

[www.inspq.qc.ca](http://www.inspq.qc.ca)



# Performance of AxSYM HIV Ag/Ab Combo assay, a 4<sup>th</sup> generation assay for routine HIV screening

Christiane Claessens, Claire Béliveau  
HIV Testing Conference, March 25, 2010

# Québec and HIV

---



## Population

- 8,000,000 (23 % of Canada)

## Persons living with HIV/AIDS (2008)

- 14,000- 21,300 (Canada 65,000)

## Annual new cases

- 500-1,200 (Canada 2,300-4,300)

## Annual HIV tests

- $\approx$  300,000

# HIV testing in Québec

---

## 33 testing sites

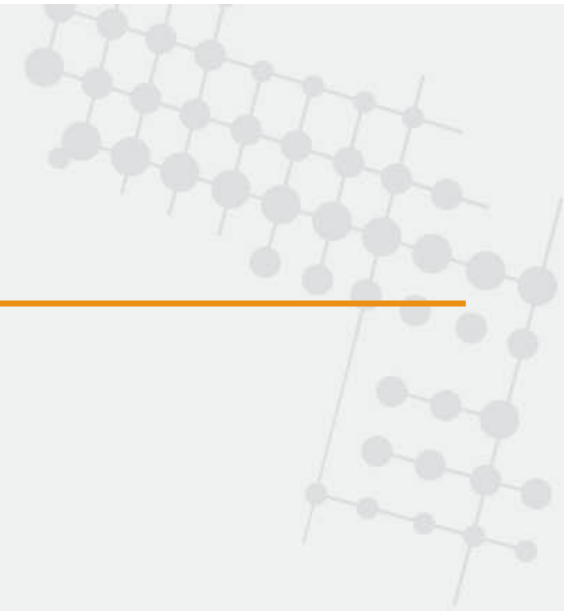
- 25 hospital laboratories
- 8 private laboratories

## In 2007, 25/33 laboratories used

- AxSYM HIV-1/2 gO (Abbott), a 3<sup>rd</sup> gen. assay

## LSPQ

- Supplemental tests on all reactive specimens
  - WB, p24 Ag EIA, HIV-2 EIA, HIV-1/HIV-2 LIA
- On request, if acute infection suspected and Abs negative
  - p24 Ag EIA



# Introduction of a 4<sup>th</sup> generation assay

---

## Fall 2007

- The first 4<sup>th</sup> generation assay was licensed for diagnosis in Canada:
  - AxSYM HIV Ag/Ab Combo\* (Abbott)
- \*not available in the US

# Assays format

---

## AxSYM HIV-1/HIV2 gO

### 3rd gen assay

- microparticle immunoassay
  - recombinant Ag
    - HIV-1 M and group O Env
    - HIV-1 core
    - HIV-2 Env
  - synthetic peptides
    - HIV-1 Env
    - HIV-2 Env.

Detection of fluorescent products

## AxSYM Ag/Ab HIV Combo

### 4th gen assay

- microparticle immunoassay
  - recombinant Ag
    - HIV-1 M and group O Env
    - HIV-1 core
    - HIV-2 Env
  - synthetic peptides
    - HIV-1 Env
    - HIV-2 Env.
- **HIV p24 monoclonal Ab**

Detection of fluorescent products



# Sensitivity assessment - Ag/Ab assay

---

Sera from patients with documented acute infection :

24 non reactive with 3<sup>rd</sup> gen test:

- 23 Ag POS
- 1 Ag NEG (3 months before WB POS)

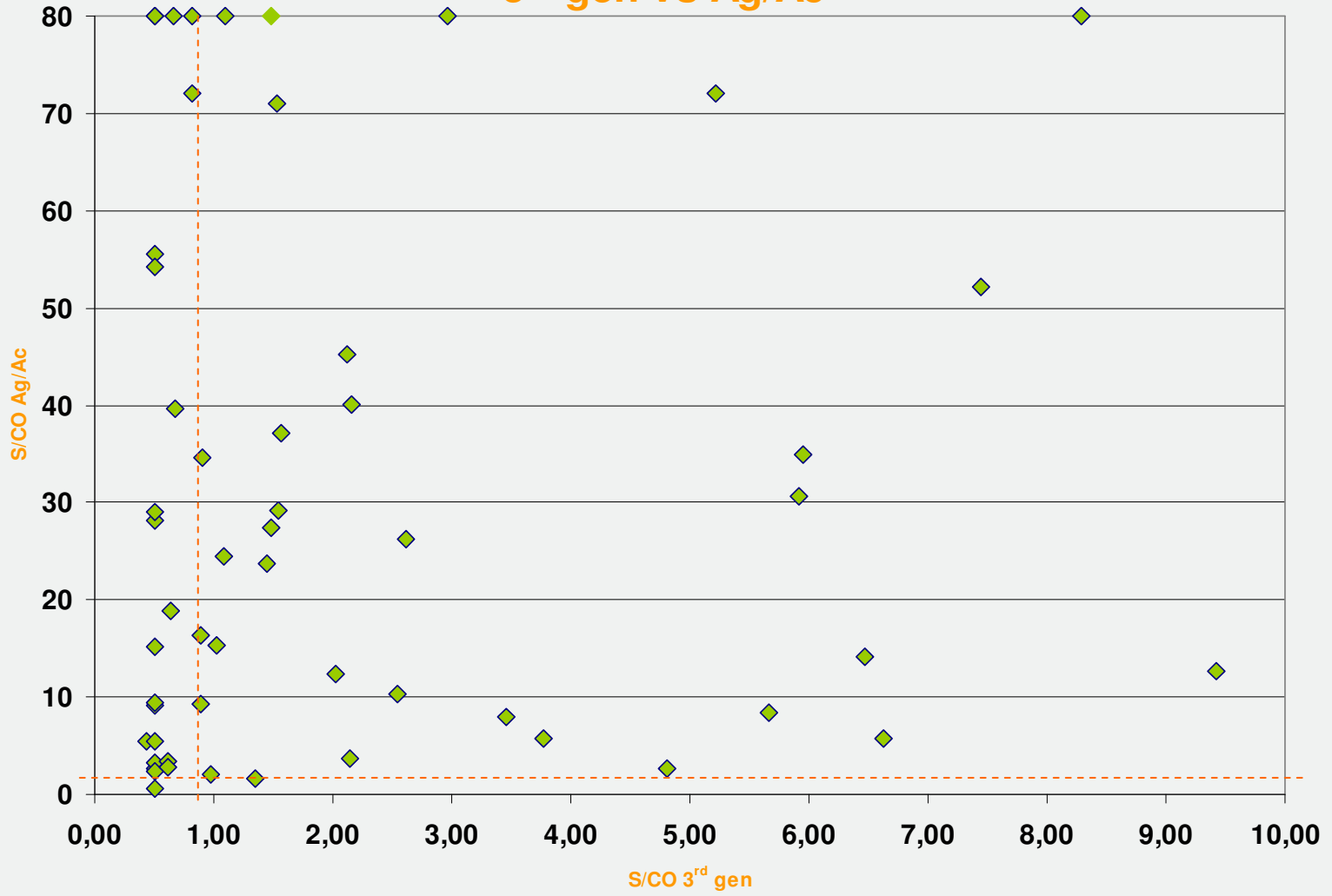
32 reactive (signal/cutoff values <10.0) with 3<sup>rd</sup> gen test:

- 32 Ag POS (15 WB NEG; 17 WB IND)

## Results

**All** p24 positive samples were reactive with 4<sup>th</sup> gen assay

# Comparison of S/CO values 3<sup>rd</sup> gen vs Ag/Ac



# Introduction of Ag/Ab in routine use

---

In 2008, sites using AxSYM platform switched to the Ag/Ab :

Algorithm adopted:

- $S/CO < 0.90$  → no further testing
  - Negative for Ab and Ag
- $0.90 \leq S/CO < 1.00$  → Greyzone (GZ) → repeat  
If GZ on repeat testing:
  - Equivocal, follow-up specimen (3 – 6 weeks) requested
- $S/CO > 1.00$  on repeat testing
  - Referred to LSPQ for supplemental testing



# Objectives

---

Assess the impact of the introduction of this new Ag/Ab test on

- the specificity of HIV serological results
- the detection of acute infections.



# Methods

---

## Data examined:

- Trends in reactivity and confirmatory rates before and after implementation of the Ag/Ab test
- Cases of acute infection detected

## Limitation of the study:

- Sequential data, no parallel testing of specimens

# Trends in reactive and HIV confirmed specimens

---

	2007 3 <sup>rd</sup> gen	2008 Ag/Ab
Tested	294 308	300 923
Reactive	0.78 % (2 290)	0.74 % (2 228)
Confirmed	70.8 % (1 622)	71.6% (1 595)

# Trends in selected sites

	Site A		Site B		Site C	
	3 <sup>rd</sup>	Ag/Ab	3 <sup>rd</sup>	Ag/Ab	3 <sup>rd</sup>	Ag/Ab
Tested	30,530	32,193	29,773	31,226	47,230	48,688
Reactive	371 (1.2%)	350 (1.1%)	147 (0.49%)	145 (0.46%)	229 (0.48%)	226 (0.46%)
Confirmed	304 (81.9%)	332 (94.8%)	74 (50.3%)	92 (63.4%)	126 (55.0%)	144 (63.7%)

# Trends in false positive p24 EIA tests

---

	2007 3 <sup>rd</sup> gen	2008 Ag/Ab
p24 Ag EIA (NT <50%)	10 (0.0034 %)	9 (0.0030 %)

# Detection of acute HIV infection

---

	2007 3 <sup>rd</sup> gen	2008 Ag/Ab
Ab (WB) Neg /Ag Pos	12	22
WB Ind /Ag Pos	22	29

# Acute HIV infection

Case 644120

53 y. old male presenting with AHI symptoms , ART initiated on day 16

Day	Ag/Ab	WB	p24 Ag	LIA
0	52.2	NEG	POS	
10	6.9	IND p24+,p18+	POS	
26	6.1	IND gp160±, 55±,p40+,p24+++,p18++	NEG	POS-1
42	4.8	IND gp160± p55± p40+ p24+++ p18±	NEG	POS-1

# Observations

---

In cases of acute HIV infection we noticed:

- ✓ Wide range of S/CO values (1.02 – 52.2) for WB negative/Ag p24 positive specimens,
- ✓ A significant decrease in the S/CO values between sera obtained days apart.



# Transient reduction of reactivity

Case 626973

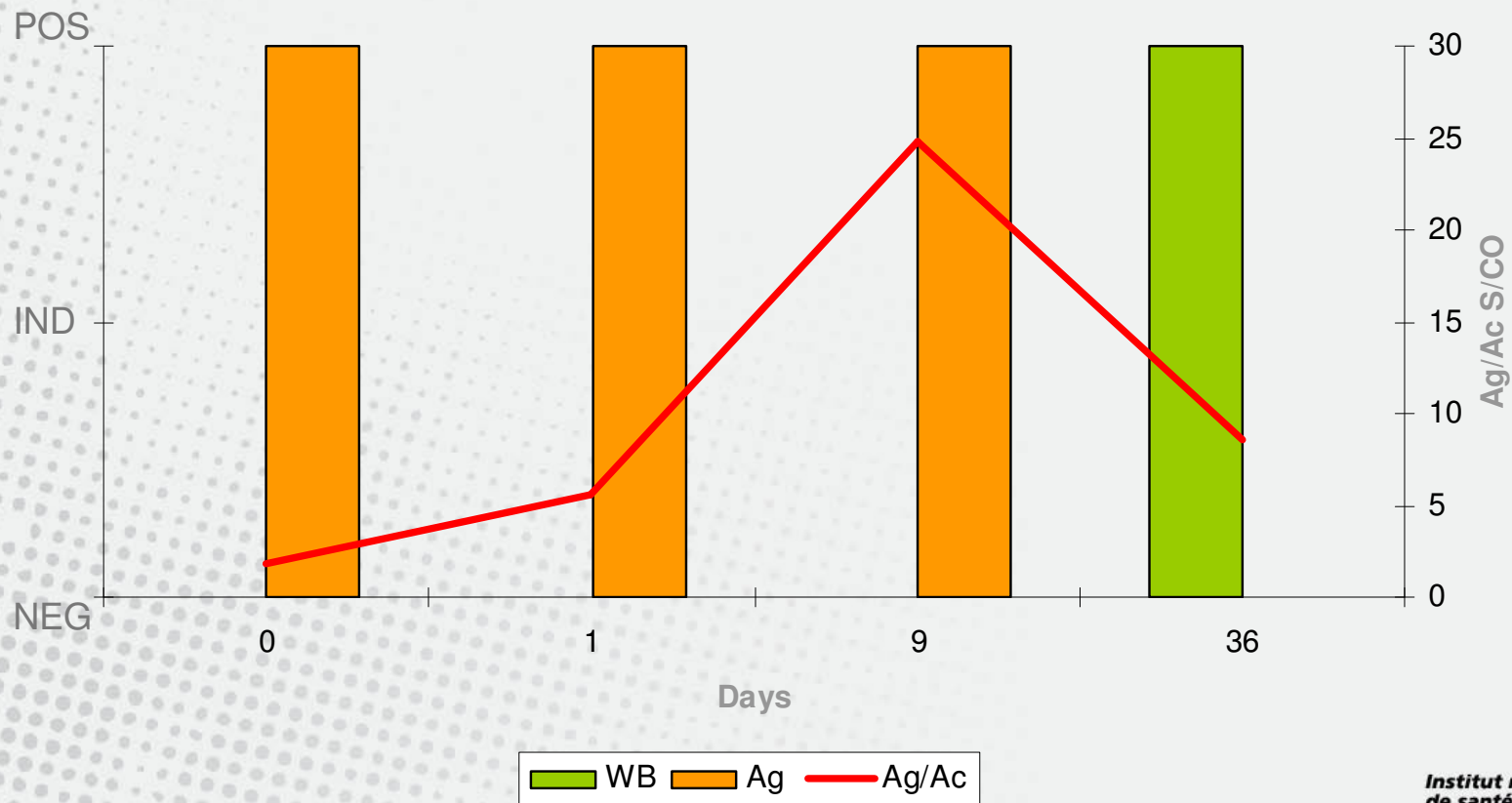
Transition zone between decreasing levels of p24 Ag and increasing levels of Abs



# Transient reduction of reactivity

Case 637587

Transition zone between decreasing levels of p24 Ag and increasing levels of Abs



# Window period exists...

CASE 618350

Day	Ag/Ab	WB	p24
0	< 0.9		
10	6.4	NEG	POS
38	18.6	POS	

... but is reduced compared to 3<sup>rd</sup> gen

CASE 637587

Day	Ag/Ab	3 <sup>rd</sup> gen	WB	p24 Ag
0	1.87		NEG	POS
1	5.6		NEG	POS
7		1.39	NEG	POS
9	24.8		NEG	POS
36	9.13		POS	

# Conclusion

---

Implementation of 4<sup>th</sup> generation assay in routine use in the Québec Provincial program :

- Allowed early identification of individuals with acute infection
- Did not result in more false-positives

# Discussion

---

Ag/Ab detected more patients with acute infection.

- “One size fits all” approach is simple for detection of acute infection, no need for specific request of p24 Ag test.

Confirmatory algorithm must be reviewed.

- Tests to detect HIV RNA or p24 Ag are needed to clarify the status of a Ag/Ab reactive/WB negative sample.

Window period is reduced.

- A second window period can occur.
- Follow-up is suggested for all greyzone reactors.

# Note to laboratory personnel...

---

With Ag/Ab assays,  
sample mismatch  
is no longer  
the most probable explanation  
for a strongly reactive, WB negative  
specimen.

# Acknowledgments

---

- Members of *Programme provincial de diagnostic de laboratoire du VIH*
- Abbott Laboratories provided Ag/Ab kits for sensitivity assessment