days 10 – 13

- A new XM is ordered, the screen is NEG
  - The patient receives his final unit of RBC for this admission

Day	9 (Morning)	10	11	13
HGB (g/dL)	3.6	4.9	5.8	6.1
T. Bili (mg/dL)	3.4	2.6	1.5	1.2
LDH (U/L)	708	634	527	437
Hapto (mg/dL)	< 30	< 30	< 30	< 30

Case Study 1: Day 13 – 30 (Discharge)

- The patient would continue to improve and planned to find a different skilled nursing facility to better take care of his needs

Day	13	15	18	21	25	30
HGB (g/dL)	6.1	6.1	6.5	6.6	7.1	8.1
T. Bili (mg/dL)	1.2	1.0	0.7	0.5	0.5	0.5
LDH (U/L)	437	406	315	217	303	186
Hapto (mg/dL)	< 30	< 30	47	74	128	174

Case Study 1: Timeline

XM Ordered

Drug Stopped

XM SCN NEG
1 RBC
Transfused

Day	0	4	6	7	8	9	10	11	30
HGB (g/dL)	13.9	11.6	8.8	5.0	3.8	3.6	4.9	5.8	8.1
T. Bili (mg/dL)	0.7	-	-	11.8	8.0	6.2	2.6	1.5	0.5
BUN (mg/dL)	20	27	37	51	72	76	83	60	47
LDH (U/L)	-	-	-	1377	-	708	634	514	186
Hapto (mg/dL)	-	-	-	-	< 30	< 30	< 30	< 30	174

Piperacillin
Tazobactam

17 FFP TPE Performed

2 RBC Transfused

Case Study 2

- 71 years old male
- COPD, CAD, cardiomyopathy, DVT
- Presenting with shortness of breath and labored breathing
  - Determined to be caused by pleural effusion
- Prescribed medication to empirically treat health care-associated pneumonia
  - Cefepime, 2g every 8 hours
  - Vancomycin
  - Azithromycin

Case Study 2: Days 0 – 1

- On admission, laboratory results:
  - HBG: 10.2g/dL (baseline 9.0g/dL)
    - Patient has history of iron deficient anemia
  - PLT: 344K/μL
  - WBC: 2.8K/μL
  - T. Bili: 0.6mg/dL
  - INR: > 10
- Day 1: HGB drops to 8.1g/dL
  - Patient had a bout of hematochezia while in the ED, INR has since been corrected with vitamin K

Case Study 2: Days 2 – 6

- Patient being evaluated and treated for pleural effusion
  - HGB trending lower
    - Potentially from bleeding from chest tube

Day	0	1	3	5
HGB (g/dL)	10.2	8.1	7.8	7.1

- Day 5: Cefepime discontinued and replaced with ceftriaxone, 2g every 24 hours

Case Study 2: Days 7 – 8

- Day 7: HGB reaches transfusion cutoff, XM ordered
  - Screen: NEG
  - 1 unit RBC transfused

Day	5	6	7	8
HGB (g/dL)	7.1	7.4	6.9	7.0
T. Bili (mg/dL)	0.4	0.4	0.4	0.4
BUN (mg/dL)	25	17	13	12

- Day 8: Patient appears nonresponsive to previous day's transfusion another unit RBC ordered and transfused

Case Study 2: Days 8 – 10

- Patient appears to be recovering, HGB trending to baseline

Day	8	9	10
HGB (g/dL)	7.0	8.5	9.1
T. Bili (mg/dL)	0.4	0.5	0.5
BUN (mg/dL)	12	16	17

Case Study 2: Days 11 – 13

- Day 11: HGB again reaches transfusion cutoff, XM ordered
  - Screen: NEG

Day	10	11 (1)	11 (2)	12 (1)	12 (2)	12 (3)	12 (4)	13 (1)	13 (2)	13 (3)
HGB (g/dL)	9.1	7.1	7.1	6.5	6.9	7.9	6.5	7.1	7.1	7.0
T. Bili (mg/dL)	0.5	0.8	0.7	1.5	1.9	2.3	2.0	2.1	1.7	-
BUN (mg/dL)	17	30	30	30	30	24	26	23	24	-

- 9 units of RBC transfused over these 48 hours (at or below HGB 7.1), none apparently successful

Case Study 2: Day 14

- New XM ordered
  - Screen: NEG

Day	13 (2)	14 (1)	14 (2)	14 (3)
HGB (g/dL)	7.1	7.8	8.8	4.5
T. Bili (mg/dL)	1.7	2.5	3.2	-
BUN (mg/dL)	24	25	30	-

- Sudden drop in HGB to 4.5g/dL prompts for the transfusion of 2 units of RBC
- Patient also showing signs of delirium/metabolic encephalopathy

Case Study 2: Day 15 Workup

- Due to decrease in hemoglobin and increase in total bilirubin workup initiated
- DAT Results

Polyspecific	IgG Specific	Complement	Saline
2+	2+	1+	0/0

- Eluate: Nonreactive
- Urinalysis
  - Amber
  - Blood: 3+
  - RBC: 0 – 2/HPF
  - Urobilinogen: Increased

Case Study 2: Day 15

- Ceftriaxone quickly identified as a potential cause for hemolysis and administration stopped
- Patient transfused 2 units of RBC through the day

Day	14 (2)	14 (3)	15 (1)	15 (2)	15 (3)	15 (4)	15 (5)
HGB (g/dL)	8.8	4.5	6.7	7.6	5.4	9.3	9.3
T. Bili (mg/dL)	3.2	-	-	3.4	-	4.0	4.4
BUN (mg/dL)	30	-	68	45	50	62	68
LDH (U/L)	-	-	-	1635	-	-	-
Hapto (mg/dL)	-	-	-	< 30	-	-	-

Case Study 2: Days 16 – 27 (Discharge)

- Patient no longer requires transfusion and quickly recovers to a HGB 11.3g/dL
- Patient suffered an acute kidney injury, but did not require dialysis

Day	15 (2)	15 (3)	15 (5)	16	17	18	27
HGB (g/dL)	7.6	5.4	9.3	9.1	11.3	10.5	9.3
T. Bili (mg/dL)	3.4	-	4.4	4.4	2.5	1.6	0.6
BUN (mg/dL)	45	50	68	69	59	57	23
LDH (U/L)	1635	-	-	2513	878	-	-
Hapto (mg/dL)	< 30	-	-	< 30	38	-	-

Case Study 2: Timeline

2 RBC Transfused

2 RBC Transfused

Drug Stopped
2 RBC
Transfused

Day	0	1	5	8	9	10	13	14	15	16	17
HGB (g/dL)	10.2	8.1	7.1	7.0	8.5	9.1	7.1	4.5	7.6	9.1	11.3
T. Bili (mg/dL)	-	-	0.4	0.4	0.5	0.8	1.7	3.2	3.4	4.4	2.5
BUN (mg/dL)	-	-	25	12	16	17	24	30	45	69	59
LDH (U/L)	-	-	-	-	-	-	-	-	1635	2513	878
Hapto (mg/dL)	-	-	-	-	-	-	-	-	< 30	< 30	38

Cefepime

Ceftriaxone

9 RBC Transfused

Drug Induced Immune Hemolytic Anemia (DIIHA)

- Have been implicated in hemolytic anemias since the 1980s
- More than 130 drugs have been described as being associated with DIIHA
- Drugs most implicated in DIIHA (Garraty and Arndt, 2014)
  - 1. Piperacillin
  - 2. Cefotetan
  - 3. Ceftriaxone
  - On the rise: Platinum containing drugs

Drug Induced Immune Hemolytic Anemia (DIIHA)

- Classically, they were described and loosely organized by drug dependency in detection and theoretical mechanism of action:
  - Drug Dependent detection
    - Drug adsorption (DT)
    - "Immune complex" formation (+Drug)
  - Drug Independent detection
    - Autoantibody production (AA)
    - Nonimmunologic protein adsorption (NIPA)

Drug Adsorption (DT)

- Antibody directed against drug adsorbed to RBC
  - Screen: NEG
  - Eluate: Nonreactive
- Due to "distance" from cell surface, does not initiate complement cascade
  - DAT: IgG POS, sometimes Complement
  - Extravascular hemolysis, gradual

"Immune Complex" Formation (+Drug)

- Antibody directed against drug forms immune complexes that attach to RBC
  - Screen: NEG
  - Eluate: Nonreactive
- Immune complexes adsorbed to RBC surface can active complement cascade
  - DAT: Complement POS, sometimes IgG
  - Intravascular hemolysis, can be severe

Autoantibody Production (AA)

- Antibody directed against self RBCs develop as a result of potential immune system changes
  - Indistinguishable from WAIHA
  - Screen: POS
  - Eluate: Panreactive
- Autoantibodies develop
  - DAT: IgG POS, often Complement POS
  - Clinical presentation varies, typically extravascular hemolysis

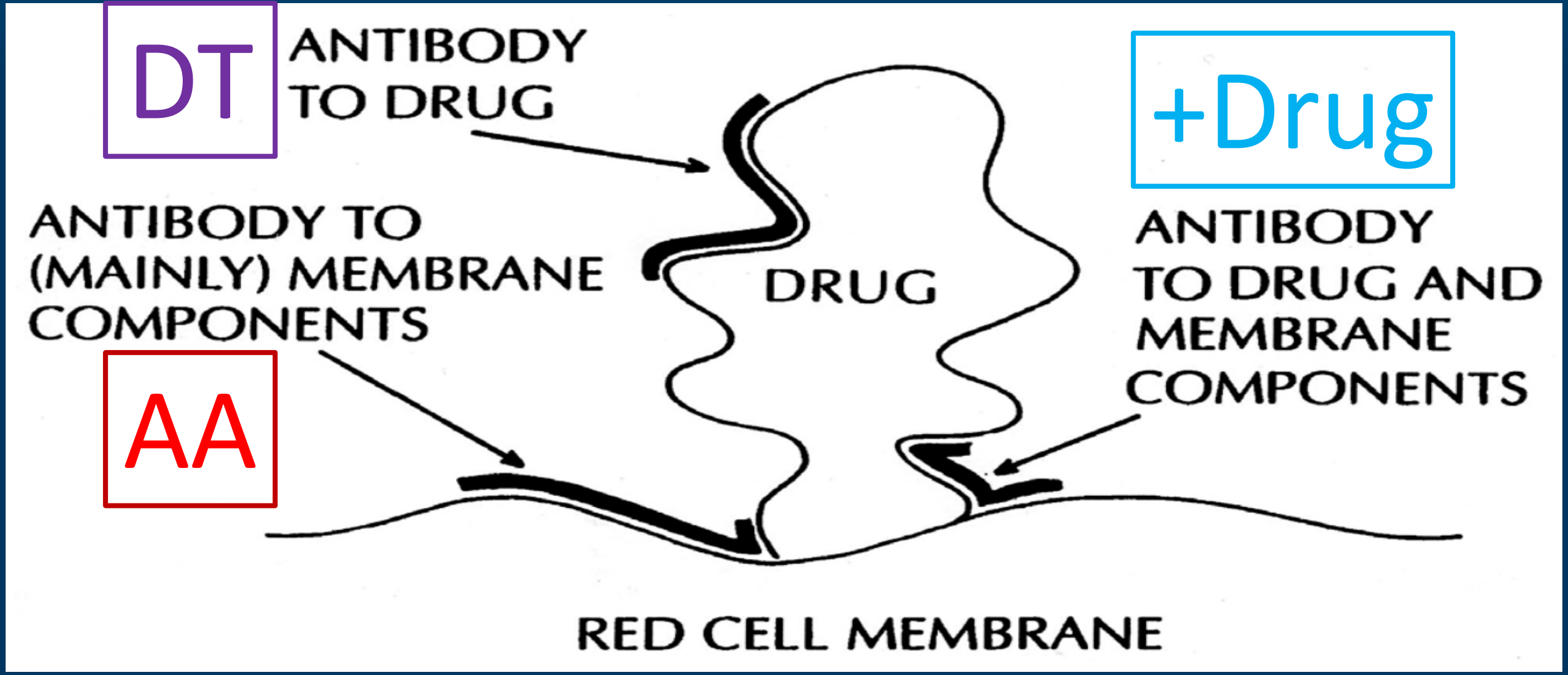
Nonimmunologic Protein Adsorption (NIPA)

- No antibody to any particular antigen is produced. Membrane modification by the drug causes nonspecific protein adsorption
  - Screen: NEG
  - Eluate: NEG
- RBC membrane will adsorb immunoglobulins, complement proteins, etc.
  - DAT: IgG POS, Complement POS
  - Can present with hemolysis, not well documented

Drug Induced Immune Hemolytic Anemia (DIIHA)

- Nowadays, there is not enough evidence to support DIIHA mechanisms being discreet and individual
- Each of the theoretical mechanisms have weaknesses and are at times, unable to explain things
- Several, similar unifying hypotheses have been proposed

DIIHA: A Unifying Hypothesis



Summary of DIIHA

	Serological Classification	Theoretical Mechanism	DAT		Serum		Eluate		Commonly Associated Drugs
			IgG	C3	DT	UT	DT	UT	
Drug-Dependent	Reactive with Drug-Treated Cells (DT)	Drug Adsorption	+	-/+	+	-	+	-	Penicillin, Other/Older Cephalosporins and Beta-Lactams
Drug-Dependent	Reactive in the Presence of Drug (+Drug)	"Immune Complex" Formation	-/+	+	-	-	-	-	Piperacillin, Ceftriaxone, Other Cephalosporins, Quinine, Quinidine
Drug-Independent	Autoantibody Production (AA)	Stimulate Autoantibody Production	+	+/-	+	+	+	+	Fludarabine, Methyldopa
Drug-Independent	Nonimmunologic Protein Adsorption (NIPA)	Membrane Modification, Nonspecific Protein Adsorption	+	+	+	-	-	-	Cephalothin, Platinum Containing Drugs

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