



### Abstract (Revised)

**Introduction:** Co-infection with hepatitis C virus (HCV) and human immunodeficiency virus type 1 (HIV-1) is a significant international health problem. We evaluated the prevalence of HCV infection among HIV-infected pregnant women screened for the Breastfeeding, Antiretrovirals, and Nutrition (BAN) Study in Lilongwe, Malawi ([www.thebanstudy.org](http://www.thebanstudy.org)).

**Methods:** Serum from 2,043 HIV-infected pregnant women was screened for HCV using a chemiluminescent immunoassay (CIA) for anti-HCV IgG (Vitros ECI, Ortho CD, Rochester, NY). All specimens with a signal-to-cut-off (S/CO) ratio of  $\geq 1.0$  were considered anti-HCV CIA reactive. All anti-HCV reactive specimens were subject to confirmatory testing with a recombinant immunoblot assay (RIBA HCV 3.0, Novartis V&D, Emeryville, CA) and HCV RNA test. Proportions and confidence intervals were calculated using binomial exact approximations.

**Results:** Of the 2,043 women tested, 110 (5.4%) were anti-HCV reactive; 13 (11.8%) had an S/CO ratio above 8.0. Of the 110 anti-HCV reactive, 109 were tested with RIBA. Only 2 (1.8%, 95% CI: 0.02%-6.4%) were positive, indicative of past or current HCV infection; 28 (25.7%, 95% CI: 17.8% - 34.9%) were indeterminate; and 79 (72.5%, 95% CI: 63.1% - 80.6%) were negative. All anti-HCV positive (110) were HCV RNA negative. The median (IQR) age of study participants was 25 (22-29) years, and the median CD4 count and alanine aminotransferase (ALT) level was 417 (IQR: 300-517) cells/mm<sup>3</sup> and 13 (IQR: 10-15) mg/dl, respectively.

**Conclusions:** Although this population of HIV-infected pregnant women in Malawi demonstrates a high frequency of anti-HCV reactivity (5.4%), the prevalence of confirmed past or current HCV infection is very low (0.1%). The higher HCV reactivity is most likely a false positive result in this low prevalence population.

### Introduction

- Among the 40 million HIV-infected worldwide, 4 - 5 million are also infected with HCV (Alter 2006). Co-infection with HIV has been associated with increased progression to liver disease (Thio 2002).
- Increasing access to ART in resource-limited settings will likely increase occurrence of liver morbidities. Yet, current public health interventions in these settings do not delineate ART treatment based on HCV co-infection (MOHP, 2008).

### Methods



#### Study Population

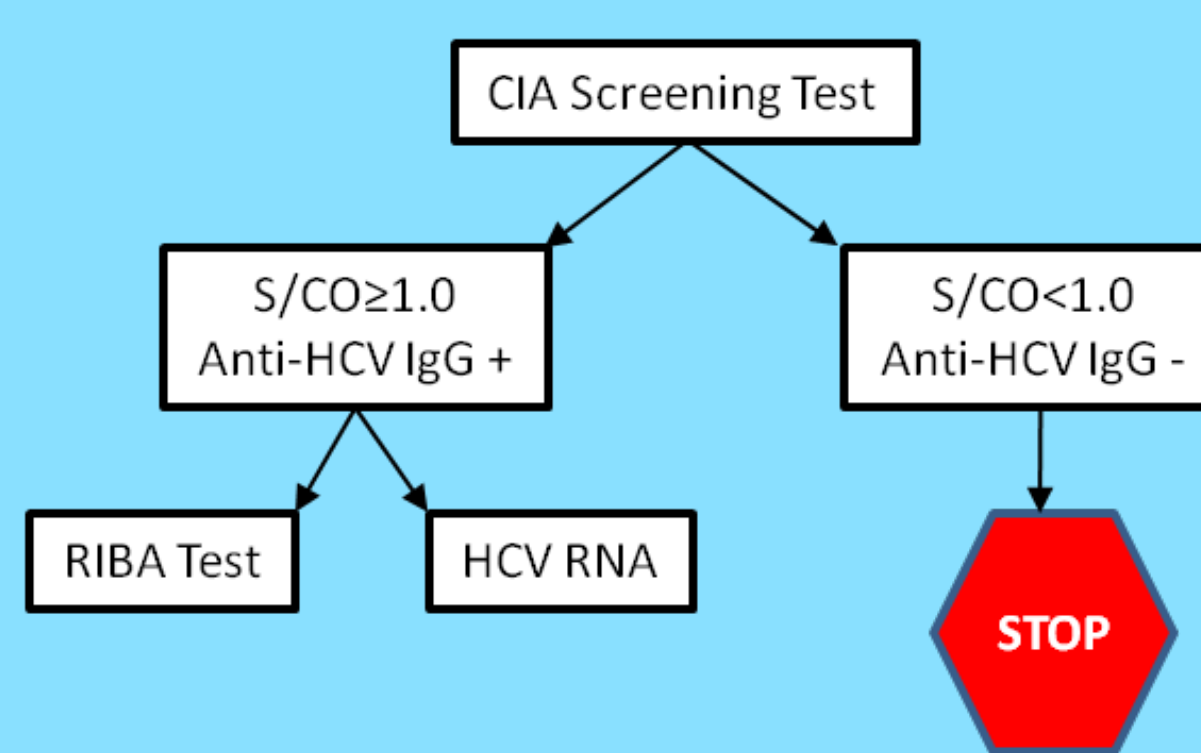
The current study used stored plasma from HIV-positive women screened for enrollment to the Breastfeeding, Antiretrovirals, and Nutritional (BAN) study (van der Horst 2009; Chasela, 2009) who consented to specimen storage and future laboratory studies. We used specimens collected at study screening (<30 weeks gestation), and therefore, prior to study administration of ART for prevention of perinatal HIV transmission.

### Methodology

#### Laboratory Procedures

All HCV testing was conducted in a central CDC laboratory. Anti-HCV IgG testing used a chemiluminescent immunoassay antibody (CIA) screening test (Vitros ECI, Ortho CD; Rochester, NY). All anti-HCV IgG reactive specimens with a Signal Cut off (S/CO) ratio  $\geq 1.0$  were tested with both a recombinant immunoblot assay (RIBA HCV 3.0, Novartis V&D; Emeryville, CA) and HCV RNA test (Figure 1).

Figure 1: HCV testing algorithm.



#### Statistical Analysis

The population characteristics were described with medians (interquartile range) and frequencies. The prevalence and 95% confidence interval of HCV according to the CIA screening and RIBA tests was calculated using binomial exact methods.

### Results

Stored blood specimens were available from 2054 HIV-infected women out of the 3572 women screened for the BAN study. Out of these women, 2043 (99.5%) had sufficient sample and were screened for HCV. The study population was relatively healthy with a median (IQR) CD4 count of 417 (300-571).

Table 1: Baseline characteristics of the HIV infected women screened for HCV (N= 2043)

Characteristics	Median ( Interquartile range [ IQR ] )
Age ( years )	25 (22-29)
CD4 count ( cells/μl )	417 (300-571)
Haemoglobin ( mg/dl )	10.7 (9.8-11.5)
Alanine Aminotrasferase ( mg/dl )	13 (10-15)
Mid Upper arm circumference ( cm )	26 (24.5-27.6)
Triceps Skin fold ( mm )	15.3 (12.3-19.3)

### Results

#### Prevalence of HCV in the study population and the Positive Predictive Value (PPV).

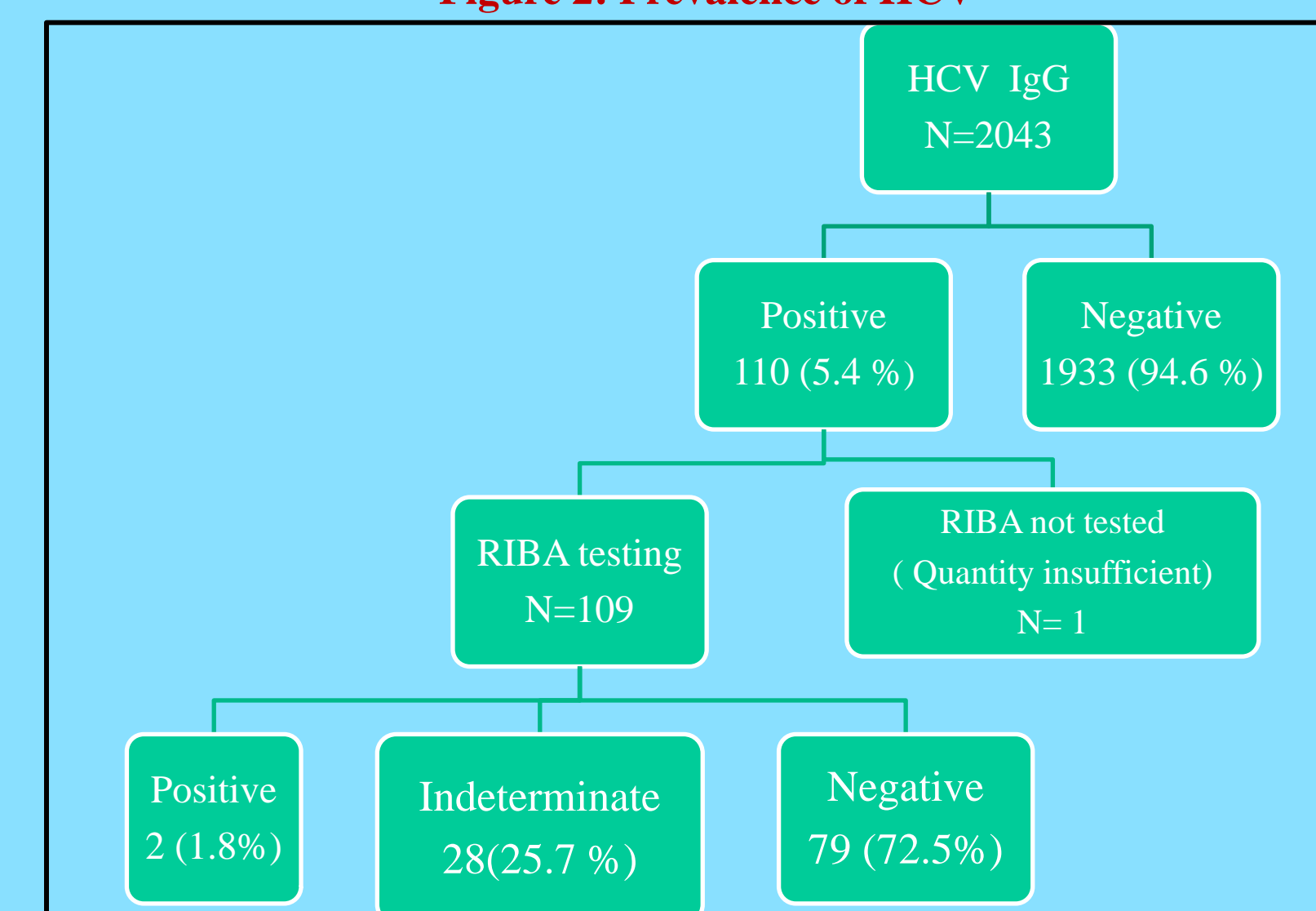
Of the 2043 women screened with CIA, 110 (5.4%, 95% CI: 4.4-6.5%) were anti-HCV IgG positive. Of these, 109 were tested by RIBA, and 79 were negative (72.5%, 95% CI: 63.1-80.6%), 28 were indeterminate (25.7%, 95% CI: 17.8-34.9%), and 2 were confirmed positive (1.8%, 95% CI: 0.02-6.4%) (Figure 2). None of the anti-HCV IgG positive (n=110) were HCV RNA positive. The estimated HCV prevalence in this population was 0.1% (95% CI: 0.01-0.4%). Excluding the indeterminate RIBA results (n=27), the PPV for the CIA screening test was 2.5% (2/81, 95% CI: 0.3-8.5%), indicating a high false positive rate of 97.5% (79/81, 95% CI: 91.4-99.7%). The anti-HCV reactive and non-reactive women were statistically no different in median age, CD4 count, or ALT level (Table 2).

Table 2: Selected variables and HCV status (N= 109)

Variable	Total %	Positive %	Indeterminate %	Negative %	P*
<b>Total</b>	109	2 (1.8)	27 (25.7)	79 (72.5)	
<b>Age (years) median</b>	25	20	25	25	0.52
<b>CD4 count (cells/μl)</b>					
<b>Median</b>	430	562	472	430	0.45
<b>&lt;250</b>	17 (15.6)	0 (0)	3 (10.7)	14 (17.7)	0.89**
<b>250-499</b>	54 (49.5)	1 (50)	14 (50)	39 (49.4)	
<b>≥ 500</b>	38 (34.9)	1 (50)	11 (39.3)	26 (32.9)	
<b>Median ALT</b>	13	29	11.5	13	0.33
<b>Median MUAC (cm)</b>	25	28	25.8	25.5	0.50
<b>Education Level</b>					
<b>No education</b>	13 (11.9)	0 (0)	3 (10.7)	10 (12.7)	0.93**
<b>Primary school</b>	55 (50.5)	1 (50)	13 (46.4)	41 (51.9)	
<b>Secondary/tertiary</b>	41 (37.6)	1 (50)	12 (42.9)	28 (35.4)	

\* Values shown are for the comparison of the HCV positive and negative groups  
\*\* Fisher exact test comparing all groups.

Figure 2: Prevalence of HCV



### Conclusion and Discussion

- The prevalence of HCV was very low (0.1%) in this population. This finding is consistent with other study findings conducted in the region using stringent methodologies (Pirillo 2007, Tess 2000, Menendez 1999).
- The high reactivity of the HCV CIA screening test (5.4%) is most likely a false positive result, a common occurrence in low prevalence areas.

### Reference

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