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Evaluation of HIV-1 Recency Assays Among Prospectively Observed HIV-1 Seroconversions

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Introduction

- The estimation of HIV incidence is a useful measure for evaluating country-specific HIV epidemics and the effectiveness of interventions
- Recency assays use measurements of HIV antibody (Ab) avidity or quantity to determine whether individuals acquired HIV recently
- When incorporated into a recent infection testing algorithm (RITA), recency assays can be used to estimate population-level HIV incidence rates[1-3]
- HIV-1 RITAs using recency assays are currently being used to determine comparator background HIV incidence rates in Phase 3 pre-exposure prophylaxis (PrEP) trials (NCT04994509, NCT04925752, and NCT04652700)
- The DISCOVER study (NCT02842086), a large Phase 3 randomized controlled trial that demonstrated the noninferiority of emtricitabine/tenofovir alafenamide (F/TAF) to emtricitabine/tenofovir disoproxil fumarate (F/TDF) for PrEP, provides a unique context to evaluate the performance of recency assays in well-documented HIV seroconversion cases

Objective

- To evaluate the performance of recency assays in well-documented seroconversion cases from the DISCOVER study

Methods

- The performance of 3 different recency assays was evaluated on 42 uniquely dated plasma samples from 25 participants from the DISCOVER study
  - HIV testing was conducted at screening, baseline, and every 12 weeks during DISCOVER
  - Time in days since infection was determined for each sample by subtracting the date of the sample from the date of the last negative HIV test (Day 0)
  - This duration of time between the positive and negative tests determined recent vs long-term infection, and was used as the reference against which to compare the recency assays
  - Of 25 participants with available samples, 15 had samples from a single visit and 10 from multiple visits over a period allowing evaluation of recent and long-term samples from the same participant

Recency Assays

Recency Platform	Method	Unit of Measurement	IA Threshold	MDRI
LAg-EIA*	Ab avidity, EIA	Normalized ODn	1.5 ODn	130 d[4]
ARCHITECT†	Ag/Ab chemiluminescent IA	Signal–cutoff ratio (S/CO)	200 S/CO	186 d[5]
Asante‡	Ab avidity, lateral flow IA, interpreted with electronic reader	LT/R band intensity	3.0 LT/R	180 d[6]

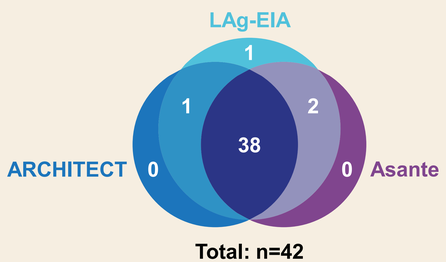
*Sedia® HIV-1 Limiting Antigen Avidity EIA (Sedia Biosciences Corporation, Beaverton, OR); †Abbott™ ARCHITECT™ HIV Ag/Ab Combo assay (Abbott, Abbott Park, Chicago, IL); ‡Asante® HIV-1 Rapid Recency® Assay (Sedia). Ag, antigen; EIA, enzyme immunoassay; IA, immunoassay; MDRI, mean duration of recent infection; ODn, optical density.

- The determination of recent or long-term infection was based on assay-specific IA threshold and MDRI
  - IA threshold: assay result cutoff defining recent infection (below IA = recent; above IA = long term)
  - MDRI: average time post-infection that individuals were classified as recently infected; differs based on the assay used
- The number of days since infection determined for each DISCOVER sample was compared with the MDRI for each assay to make the recent vs long-term determination

Results

Samples Evaluated by Assay

Samples Evaluated in:	LAg-EIA	ARCHITECT	Asante	Samples Tested
3 recency assays	38	38	38	38
2 recency assays				
LAg EIA + ARCHITECT	1	1		1
LAg EIA + Asante	2		2	2
1 recency assay				
ARCHITECT		1		1
Total	41	40	40	42



- Of 42 samples, 38 were tested by all assays, 3 were available for testing by 2 assays, and 1 sample was tested by only 1 assay
- Samples were predominantly subtype B (22/25 participants); 2 participants were subtype F1 and 1 was AG

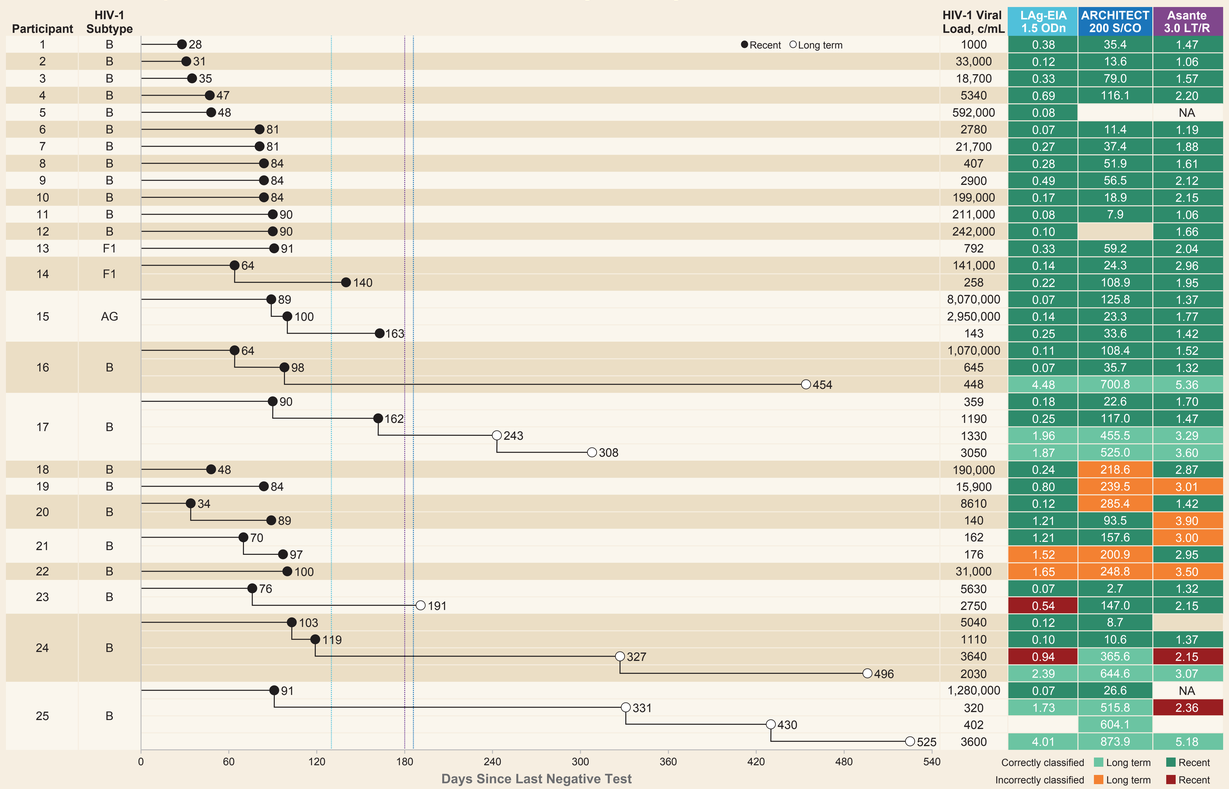
Recency Assay Results

	LAg-EIA n=41	ARCHITECT n=40	Asante n=38*
Classified correctly, n (%)	37 (90)	35 (88)	32 (84)
Classified incorrectly, n (%)	4 (10)	5 (12)	6 (16)
Recent called long term, n	2	5	4
FRR (long term called recent), n	2	0	2

*2 samples were called negative for HIV-1 by Asante. FRR, false recency rate.

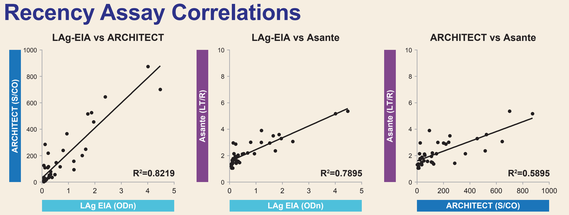
- LAg-EIA correctly classified the most samples (90%); all assays correctly classified >84% of samples
- Similar numbers of incorrectly classified samples were seen between assays (10–16%)
- Of the 3 samples that were incorrectly classified as recent, 1 was incorrectly classified by 2 assays; all participants had documented history of antiretroviral use (on antiretroviral therapy after diagnosis) while in the trial
- Of the 5 participants' samples that were incorrectly classified as long term, 2 had samples from 2 different visits that were incorrectly classified by ≥1 assay, possibly suggesting a participant-specific attribute
- Of the 3 remaining incorrectly classified samples, 2 were incorrectly classified as long term by 2 assays

Timeline of Samples Since Infection and Classification by 3 Assays*



*Day 0 is date of last negative HIV test; numbers to *right of circles* are days since last negative test; MDRI for LAg-EIA in *light blue dotted line*, ARCHITECT in *dark blue dotted line*, and Asante in *purple dotted line* demonstrating cutoff for recent vs long-term infections for each assay; any samples that were close to MDRI cutoff were considered recent as infection most likely did not occur at Day 0; *raw* IA call found in *right columns* for each assay; *blank cells* indicate samples were not tested by assay. NA, not applicable (Asante assay determined sample to be HIV-1 negative so assay call is NA).

Recency Assay Correlations



- Strong correlations were seen between LAg-EIA and ARCHITECT, and Asante
- Less correlation was seen between ARCHITECT and Asante

Conclusions

- LAg-EIA, Asante, and ARCHITECT recency assays were able to distinguish between recent and long-term infections observed during the DISCOVER study
- All 3 assays identified the recent infections with similar high degrees of accuracy
- While HIV incidence was unable to be determined with these assays in sites where DISCOVER was conducted due to lack of appropriate screening of the entire population, use of well-documented seroconversion samples from the DISCOVER study allowed for a thorough analysis of 3 recency assays
- Overall, these analyses support the use of recency assays in determining the counterfactual background HIV incidence rates in future PrEP trials

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