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Reasons for HIV testing in women diagnosed with HIV (2000–2023): insights from a 24-year retrospective cohort study in Barcelona, Spain

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Abstract

Background Determining the reasons for HIV test performance in women living with HIV (WLWH) is crucial for evaluating areas for improvement in early diagnosis.

Methods The primary objective of the study was to define the reasons for HIV testing in cisgender WLWH who had their first visit in Hospital Clínic, Barcelona, between January 2000 and December 2023. Reasons were classified into: pregnancy and preconception care, AIDS-defining events (ADE), diagnosis in a sexual partner, indicator condition (IC), patient-initiated testing, physician-initiated testing, and "other". Secondary objectives included the proportion of late and advanced diagnosis, and analysing viral load (VL), CD4 count, and patient status at 48 weeks. Data were compared between two periods (January 2000 to December 2014 and January 2015 to December 2023).

Results The study included 537 women. The main reasons for HIV testing were pregnancy and preconception care (24%) and indicator conditions (22%). Overall, late and advanced diagnoses were observed in 56% and 32% of women, respectively. As expected, women diagnosed following an AIDS defining event presented with the lowest CD4 counts, while those tested due to an indicator condition also showed high proportions of late (66%) and advanced (43%) diagnoses. Spanish women had a lower risk of late diagnosis compared with non-Spanish women, particularly when diagnosed during pregnancy or preconception care, 0.39 (95% CI: 0.18–0.87, $p = 0.021$). At 48 weeks, CD4 counts remained below 400 cells/mm^3 in late-diagnosed women across both periods.

Conclusion These findings underscore the importance of targeted strategies to promote earlier HIV detection and improve long-term health outcomes in women.

Keywords HIV, Prevention, Women, Test, HIV testing, Early diagnosis

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Text box 1. Contributions to literature

- Evidence on reasons for HIV testing among women in low prevalence, male dominated epidemics is scarce; this study provides 24 years of real-world data from a European setting.
- The findings highlight persistent gaps in early HIV diagnosis among women, despite universal antenatal screening and expanded testing recommendations.
- The study shows that indicator-condition-guided testing remains under implemented, contributing to high rates of late and advanced diagnosis.
- Results underscore the need for gender responsive HIV testing strategies that address structural barriers, provider-initiated testing gaps, and disparities by region of origin.

Background

Late HIV diagnosis remains a major public-health challenge worldwide. In the WHO European Region, more than half of the people newly diagnosed with HIV in 2023 were diagnosed late (CD4 < 350 cells/mm^3 or diagnosis with an AIDS-defining event, regardless of CD4 count) [1].

Interventions to improve early diagnosis include promoting sexual-health literacy in the general population and guidelines for general practitioners and emergency department healthcare professionals to identify HIV-related indicator conditions [2].

In countries where the HIV epidemic disproportionately affects men, such as Spain, women are often underrepresented in targeted prevention strategies.

Understanding the reasons for HIV testing is crucial for identifying areas for improvement in early diagnosis [3].

Late and advanced HIV diagnoses are key health indicators, typically defined according to CD4 thresholds at diagnosis [4]. These categories reflect missed opportunities for earlier testing and are associated with poorer clinical outcomes, delayed access to treatment, and increased morbidity.

The present study aimed to analyze the main reasons for HIV screening among women living with HIV (WLWH) who had their first visit from January 2000 to December 2023 at Hospital Clinic, a tertiary hospital in Barcelona, Spain; to describe the proportions of late and advanced disease stages at diagnosis by reason; and to compare trends between two time periods.

Methods

This is a descriptive, retrospective study of cisgender women living with HIV (WLWH) who had their first visit between January 1, 2000, and February 5, 2024 (database closure), at the HIV Unit of Hospital Clínic (Barcelona, Spain). Women were included if they had documented reasons for HIV testing and at least two viral load measurements (the initial one and another within 14 months). Although patient selection extended to February 5, 2024, no new diagnoses were recorded in 2024.

Clinical records were evaluated and double-checked by two clinical investigators.

Reasons for HIV testing were grouped into seven categories: pregnancy and preconception care (including first-trimester antenatal HIV screening and consultations for women seeking pregnancy), AIDS-defining event (ADE), diagnosis in a sexual partner, indicator condition, non-ADE (IC), patient-initiated testing, physician-initiated testing, and other reasons. The indicator-condition category (IC) was subdivided according to the definitions provided in the HIV Indicator Condition Guidelines developed by the HIV in Europe initiative. (Members to the panel on Guidance on Indicator Condition- Guided HIV testing in Adults & WHO and the European Centre for Disease Prevention and Control (ECDC), 2012).

Epidemiological and laboratory data were extracted from the electronic medical record system of the HIV Unit. An electronic case report form was also created in REDCap Software [5, 6], hosted at the Hospital Clinic, to systematically collect information on the reasons for HIV testing.

We also analyzed the following variables across testing-reason categories: age at diagnosis, CD4 count at diagnosis, and place of birth. The diagnosis period was categorized into two intervals: period 1 (January 2000 to December 2014) and period 2 (January 2015 to December 2023). These periods reflect the implementation of universal treatment regardless of CD4 count, which became standard practice around 2015. Late HIV diagnosis was defined as a first HIV diagnosis with a CD4 count < 350 cells/mm^3 or with an AIDS-defining event, regardless of CD4 count, according to the 2022 EuroTEST consensus definition [4]. Individuals with documented primary HIV infection who presented with a CD4 count < 350 cells/mm^3 were excluded from all analyses that included CD4 count.

Qualitative variables were expressed as frequencies and percentages. Quantitative variables were expressed as median and interquartile range (IQR). We used multinomial logistic regression with 'reason for testing' as the dependent variable and pregnancy and preconception care as the reference group. Results are presented as relative-risk ratios (RRRs). Relative Risk Ratio (RRR) is defined as the exponentiated coefficient e(b), representing the ratio of the probability of each outcome category relative to the reference category. Logistic regressions were performed with late and advanced diagnosis as dependent variables to show the possible differences in these variables between reasons for testing, place of birth, and the period of diagnosis, and odds ratios (OR) were reported. Data regarding HIV VL, CD4 count, and continuum of care were analyzed at 48 weeks of

follow-up after the first HIV visit to the hospital for ART-naïve women and for those patients whose baseline CD4 count was reported in the clinical records and could be confirmed. ART-naïve women were defined as those who had not received antiretroviral therapy before their first visit to the Hospital Clinic. Logistic regression was used for binary outcomes and linear regression for continuous outcomes. Age was included as an adjustment variable because patients with late or advanced diagnosis were significantly older than those without, and age was therefore considered a potential confounder in the association between mode of diagnosis and diagnostic stage.

The tests were two-tailed, and the significance level was set at 5%. Stata 18 software was used for statistical analyses [7].

Results

Of the 660 eligible women, 123 lacked information on the reason for HIV testing; therefore, 537 WLWH were included.

Baseline characteristics

The median age at HIV diagnosis was 34 years (IQR 28–42). Among women with available country-of-birth data ($n=534$), 52% (276/534) were from Spain, 29% (154/534) from Latin America and the Caribbean, 11% (61/534) from Africa, and 7% (36/534) from other countries in Europe except for Spain. 83% (445/537) of women were ART-naïve at their first visit to the center. The remainder had been diagnosed elsewhere and were already receiving ART at first visit.

Table 1 Baseline characteristics of women diagnosed with HIV at Hospital Clínic Barcelona, 2000–2023

Variable		Period of diagnosis		Total
		2000–2014 $n=440$	2015–2023 $n=97$	$n=537$
Age (median, IQR)		33 (27; 41)	37 (28; 43)	34 (28; 42)
Origin	Spain	247 (57%)	29 (30%)	276 (52%)
	Latin America	112 (26%)	42 (43%)	154 (29%)
	Africa	49 (11%)	12 (12%)	61 (11%)
	Europe	26 (6%)	10 (10%)	36 (7%)
	Other origin	3 (1%)	4 (4%)	7 (1%)
	Total	437* (100%)	97 (100%)	534 (100%)
First CD4** (median, IQR)		314(157; 529)	266 (149; 497	300 (156; 529)
First VL** (median, IQR)		27,624 (4700; 158000)	22,950 (3380; 118000)	25,000 (4600; 158000)
Naïve at first visit		363 (82%)	82 (85%)	445/537 (83%)

Abbreviations: *IQR* interquartile range, *VL* viral load

*3 women did not have information about country of birth (origin)

**CD4 count and HIV viral load at diagnosis were calculated only for ART-naïve women, as specified in the Methods section

When comparing the periods 2000–2014 and 2015–2023, the median age at diagnosis increased from 33 years (IQR: 27–41) to 37 years (IQR: 28–43). In 2000–2014, 62% of women were from Europe (including 57% from Spain), whereas in 2015–2023 this proportion decreased to 40% (30% from Spain). In contrast, women from Latin America and the Caribbean accounted for 26% in the first period, increasing to 43% in the second. The proportion of women from Africa remained relatively stable across periods (11% vs. 12%). The percentage of ART-naïve women did not change across periods (Table 1).

Reason for HIV test

Overall, the main reason for HIV testing in both periods was pregnancy and preconception care (24%; 128/537), followed by indicator conditions (22%; 120/537), physician-initiated testing (19%; 100/537), diagnosis in a sexual partner (18%; 94/537), AIDS-defining events (10%; 56/537), other reasons (4%; 24/537), and patient-initiated testing (3%; 15/537). Other reasons are detailed in Supplementary Table 1.

The most frequent ICs were generalized lymphadenopathy (25%; 30/120), unexplained weight loss or toxic syndrome (11%; 13/120), and unexplained fever (8%; 9/120).

As for the ADEs, the most frequent diagnoses were Pneumocystis jirovecii pneumonia (29%; 16/56), Mycobacterium tuberculosis infection (25%; 14/56), cerebral toxoplasmosis (16%; 9/56), esophageal candidiasis (9%; 5/56), chronic cryptosporidiosis (4%; 2/56), and recurrent bacterial pneumonia (4%; 2/56). Other opportunistic diseases each accounted for < 2%.

The proportion of diagnoses attributed to pregnancy and preconception care and to indicator conditions remained stable across periods. In contrast, diagnoses following a partner's diagnosis increased from 16% in 2000–2014 to 25% in 2015–2023. Patient-initiated testing also increased, from 2% to 4%. Conversely, ADE-related diagnoses decreased from 11% to 8%. After age adjustment, physician-initiated testing was significantly less likely in the later period (RR: 0.55; 95% CI: 0.31–0.99), with no significant differences in other categories. (Fig. 1).

Immunological status at HIV diagnosis

For immunological analyses, we included data from women with available baseline CD4 counts. Five women diagnosed during primary infection with CD4 < 350 cells/mm³ were excluded, as these counts were considered transiently low due to acute infection.

Median CD4 count at diagnosis was 300 cells/mm^3 (IQR 157–529). The best immunovirological status was observed in women in whom the HIV test was carried out on their own initiative (median CD4 count of 436 cells/mm^3 [IQR 389–567], followed by HIV test

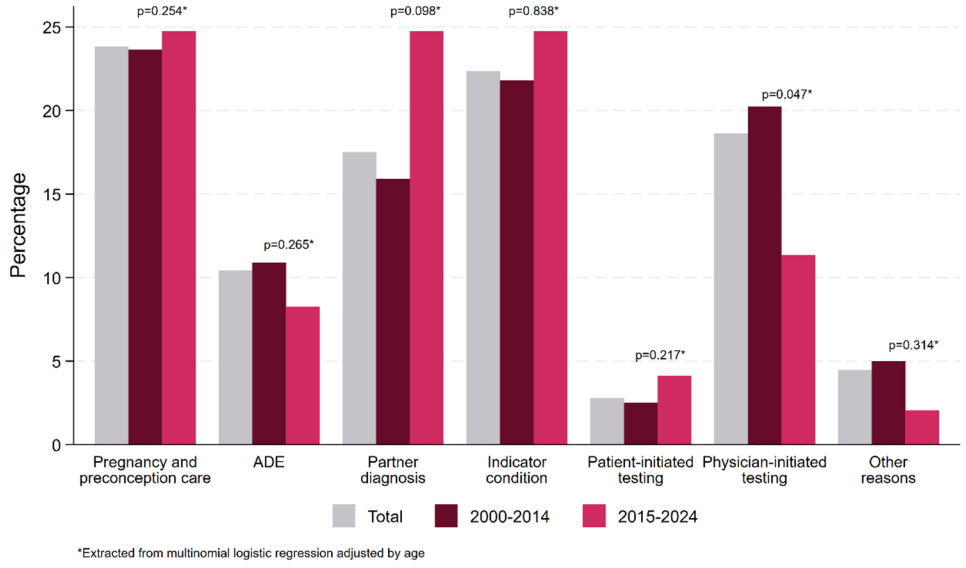

Fig. 1 Distribution of reasons for HIV testing among women diagnosed with HIV at Hospital Clínic Barcelona, 2000–2023. Abbreviations: ADE: AIDS-defining event

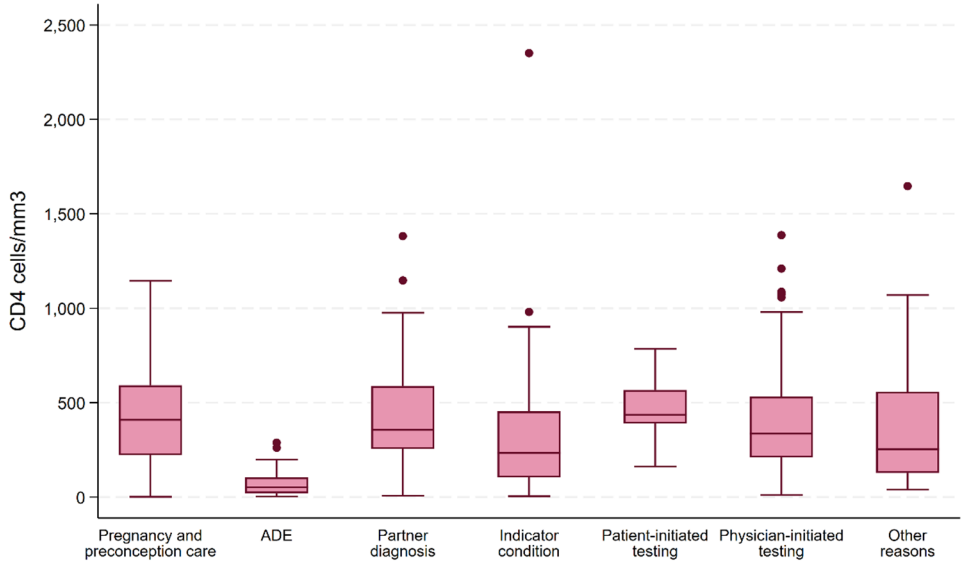

Fig. 2 CD4 cell count at HIV diagnosis by reason for testing among women at Hospital Clínic Barcelona, 2000–2023. Data was available for 459 patients. Box-Whiskers plot. Box lines represent the 25th (Q1), 50th (median), and 75th (Q3) percentiles. The whiskers represent the lower and upper adjacent values (i.e., lowest and highest observations within the range defined as follows: Q1 – 1.5×IQR for the lower limit; Q3 – 1.5×IQR for the upper limit). Dots represent outliers. Abbreviations: IQR: interquartile range; ADE: AIDS-defining event

performed due to pregnancy and preconception care (CD4 count 409 cells/mm^3 [IQR 222.5–592], diagnosis in a sexual partner (CD4 count 356 cells/mm^3 [IQR 255–588], physician-initiated testing (CD4 count 336 cells/mm^3 [IQR 210–532]), 'other reasons' (CD4 count 253 cells/mm^3 [IQR 127.5–557.5]), indicator conditions (CD4 count 234 cells/mm^3 [IQR 104–446], and presence of an ADE (51 cells/mm^3 [IQR 20–104]) (Fig. 2).

Late diagnosis (CD4 < 350 cells/mm^3) was observed in 57% (264/459) of women with available baseline CD4. When the reason for testing was the presence of an indicator condition, this percentage increased to 66% (66/100) and for the groups "physician-initiated-testing", "diagnosis in a sexual partner" and "pregnancy and preconception care", the percentages were 51% (42/82), 48% (41/86), and 43% (48/111), respectively. Among women who underwent testing on their own initiative, only 13% had a late diagnosis (2/15). As expected, all ADE-related diagnoses were late ($n = 49$) (Fig. 3).

Advanced diagnosis (CD4 < 200 cells/mm^3) occurred in 32% (150/459). By category: ADE, 96% (47/49); indicator condition, 43% (44/100); other reasons, 38% (6/16);

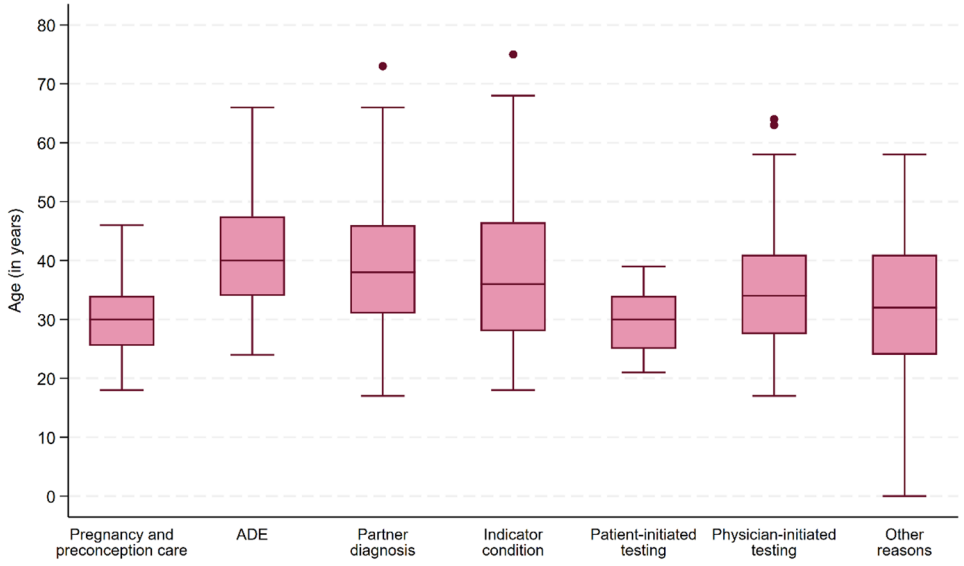

Fig. 3 Age at HIV diagnosis by reason for testing among women at Hospital Clínic Barcelona, 2000–2023. Box-Whiskers plot. Box lines represent the 25th (Q1), 50th (median), and 75th (Q3) percentiles. The whiskers represent the lower and upper adjacent values (i.e. lowest and highest observations within the range defined as follows: Q1 – 1.5×IQR for the lower limit; Q3 – 1.5×IQR for the upper limit). Dots represent outliers. Abbreviations: IQR: interquartile range; ADE: AIDS-defining event

Table 2 Age-adjusted regression models for late and advanced HIV diagnosis by reason for testing among women at Hospital Clínic Barcelona, 2000–2023

Late diagnosis (CD4 < 350 cells/mm^3)	**RRR**	**[95% CI]**
Pregnancy and preconception care	1	
Partner diagnosis	0.80	[0.43, 1.47]
Indicator condition	1.77	[0.98, 3.19]
Patient's initiative	0.21	[0.04, 0.98]
Physician's initiative	1.07	[0.59, 1.94]
Other	2.16	[0.68, 6.81]
Advanced diagnosis (CD4 < 200 cells/mm^3)	**RRR**	**[95% CI]**
Pregnancy and preconception care	1	
Partner diagnosis	0.40	[0.18, 0.90]
Indicator condition	2.07	[1.09, 3.93]
Patient's initiative	0.29	[0.04, 2.35]
Physician's initiative	0.76	[0.37, 1.58]
Other	1.68	[0.53, 5.27]

Abbreviations: *RRR* relative risk ratio, *CI* confidence interval

pregnancy and preconception care, 21% (23/111); physician's initiative, 21% (17/82); diagnosis in a sexual partner, 14% (12/86); and patient's initiative, 7% (1/15).

Because women with late or advanced diagnosis were significantly older than those diagnosed earlier, age acted as a potential confounder in the association between diagnostic category and diagnostic stage. Therefore, all multivariable analyses were adjusted for age to isolate the independent effect of the reason for HIV testing. For transparency, unadjusted models and supplementary figures illustrating the number of women with late or advanced diagnosis across diagnostic categories by age are provided in the Supplementary Material (Supplementary Table 2, Supplementary Figs. 1 and 2).

Compared with pregnancy and preconception care (reference category), only patient-initiated testing was associated with a lower risk of late diagnosis (RRR: 0.21; 95% CI: 0.04–0.98). For advanced diagnosis (CD4 < 200), testing prompted by an indicator condition was associated with a higher risk of advanced disease compared with pregnancy and preconception care (RRR: 2.07; 95% CI: 1.09–3.93) (Table 2).

No differences in the distribution of testing reasons were observed by place of birth. Spanish-born women had lower odds of late and advanced diagnosis than non-Spanish women, particularly within pregnancy and preconception care category: late diagnosis OR: 0.39 (95% CI: 0.18–0.87) and advanced diagnosis OR: 0.28 (95% CI: 0.09–0.82).

Follow-up at 48 weeks after HIV diagnosis

We examined three outcomes at 48 weeks from the first HIV visit among ART-naïve women—viral load, CD4 count, and retention in care—across the two periods and by baseline CD4 strata.

During the 2000–2014 period, women with late diagnosis had a significantly higher percentage of undetectable viral load at 48 weeks compared to those without late diagnosis (80% vs. 45%; OR: 4.69; 95% CI: 2.77–7.93). This was likely influenced by the ART initiation criteria based on CD4 count at the time, as in the 2015–2023 period, no significant differences were observed between

Table 3 Linear regression models of CD4 cell count at 48-week follow-up by late and advanced HIV diagnosis among women at Hospital Clínic Barcelona, 2000–2023

Period	Late diagnosis (CD4 < 350 cells/mm^3)			
	No	Yes	Coefficient	[95% CI]
2000–2014	585.5 (462; 842)	362 (238; 511)	-266.18	[-324.32; -208.04]
2015–2023	805 (653; 1078)	312.5 (232; 462.5)	-488.61	[-617.23; -360.0]
Period	**Advanced diagnosis (CD4 < 200 cells/mm^3)**			
	No	**Yes**	**Coefficient**	**[95% CI]**
2000–2014	528 (427; 756)	272.5 (183; 395.5)	-291.22	[-353.25; -229.18]
2015–2023	653 (501; 880)	264 (218; 318)	-426.97	[-551.23; -302.71]

Abbreviations: *CI* confidence interval

late and non-late diagnosis groups (83 vs. 89%; OR: 0.56; 95% CI: 0.13–2.42).

In terms of immunological recovery, women with late diagnosis consistently had lower CD4 counts at 48 weeks compared to those without late diagnosis across both periods. For advanced diagnosis, similar trends were observed (Table 3).

Retention at 48 weeks was higher among late presenters in 2000–2014 (92% vs. 81%), but similar in 2015–2023 (96% vs. 97%).

Discussion

This retrospective study aimed to analyze the main reasons for HIV testing among women living with HIV (WLWH) and to describe the proportions of late and advanced disease stages at diagnosis based on these reasons. The study focused on WLWH visiting the HIV unit of a tertiary hospital in Barcelona, Spain, between 2000 and 2023, and compared trends between 2000–2014 and 2015–2023.

While there is limited literature on reasons for HIV testing among people living with HIV (PLWH) [8, 9], more emphasis has been placed on understanding the general population's willingness to undergo testing and developing strategies to improve test uptake [10, 11]. Insights from these key points can help identify gaps in testing performance and devise interventions to promote testing.

In the European Union, where the male-to-female ratio of HIV infection was 2.7 in 2023, women are often diagnosed later than men [1]. This pattern may reflect lower testing frequency and lower perceived risk among women.

Of the 537 women included in our study, over one in five diagnoses occurred during pregnancy or preconception care—mostly at routine first-trimester screening—highlighting antenatal care as a key entry point for timely diagnosis and ART initiation. Despite guideline recommendations [12], antenatal HIV screening is not universally implemented. Systematic screening in pregnancy enables timely diagnosis and rapid ART initiation in women who might otherwise be missed [13], and it is crucial to prevent vertical transmission.

Over time, we observed a notable decline in HIV diagnoses initiated by physicians, along with a reduction in diagnoses following AIDS-defining events. These trends may reflect a shift towards earlier detection and a reduced burden of advanced disease at presentation in Spain. However, the significant decrease in physician-initiated testing raises concerns about missed opportunities for diagnosis, particularly in women who may not perceive themselves at risk. At the same time, the increase in tests performed due to a partner's diagnosis and, to a lesser extent, patient-initiated testing—although not statistically significant—may indicate greater empowerment and reduced HIV stigma [14]. In both groups—those diagnosed due to a partner's diagnosis or through their own initiative—the likelihood of late diagnosis was significantly lower, suggesting the importance of these testing pathways in promoting earlier detection. In the same way, the downward trend in tests following AIDS-defining events, although not significant, could indicate improvements in early detection and intervention strategies, potentially reducing disease progression. However, more than half of the women in our study were diagnosed late, consistent with WHO data showing high rates of late diagnosis globally and in Europe [1]. This trend emphasizes the need for gender-specific healthcare strategies, as late presentation impacts clinical outcomes and epidemic control [15–19].

European guidelines recommend routine HIV testing for patients with non-AIDS conditions, known as indicator conditions (ICs), that are conditions more commonly found in people with HIV than in people without HIV, to facilitate timely diagnosis [20]. However, women diagnosed with indicator conditions in our study showed the highest percentage of late and advanced disease. We hypothesized that many ICs prompting tests in our study were associated with advanced disease, such as generalized lymphadenopathy and unexplained weight loss, while milder ICs might not have been screened. In fact, the HIDES 2 study showed low testing rates for well-established HIV ICs across Europe [21] and a more recent study showed low coverage of HIV indicator condition guidelines also in Europe [22].

Our logistic regression analysis did not reveal statistically significant changes in late diagnosis percentages between 2000 and 2014 and 2015–2023. Despite some improvements mentioned above - such as fewer diagnoses after AIDS-defining events and more tests following

a partner's diagnosis or initiated by the patient - the proportion of late diagnoses remained high, indicating that pathway shifts were insufficient to impact late presentation at population level within this setting. This finding underscores the need for continued efforts to promote earlier testing and better implementation of screening strategies, particularly those targeting women.

The 48-week follow-up analysis of women diagnosed with HIV in our center reveals both encouraging and concerning trends. On the positive side, a significant proportion of patients diagnosed at late and advanced stages achieved an undetectable viral load (< 50 copies/mL) by 48 weeks, reflecting the effectiveness of modern ART. Additionally, the high rate of patients remaining actively engaged with the healthcare system underscores the success of retention strategies. This observation is consistent with the results described in a recent publication performed in a cohort study in Spain between 2006 and 2020, with a maximum rate of 8.5% per year of people living with HIV who were lost to follow-up during this period [23]. This continued engagement is crucial for long-term health outcomes and preventing disease progression.

As expected, CD4 recovery during the first year remained limited in women diagnosed at late or advanced stages. Although most women achieved and maintained virologic suppression, which provides additional immunological protection, persistently low CD4 counts still carry prognostic implications and may increase vulnerability to opportunistic infections and other HIV-related complications despite effective ART [24, 25]. This pattern is consistent with current evidence and guideline recommendations, which acknowledge that virologic suppression mitigates, but does not fully eliminate, the risks associated with low CD4 counts.

The study's strength lies in its focus on women and reasons for testing in a setting where they represent a low percentage of PLWH compared to men. However, as a single-center study, results may not be extrapolated to areas with different HIV prevalence and cultural factors. Other limitations are potential residual confounding, and lack of a male comparison group.

Conclusions

Pregnancy and preconception care and indicator conditions were primary reasons for HIV diagnosis in WLWH. Late diagnosis rates were high across all groups. Beyond the low proportion of women's initiative for testing, our findings indicate that they are frequently omitted from provider-initiated HIV testing. Efforts to promote earlier testing and diagnosis in women should include targeted interventions to improve access, increase awareness, and address stigma, especially where women are a lower percentage of PLWH than men.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s13690-026-01913-3.

Supplementary Material 1: Supplementary Table 1. Category 'Other reasons' for HIV testing among women diagnosed at Hospital Clínic Barcelona, 2000–2023. Supplementary Table 2. Unadjusted multinomial regression models for late and advanced HIV diagnosis by reason for testing among women at Hospital Clínic Barcelona, 2000–2023. Figure 3. Age at HIV diagnosis by reason for testing among women at Hospital Clínic Barcelona, 2000–2023. Box-Whiskers plot. Box lines represent the 25th (Q1), 50th (median), and 75th (Q3) percentiles. The whiskers represent the lower and upper adjacent values (i.e. lowest and highest observations within the range defined as follows: Q1−1.5×IQR for the lower limit; Q3−1.5×IQR for the upper limit). Dots represent outliers. Supplementary Figure 1. Age distribution of women with and without late HIV diagnosis by reason for testing at Hospital Clínic Barcelona, 2000–2023.

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Authors' contributions

ST , JM and BT designed the study, interpreted and discussed the data, and wrote the initial manuscript.ST and BT revised and cleaned the patient data. LB designed the study, performed data analysis, interpreted and discussed the data, and wrote the initial manuscript. AGC, AI, LM, MMR, ML, JA, AF, IC, JC, EL, JLB, EM and JMM interpreted and discussed data analysis, critically revised the initial manuscript and contributed with changes and improvements. EF collected patients' data, revised the initial manuscript and contributed with changes and improvements.

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Data availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

Declarations

Ethics approval and consent to participate

This study was approved by the Institutional Review Board (IRB) of Hospital Clínic.

Consent for publication

Not applicable.

Competing interests

The authors declare no competing interests.

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