Carter BloodCare

My Patient, Your Patient, Our Patient-
Immunohematology Reference Lab

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Ultimate Goal

Carter BloodCare Mission Statement:
We save lives by making transfusion possible.

This applies to every aspect from the collection of the blood, to processing, testing, distributing and everything in between.

In the Reference and Transfusion Department(R&T), one area we are specifically interested in is pre-transfusion testing. Our goal is to identify any unexpected antibodies in the patient's sample and to provide a safe blood product to the patient in a timely manner.

- *What is the antibody?*
- *Is the antibody clinically significant?*
- *Can we find compatible blood?*
- *Does this antibody cause hemolytic disease of the newborn?*
- *How long is it going to take?*

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Let's get started

- **Receiving/Processing**
  - Lab assistants receive in specimen
  - Call to notify client receipt of specimen (if reference facility)
  - Centrifuge sample
  - Place sample/order in designated racks

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Now the fun begins

Getting started

- The next available tech will select a workup based on the order it was received and/or status or continue a workup from a previous shift
- A workup sent for antibody identification typically begins with a **D**irect **A**ntiglobulin **T**est (DAT) and antibody screen

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Testing considerations

Methodology

There is no perfect, one-size-fits-all test method.

- **Tubes (LISS, PeG)**

  Pros: test different phases (IS,RT,37,IAT), resolve discrepancies/rouleaux, specialized testing
  Cons: tech/technique dependent, labor intensive, subjective, unstable (must be read immediately)

- **Gel**

  Pros: small sample/reagent volume, less subjective interpretation, stable, can be automated
  Cons: time limitation, sensitivity (not enough/too much), equipment requirements/maintenance

- **Solid Phase (ECHO)**

  Pros: automated, stable, standardization, detect Kidd antibodies well, Knops system not detected
  Cons: larger volume requirement, unable to choose selected cells, sensitivity, time limitation

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Collaboration

- **General Considerations**
  - Methodology used at facility (gel, tubes, solid phase)
  - Results from testing performed at facility
    - +DAT?
    - Anagram results? All cells pos? 1 cell? Incompat xm?
  - Previous history check
    - WAA/CAA
    - Allo Ab? Ex. -K, -Js(b)
  - Additional Details
    - Recently transfused? BMT?
    - Diagnosis?
      - Multiple Myeloma, ITP, Leukemia, Sickle Cell
    - Ethnicity?
    - Medication?
      - WinRho, Daratumumab (DARA), anti-microbial (Cephalosporin)

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Generalizations

Test selection

- Based on assessment of the aforementioned criteria, experience, and gut instinct

Examples:

- *Typically* solid phase detects anti-D's and Kidd antibodies well; therefore, we may choose to use the ECHO although the client uses tubes or gel
- Antibodies in the Knops system are sometimes undetectable on the ECHO
- Warm auto antibodies (and often cold antibodies) cannot be resolved in solid phase and are often difficult to complete in gel; therefore, WAA/CAA's are *usually* resolved using tube testing
- High titer low avidity antibodies (HTLA) are *generally* best reactive in gel (may not be detectable using tube testing)
- Rouleaux cannot be resolved in gel resulting in a different choice of methodology

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Ex. 1 Antibody Screen (tubes)

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN					
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	LISS 37°	LISS IAT	PeG IAT
1	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	0	0	+	0	0✓	0✓
2	+	0	+	+	0	0	0	+	+	+	0	+	0	+	+	0	+	0	+	0	2+	2+
3	0	0	+	0	+	+	+	+	0	+	0	+	0	0	+	+	0	+	0	0	0✓	0✓
AC																				0	0✓	0✓

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Selected cells

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN					
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	LISS 37°	LISS IAT	PeG IAT
4	+	+	0	0	+	0	0	+	+	0	+	0	0	+	0	+	0	0	+	0	0✓	0✓
6	+	0	+	+	0	0	0	+	+	+	0	+	0	+	+	+	0	+	0	0	2+	2+
9	0	0	+	0	+	+	0	+	0	+	0	+	0	0	+	0	+	+	0	0	0✓	0✓

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Simple antibody ID

Name: | Patient ID#: | Case Number: | Testing Date:
DOB: | Diagnosis: | Requesting Facility: | Previous Record Check:

ABO/Rh TYPING

Antigens						Cells			Interp	Tech
A	B	A,B	A_1	O	New A_1		A_1	B		

ADDITIONAL Rh TYPING

Antigens				Interp	Tech
C	E	c	e		AJ

DAT

	Poly	IgG	C3	Ctl	Tech
IS	0				AJ
RT	0+				

ANTIBODY SCREEN

Mfg/Lot #	Tech
I	
II	
III	
AUTO	

Interp: ☐Pos ☐Neg

ADDITIONAL RED BLOOD CELL PHENOTYPES ☐ Cell Treatment

Antisera	M	N	S	s	K	k	Fy^a	Fy^b	Jk^a	Jk^b	P_1	Le^a	Le^b	Tech
Patient														
Pos Ctl														
Neg Ctl														

Mfg, Lot #, & Exp. Date

MISCELLANEOUS TESTING Tech

RESULTS / CONCLUSIONS:

Anti-E

LifeTrak™ Lab Entry: | Tech 1 | Tech 2 | Worksheet #: | Donor Unit Disposition:

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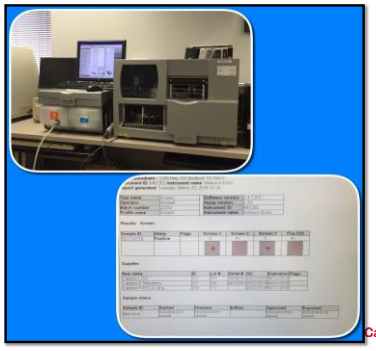
ECHO (Ex. Anti-E)



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Gel (Ex. Anti-E)

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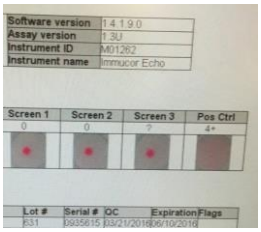
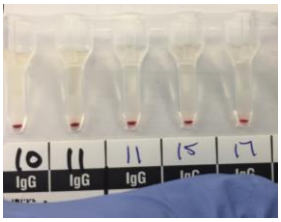
Weak reactions/non-specific rxns





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Example 2

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN					
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	LISS 37°	LISS IAT	PeG IAT
1	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	0	0	+	0	2+	2+
2	+	+	0	0	+	0	+	+	0	+	0	+	0	+	+	+	+	+	0	0	2+	2+
3	+	0	+	+	0	0	0	+	+	+	+	+	0	0	0	0	+	+	+	0	2+	2+
4	+	0	+	0	+	+	0	+	+	0	+	0	0	+	+	0	+	0	+	0	2+	2+
5	0	+	+	0	+	+	0	+	+	0	0	+	+	0	+	+	+	0	+	0	2+	2+
6	0	0	+	+	+	+	0	+	0	+	+	+	0	0	0	0	+	+	0	0	2+	2+
7	0	0	+	0	+	+	+	+	0	0	+	0	0	+	+	+	+	0	+	0	2+	2+
8	0	0	+	0	+	+	0	+	0	+	+	+	+	0	+	0	+	0	+	0	2+	2+
9	0	0	+	0	+	+	0	+	+	+	+	0	0	+	0	+	+	+	0	0	2+	2+
10	0	0	+	0	+	+	0	+	0	+	+	0	0	0	+	+	0	0	+	0	2+	2+
AC																				0	2+	2+

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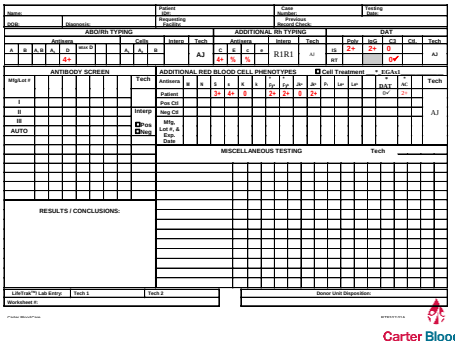
Next step



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Conclusion

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN						
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s		Low Ph EL	LW	2X PeG Ads plasma *
1	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	0	0	+		2+	0✓	0✓
2	+	0	+	+	0	0	0	+	+	+	0	+	0	+	+	0	+	0	+		2+	0✓	0✓
3	0	0	+	0	+	+	+	+	0	+	0	+	0	0	+	+	0	+	0		2+	0✓	0✓
*				%	%			0				0									2+		0✓

*W035215235237-E (adsorbing cell)

Warm auto Ab with no underlying Allo antibodies

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Case study 1

- 37 year old pregnant, Hispanic female
- Presented to hospital for delivery with a 3g/dl Hgb
- Referred to our facility by a reference account

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN							
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	LISS 37°	LISS IAT	PeG IAT	Gel
1	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	0	0	+	0	1+	2+s	2+
2	+	0	+	+	0	0	0	+	+	0	0	+	0	+	+	0	+	0	+	0	1+s	3+	2+
3	0	0	+	0	+	+	+	+	0	+	0	+	0	0	+	+	0	+	0	0	1+	2+s	3+
AC																				0	0✓	0✓	0

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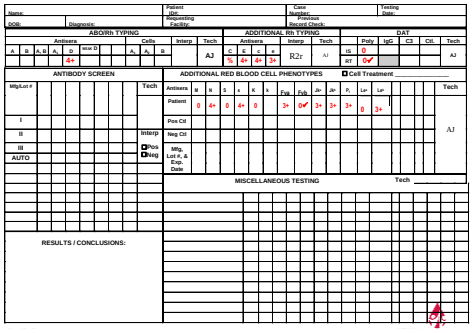
Next step



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Enzymes/Adsorption

	Rh-Hr						Kell		Duffy		Kidd		Lewis		P	MN						
	D	C	c	E	e	f	K	k	Fy^a	Fy^b	Jk^a	Jk^b	Le^a	Le^b	P_1	M	N	S	s	PeG IAT	.2M DTT	Ficin 37/IAT	#
1	+	+	0	0	+	0	0	+	+	0	+	0	+	0	0	+	0	0	+				0 ✓
2	+	0	+	+	0	0	0	+	+	0	0	+	0	+	+	0	+	0	+				0 ✓
3	0	0	+	0	+	+	+	+	0	+	0	+	0	0	+	+	0	+	0				0 ✓
***W03521 6084820**		0					0			0								0		3+	3+	0/0✓	0 ✓
W03521 5246554	(Dib=)																			2+s			
O cord																				3+			

***phenotypically matched**
 # 2x PeG adsorbed plasma w/ *

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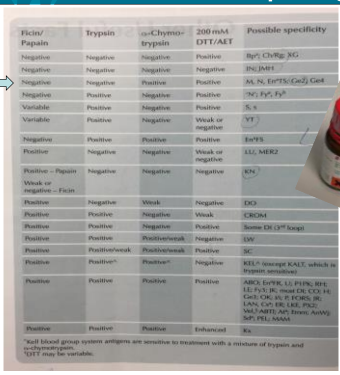
The Blood Group Antigen Facts Book



Ficin/ Papain	Trypsin	α-Chymo-trypsin	200 mM DTT/AET	Possible specificity
Negative	Negative	Negative	Positive	Bp^a; Ch/Rg; XG
Negative	Negative	Negative	Negative	IN; JMH
Negative	Negative	Positive	Positive	M, N, En^{a}TS; Ge2; Ge4
Negative	Positive	Negative	Positive	'N'; Fy^a, Fy^b
Variable	Positive	Negative	Positive	S, s
Variable	Positive	Negative	Weak or negative	YT
Negative	Positive	Positive	Positive	En^{a}FS
Positive	Negative	Negative	Weak or negative	LU, MER2
Positive – Papain Weak or negative – Ficin	Negative	Negative	Negative	KN
Positive	Negative	Weak	Negative	DO
Positive	Positive	Negative	Weak	CROM
Positive	Positive	Negative	Positive	Some DI (3rd loop)
Positive	Positive	Positive/weak	Negative	LW
Positive	Positive/weak	Positive/weak	Positive	SC
Positive	Positive^	Positive^	Negative	KEL^ (except KALT, which is trypsin sensitive)
Positive	Positive	Positive	Positive	ABO; En^{a}FR; LI; P1PK; RH; LE; Fy3; JK; most DI; CO; H; Ge3; OK; I; P; FORS; JR; LAN; Co^a; ER; LKE; PX2; Vel,† AB[illegible]; At^a; [illegible]; AnWj; Sd^a; PEL; MAM
Positive	Positive	Positive	Enhanced	Kx

^Kell blood group system antigens are sensitive to treatment with a mixture of trypsin and α-chymotrypsin.
†DTT may be variable.

Reid ME and Lomas-Francis C. The Blood Group Antigen Facts Book. San Diego, CA: American Press, 3rd Edition, 2012

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Confirmation

➢ Type patient for Ge2 Ag

➢ Test Ge:-2 cells against patient's plasma

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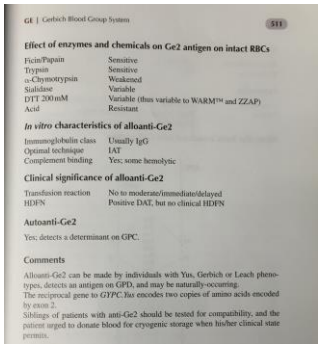
Blood selection

Reid ME and Lomas-Francis C. The Blood Group Antigen Facts Book. San Diego, CA: American Press, 3rd Edition, 2012



GE | Gerbich Blood Group System 511

Effect of enzymes and chemicals on Ge2 antigen on intact RBCs

Ficin/Papain	Sensitive
Trypsin	Sensitive
α-Chymotrypsin	Weakened
Sialidase	Variable
DTT 200 mM	Variable (thus variable to WARM™ and ZZAP)
Acid	Resistant

***In vitro* characteristics of alloanti-Ge2**

Immunoglobulin class	Usually IgG
Optimal technique	IAT
Complement binding	Yes; some hemolytic

Clinical significance of alloanti-Ge2

Transfusion reaction	No to moderate/immediate/delayed
HDFN	Positive DAT, but no clinical HDFN

Autoanti-Ge2

Yes; detects a determinant on GPC.

Comments

Alloanti-Ge2 can be made by individuals with Yus, Gerbich or Leach phenotypes, detects an antigen on GPD, and may be naturally-occurring. The reciprocal gene to GYPC.Ile encodes two copies of amino acids encoded by exon 2. Siblings of patients with anti-Ge2 should be tested for compatibility, and the patient urged to donate blood for cryogenic storage when his/her clinical state permits.

- Import/ARDP
- Type siblings
- Autologous donation when available

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What's new and exciting?

- **Molecular**
  - Screening donors
  - Testing patients
- **Daratumumab (DARA)**
  - Monoclonal anti-CD38
  - Promising for Multiple Myeloma patients

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It takes a TEAM effort

- **Everyone plays an integral role in getting the proper blood to the patient**
  - Phlebotomist
  - Referring facility
  - Physician
  - Nurse
  - Lab
  - Family

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Questions?

Photo credit: Bernard Simon

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References

1. Reid ME and Lomas-Francis C. The Blood Group Antigen Facts Book. San Diego, CA: American Press, 3rd Edition, 2012

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