



TO DIFF OR NOT TO DIFF.....

A Guide to Truth Table Analysis

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OBJECTIVES

- ❖ Why re-evaluate diff review criteria?
- ❖ What is a truth table?
- ❖ What is required for a valid truth table analysis
- ❖ Look at truth table results to establish what is significant and what is not
 - ❖ What is TP, TN, FP, FN?
- ❖ Making adjustments to review criteria
- ❖ Examples

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Why re-evaluate diff review criteria?





- ❖ Using the same review criteria from 1960's
- ❖ Aren't enough warm bodies to do the work
- ❖ Feel like you are doing diffs on every slide and the majority are "normal"

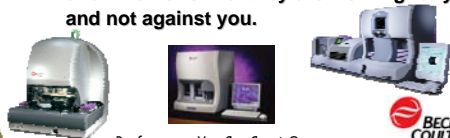


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What is a truth table?


- ❖ Determines the sensitivity, specificity and efficiency of the automated differential in distinguishing normal from abnormal specimens
- ❖ Looks at the Laboratory's Definitive flags and whether or not they are working for you and not against you.



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Variables to Take Into Consideration




- ❖ **Patient Population**
 - ❖ Primarily Normal or Abnormal?
 - ❖ Good representative mix during data collection?
- ❖ Sensitivity settings on the instrument
- ❖ Cut-offs to determine a positive / negative result
- ❖ Manual diff results – Rumke's Rules & Tech to Tech variation
- ❖ What is the medical significance of a band?

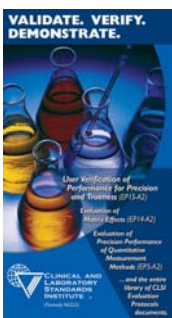
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What is necessary for a valid truth table analysis




- ❖ Sources such as:
 - ❖ CLSI-H20A - "Reference Leukocyte Differential Count and Evaluation of Instrumental Methods"
 - ❖ CLSI-EP09A - "Method Comparison and Bias Estimation Using Patient Samples"
 - ❖ www.clsi.org




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What is necessary for a valid truth table analysis




❖ Guideline Recommendations:

- ❖ Make sure the instrument is in good operating order
 - ❖ Clean
 - ❖ Recent Calibration
 - ❖ Controls are in range
- ❖ 200 specimens are necessary for valid statistical results (minimum of 40)
 - ❖ Not more than 25% of the total number on a single day
- ❖ Samples should be a good representation of normal workload; including both normal and abnormal patients
- ❖ Specimens and slides should be run / made within 4 hours of collection


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What is necessary for a valid truth table analysis




❖ Guideline Recommendations:

- ❖ Specimen should be inverted at least 20 times before slide preparation
- ❖ 3 wedge slides for each specimen – A, B and C
- ❖ Stain slides within one hour of preparation
- ❖ Technologist should be "qualified"
 - ❖ Be able to classify all common cells
 - ❖ Know most WBC and RBC variations both congenital and acquired
 - ❖ Attitude, motivation and concentration are key factors


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What is necessary for a valid truth table analysis



❖ Guideline Recommendations:

- ❖ 400-cell differentials should be performed on each specimen
 - ❖ 200-cell differential on one of the two slides by each Technologist; one uses slide **A** and one uses slide **B**
- ❖ Differentials should be "blind"
- ❖ Call Morphology if it is something you would act on or consider medically significant
- ❖ Spare slides are for discrepancy results or recounts by a third qualified Technologist


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Definitive and / or Action Limits

	Reference Limits		Definitive (Action) Limits		Critical Limits	
	Low	High	Low	High	Low	High
WBC						
RBC						
Hgb						
HCT						
MCV						
MCH						
MCHC						
RDW						
Plt						
MPV						
Ne %						
Ly %						
Mo %						
Eo %						
Ba %						
NRBC %						
Ne #						
Ly #						
Mo #						
Eo #						
Ba #						
NRBC #						

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Manual Differential Abnormal Limits

❖ Define manual diff abnormal limits for each cell type

- ❖ Consult with Pathologist
- ❖ Be specific; if 15% is the limit for bands then 14% is normal or negative and 16% is abnormal or positive


Segmented neutrophils	_____	Metamyelocytes	_____
Band neutrophils	_____	Myelocytes	_____
Lymphocytes	_____	Promyelocytes	_____
Variant Lymphocytes	_____	Blasts	_____
Monocytes	_____	NRBCs	_____
Eosinophils	_____	Other (specify)	_____
Basophils	_____	Other (specify)	_____

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CLSI Clinical Sensitivity Study

H20-A Specimen Types for Clinical Sensitivity Study

Clinical Condition	Characteristic Leukocyte Differential Count Finding	Absolute Cell Count	Proportional Cell Count
Acute Inflammation Bacterial Infection	Granulocytosis and/or	$\geq 9.0 \times 10^9/L$	>80 %
	Left Shift (band forms)	$\geq 0.9 \times 10^9/L$	>6 %
Chronic Inflammation	Monocytosis	$\geq 0.8 \times 10^9/L$	>10 %
Parasitic Infection	Eosinophilia	$\geq 0.5 \times 10^9/L$	>7 %
Allergic Reaction			
Viral Infections	Lymphocytosis and/or	$\geq 3.5 \times 10^9/L$	>50 %
	Lymphocytes, variant forms**	$\geq 0.7 \times 10^9/L$	
infectious mononucleosis cytomegalovirus infection infectious hepatitis			
Aplastic anemia, chemotherapy	Granulocytopenia	$\leq 1.5 \times 10^9/L$	<10 %
HIV infection	Lymphopenia	$\leq 1.0 \times 10^9/L$	<7 %
Acute leukemia	Immature cells, including blasts**	$\geq 0.1 \times 10^9/L$	>2 %
Severe anemia myelophthisic anemia	Nucleated red blood cells**	$\geq 0.02 \times 10^9/L$	>2 %



International Society for Laboratory Hematology

INTERNATIONAL CONSENSUS GROUP FOR HEMATOLOGY REVIEW

The International Consensus Group for Hematology Review is pleased to publish the following guideline:

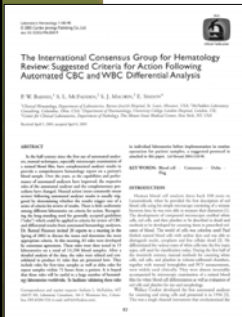
Suggested Criteria for Action Following Automated CBC and WBC Differential Analysis

<http://ISLH.org/2004/Committees/ConsensusGroup/CGCCConsensusRules.htm>





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ISLH CONSENSUS RULES




- ❖ Group representing 6 countries / 17 hospitals
- ❖ Reviewed >13,000 samples using the same review criteria
- ❖ Established 41 “rules” or “guidelines” for smear review
- ❖ Incorporate “delta” checks
- ❖ Bands are not evaluated



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ISLH Consensus Report ISLH.org



Rule #	Parameter	Primary and/or	Secondary and/or	Tertiary and/or	Fourth	Action 1	Action 2	Action 3
1	Neonate	First sample					Slide Review	
2	WBC, RBC, HGB, PLT, Retics	Exceeds in any					Dilute sample and rerun	
3	WBC, PLT	Lower than Lab verified reference in any					Follow lab SOP	
4	WBC, RBC, HGB, PLT	Vols Cut					Check sample for clot	Rerun sample. If possible, perform alternate counting method
5	WBC	<4.0 OR >30.0	and	first time			Slide Review	
6	WBC	<4.0 OR >30.0	and	delta failed	and	within 3 days	Slide Review	
7	PLT	<100 OR >1000	and	first time			Slide Review	
8	PLT	Any value	and	Delta Check Fail			Slide Review	
9	HGB	>7g/dl or >2g/dl above lab or reference range for age and sex	and	first time			Slide Review	Verify sample integrity if indicated
10	MCV	<75fl or >105fl (Adult)	and	first time	and	specimen is < 24 hours old	Slide Review	
11	MCV	>105 fl	and	adult	and	specimen is > 24 hrs old	Slide Review for macrocytic associated changes	Request fresh sample if MCV macrocytic associated changes seen. Report with comment if fresh sample is not available
12	MCV	Any value	and	delta fails	and	specimen is < 24 hours old	Verify sample integrity/stability	
13	MCHC	>=2 units above upper limit of reference range					Check for spheria, hemolysis, RBC agglutination, spurritoxemia	

WHITE PAPERS

Laboratory Hematology 11/09/06
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**The International Consensus Group for Hematology
Review: Suggested Criteria for Action Following
Automated CBC and WBC Differential Analysis**


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HOW TO ESTABLISH A TRUTH TABLE

❖ For each specimen analyzed on the instrument

- ❖ Classify the instrument results as Normal ("negative") if **NO definitive flags** (aH, aL, cH, or cL) or **suspect messages** are present or none of the results fail your "new" review criteria
- ❖ Classify the instrument results as Abnormal ("positive") if **definitive flags** (aH, aL, cH, or cL) or **suspect messages ARE** present or any of the results fail your "new" review criteria



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NORMAL vs ABNORMAL

❖ NORMAL	❖ ABNORMAL
WBC 3.5	WBC 31.7 aH
NE % 48.6	NE % 93.7 aH
LY % 40.7	LY % 3.0
MO % 7.4	MO % 3.3
EO % 2.8	EO % 0.0
BA % 0.5	BA % 0.0
NRBC %0.0	NRBC %0.0
RBC 3.02	WBC 0.7 aL
HGB 9.1	WBC 7.6 R
HCT 27.3	LY % 88.9 R
MCV 90.3	MO % 3.2 R
MCH 30.1	EO % 0.3 R
MCHC 33.4	BA % 0.0 R
RDW 18.4	NRBC %5.2 RCH
PLT 331	PLT 6 cL
MPV 8.6	NPV 8.0


Suspect (Normal side)

Suspect (Abnormal side)

Imm. NE 2 (Abnormal side)

Imm. NE 1 (Abnormal side)

Platelet clumps (Abnormal side)




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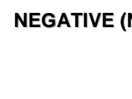
NORMAL vs ABNORMAL

❖ Indicate on the printout whether it is considered


POSITIVE (P)



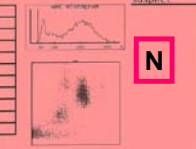
NEGATIVE (N)



WBC	14.8	WBC Est		SUSPECT
NE %	76.2	Neut		
LY %	14.2	Lymph		
MO %	7.0	Mon		
EO %	2.1	Eos		
BA %	0.5	Baso		
NRBC	0.0	NRBC		
NEC #	111.3	Band		
LY #	2.1	Imm Gran		
MO #	1.0	Var Lymph		
EO #	0.3	BTast		
BA #	0.1			
NRBC #	0.0			






WBC	4.9	WBC EST		SUSPECT
NE %	74.4	Neut		
LY %	13.5	Lymph		
MO %	10.6	Mon		
EO %	1.4	Eos		
BA %	0.1	Baso		
NRBC	0.0	NRBC		
NEC #	3.7	Band		
LY #	0.7	Imm Gran		
MO #	0.5	Var Lymph		
EO #	0.1	BTast		
BA #	0.0			
NRBC #	0.0			




HOW TO ESTABLISH A TRUTH TABLE

❖ For each manual diff:
❖ Average the differential results



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HOW TO ESTABLISH A TRUTH TABLE

❖ For each manual diff:

- ❖ Classify as Normal ("negative") any manual diff in which all cell types are normal and within your established limits
- ❖ Bands = 14 and upper acceptable limit is 15 then this differential would be considered **NEGATIVE**
- ❖ 1+ Aniso is called on RBC morphology and ≥ 2+ is considered abnormal then this differential would be considered **NEGATIVE**

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HOW TO ESTABLISH A TRUTH TABLE

❖ For each manual diff:

- ❖ Classify as **Abnormal** ("positive") **ANY** morphological or distributional abnormality present
- ❖ Bands = 16 and upper acceptable limit is 15 then this differential would be considered **POSITIVE**
- ❖ 3+ Aniso is called on RBC morphology and $\geq 2+$ is considered abnormal then this differential would be considered **POSITIVE**

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Truth Table Definitions

❖ Categorize each specimen as one of the following four categories and record the results on the Truth Table Worksheet:

- ❖ **True Positive** – results from both the instrument and the manual diff are positive
- ❖ **True Negative** – results from both the instrument and the manual diff are negative
- ❖ **False Positive** - results from the instrument are positive, but the manual diff is negative.
- ❖ **False Negative** - results from the instrument are negative, but the manual differential is positive.

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True Pos? False Pos? True Neg? False Neg?

❖ After reviewing the manual diff results add TRUE or FALSE to the report:

TRUE POSITIVE (TP)

or

FALSE POSITIVE (FP)

WBC	34.8	WBC	ELS	
HE %	76.2	HE	ELT	
LY %	24.2	LY	ELM	
MO %	7.0	MO	ELN	
EO %	2.1	EO	ELX	
BA %	0.5	BA	ELY	
NRBC	0.0	NRBC	ELZ	
MP	0.1	MP	ELAA	
LY #	2.1	LY	ELAB	
MO #	0.0	MO	ELAC	
EO #	0.3	EO	ELAD	
BA #	0.1	BA	ELAE	
NRBC #	0.0	NRBC	ELAF	

TP

TRUE NEGATIVE (TN)

or

FALSE NEGATIVE (FN)

WBC	4.9	WBC	ELS	
HE %	74.4	HE	ELT	
LY %	23.5	LY	ELM	
MO %	10.6	MO	ELN	
EO %	2.4	EO	ELX	
BA %	0.2	BA	ELY	
NRBC	0.0	NRBC	ELZ	
MP	0.2	MP	ELAA	
LY #	0.2	LY	ELAB	
MO #	0.5	MO	ELAC	
EO #	0.1	EO	ELAD	
BA #	0.0	BA	ELAE	
NRBC #	0.0	NRBC	ELAF	

TN

TRUTH TABLE WORKSHEET

❖ Tally the TN, FN, TP and FP and place these totals on the worksheet:

Reference Manual Differential	Instrument Results			Total
	Normal (Negative)	Normal (Negative)	Abnormal (Positive)	
	True Negative	False Positive	Total	
Normal (Negative)	100	3	103	200
Abnormal (Positive)	2	95	97	
Total	102	98	200	Total # specimens utilized for study

TRUTH TABLE WORKSHEET


❖ Using the totals placed on the previous worksheet, calculate your statistics:

PARAMETER	CALCULATION	RESULT (%)
% TN	Number of TN/Total	50.0%
% TP	Number of TP/Total	47.5%
% FN	Number of FN/Total	1.0%
% FP	Number of FP/Total	1.5%
Pass Rate	% TN + % FN	51.0%
Review Rate	% TP + % FP	49.0%
Specificity	$[\# \text{ TN} / \# (\text{TN} + \text{FP})] \times 100$	97.1%
Sensitivity	$[\# \text{ TP} / \# (\text{TP} + \text{FN})] \times 100$	97.9%
Predictive Value of a Negative Test	$[\# \text{ TN} / \# (\text{TN} + \text{FN})] \times 100$	98.0%
Predictive Value of a Positive Test	$[\# \text{ TP} / \# (\text{TP} + \text{FP})] \times 100$	96.9%
Efficiency	$[(\# \text{ TP} + \# \text{ TN}) / \text{Total}] \times 100$	97.5%


THOUGHT QUESTION

❖ If your flagging limits are set correctly, ideally which two categories on the Truth Table should contain most of your samples?

- ❖ True Positives
- ❖ True Negatives





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THOUGHT QUESTION

❖ If your laboratory's FP rate is around 40% what corrective action do you think would help?

- ❖ Broaden the instrument limits / definitive flags



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THOUGHT QUESTION

❖ If your laboratory's FN rate is around 35% what corrective action do you think would help?

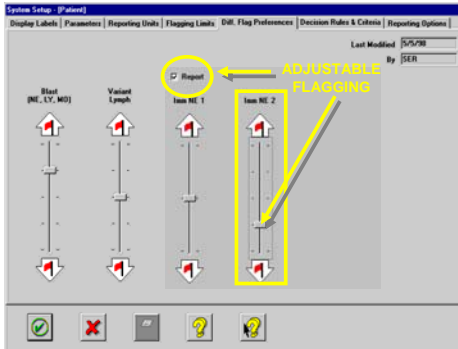
- ❖ Tighten the instrument limits / definitive flags
- ❖ Adjust sensitivity settings
 - ❖ LH700 Series
 - ❖ DxH800

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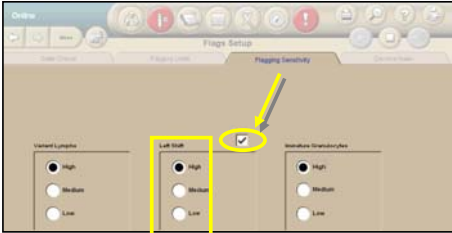


LH700 Series FLEXIBLE FLAGGING




The screenshot shows a software interface for 'System Setup - [Patient]'. It features several vertical sliders for 'Start (MLC-WL)', 'Variant Lymph', 'Item NE 1', and 'Item NE 2'. A yellow box highlights the 'Item NE 2' slider, with a yellow arrow pointing to it from the text 'ADJUSTABLE FLAGGING'. A yellow circle highlights the 'Report' checkbox, which is currently checked. At the bottom, there are icons for a green checkmark, a red X, a grey square, a yellow question mark, and a yellow lightbulb.


DxH800 Series FLEXIBLE FLAGGING



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THOUGHT QUESTION




❖ If your laboratory's **EFFICIENCY** is around **40%** what does this mean?

- ❖ The laboratory needs to re-evaluate their protocol and make adjustments to increase their TP / TN and decrease FP / FN



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REAL LIFE EXAMPLES

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