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MAJOR ARTICLE

Risk of Developing Low-Level Viral Rebound Among People With HIV Receiving 2- or 3-Drug Regimens: A Case-Control Study Nested in the ICONA Cohort

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Background. The risk of developing low-level viral rebound (LLVR) after achieving human immunodeficiency virus HIV-1 (HIV) RNA suppression with 2-drug regimens (2DR) compared to 3-drug regimens (3DR) remains uncertain.

Methods. We conducted a matched 1:3 case-control study nested within the ICONA cohort including persons with HIV (PWH) who achieved virological suppression (≥2 consecutive HIV-RNA ≤50 copies/mL over 6 months) after 11 November 2014 (baseline [BL]). Cases were defined as PWH experiencing a single HIV-RNA of 51–199 copies/mL after BL (index date); controls were defined as those who maintained HIV-RNA ≤50 copies/mL up to the index date and were matched for history of care gaps and number of drugs failed. The cumulative incidence of LLVR after virological suppression was estimated using Kaplan–Meier methods, and conditional logistic regression models were used to evaluate the association between the current antiretroviral therapy regimen (2DR [dolutegravir/lamivudine, dolutegravir/rilpivirine, dolutegravir/doravirine, or cabotegravir/rilpivirine] vs 3DR [dolutegravir, bictegravir, rilpivirine, doravirine, boosted darunavir, or boosted atazanavir–based with a backbone of tenofovir and lamivudine or emtricitabine]) and LLVR risk. In sensitivity analyses cases were defined as PWH experiencing 2 consecutive HIV-RNA of 51–199 copies/mL.

Results. Among 1033 PWH (261 cases, 772 matched controls), 21% were female, median age was 43 (interquartile range [IQR], 34–51) years, and BL CD4 count was 601 (IQR, 379–826) cells/μL; 2DR use was 25% in cases versus 29% in controls ($P = .23$). Two years after viral suppression, cumulative LLVR incidence was 2.7% (95% CI, 2.3%–3.1%) when considering single LLVR events and 1.8% (95% CI, 1.4%–2.1%) when limited to 2 consecutive values. After adjusting for confounding, evidence for an association with current regimen was inconclusive (adjusted odds ratio, 0.86 [95% CI, .57–1.29]).

Conclusions. Despite the wide range of plausibility, we can exclude a risk of LLVR higher than 29% when using 2DR versus 3DR regimens.

Keywords. 2-drug regimen; 3-drug regimen; HIV; low-level viral rebound; modern antiretroviral therapy.

Virological suppression rates in people with human immunodeficiency virus HIV-1 (HIV) have markedly improved with the advent of modern antiretroviral therapy (ART), particularly following the introduction of optimized second-generation integrase strand transfer inhibitor (INSTI)–based regimens [1]. Despite these advances, some individuals experience persistent or intermittent low-level viremia (LLV) even while on effective ART and with confirmed adherence. Various definitions of LLV have been proposed based on the magnitude and persistence of viral detectability and differ substantially across international guidelines [2]. This heterogeneity hinders direct comparison across studies, leading to wide variability in prevalence estimates [3]. A recent systematic review and meta-analysis including >120 000 participants from 33 studies reported a pooled LLV prevalence of approximately 26% [4]; however, when narrower HIV-RNA thresholds were used to define LLV, its prevalence ranged between 7% and 15% [3].

More recently, a broader definition of low-level viral rebound (LLVR) has been adopted in several studies, based on expert panels and guidelines [5–8]. LLVR generally refers to a confirmed HIV-RNA detection of 51–199 copies/mL following

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previously confirmed viral suppression [9, 10], although upper thresholds vary by guideline (HIV-RNA <999 copies/mL for World Health Organization and <199 copies/mL for European and US guidelines) [5–8]. LLVR may encompass different clinical scenarios including "viral blips" with adjacent suppressed measurements, LLVR during stable first-line ART (>6 months), or rebound occurring after long-term suppression [11]. Factors that lead to LLVR are likely multiple [12, 13]. Reported risk factors include older age, late HIV diagnosis, suboptimal adherence, reduced regimen potency, archived resistance, transient immune activation due to coinfections or vaccinations, patient-specific immunological and clinical factors (eg, preexisting comorbidities), viral reservoir dynamics, and cases of virological escape. Persistent or transient detectable HIV-RNA in plasma in previously suppressed people with HIV (PWH) may arise from 2 distinct mechanisms: (*i*) bursts of $CD4^+$ T cells harboring integrated provirus, followed by a prompt block of viral replication by ART (viral production); and (*ii*) suboptimal drug potency or exposure or reduced drug penetration in sanctuary sites, leading to viral replication [14, 15]. A better understanding of this second mechanism is the main driver of our analysis.

Over the past decade, second-generation INSTI-based 2-drug regimens (2DR) have emerged as a simplified alternative to traditional 3-drug regimens (3DR), especially for individuals with sustained virological suppression. Clinical trials conducted in the suppressed switch settings have demonstrated that combinations such as dolutegravir (DTG) plus lamivudine (3TC), DTG plus rilpivirine (RPV), or the long-acting combination of cabotegravir (CAB) and RPV can effectively maintain viral suppression, offering favorable tolerability, proving noninferiority compared to 3DRs, and very low risk of virological failure in presence of good adherence [16–18]. Nevertheless, concerns persist regarding potential differences in drug potency or drug penetration with 2DR versus 3DR that could theoretically increase the risk of LLVR.

METHODS

Study Design and Population

We conducted a nested case-control study within the ICONA (Italian Cohort Naïve Antiretrovirals) Foundation Study [19], an ongoing prospective cohort of ART-naive PWH initiating ART followed up by 61 centers across 16 regions in Italy.

The case-control study was nested within the universe of PWH of ICONA who achieved virological suppression (defined as at least 2 consecutive HIV-RNA measurements ≤50 copies/mL over a 6-month period) for the first time after the fixed calendar date of 11 November 2014. This date was chosen because it corresponds to the date of introduction of 2DR with DTG in clinical practice in Italy. The date of the first of these 2 values ≤50 copies/mL was defined as baseline (BL). Cases were defined as participants who experienced LLVR, defined as a single HIV-RNA measurement between 51 and 199 copies/mL after BL. Sensitivity analyses were performed after restricting to cases who experienced 2 consecutive HIV-RNA values of 51–199 copies/mL.

For each case, up to 3 controls from the universe described above were matched on 2 key criteria: (*i*) history of care gaps (defined as an interval of >12 months between any 2 clinical visits); and (*ii*) number of prior antiretroviral drugs failed. Previous virological failure of a drug was defined as having spent >4 months with HIV-RNA >400 copies/mL while continuously being exposed to the drug. Because of the single value of HIV-RNA, the higher cut-off of 400 copies/mL was used instead of the standard definition of 2 consecutive values >200 copies/mL. The time at which cases experienced LLVR was considered the "index date." Controls were defined as those who maintained HIV-RNA ≤50 copies/mL up to the index date of the matching case.

Ethics

The ICONA Foundation study was approved by the local ethics committees of participating clinical sites. All patients provided written informed consent for study participation and data processing, in accordance with the ethical standards of the committee on human experimentation and the Helsinki Declaration (last amended in October 2013). The latest amendment of the ICONA Foundation Study was approved centrally by the Lazio Area 4 Territorial Ethics Committee on 1 July 2024 (approval no. 83-2024).

Statistical Analysis

In the cohort analysis leading to the identification of the time of developing LLVR, participants' follow-up accrued from the time of HIV-RNA ≤50 copies/mL achieved after November 2014 and until the occurrence of viral rebound (HIV-RNA >200 copies/mL) or the last available HIV-RNA measurement. Incidence of LLVR with 95% confidence intervals (CIs) was calculated using the Kaplan–Meier method.

In the case-control study nested within this cohort, the primary exposure of interest was the type of ART regimen in use at the index date. In particular, 2DR considered were DTG-based or CAB-based combinations (DTG/3TC, DTG/RPV, DTG/doravirine [DOR], CAB/RPV); 3DR were characterized by tenofovir disoproxil fumarate (TDF) or tenofovir alafenamide (TAF) + emtricitabine (FTC) or 3TC as backbone associated with a third agent among DTG, bictegravir (BIC), RPV, DOR, boosted darunavir, or boosted atazanavir.

Descriptive statistics for categorical variables and median and interquartile ranges (IQRs) for continuous variables were used to compare BL patient characteristics according to the exposure of interest (2DR vs 3DR) and to case/control status.

Conditional logistic regression models were used to estimate the association between regimen type (2DR vs 3DR) and LLVR, accounting for the matched design. Odds ratios (ORs) and corresponding 95% CIs were reported, with and without further adjusting for confounding. Covariates to be included in

multivariable models were selected a priori based on clinical relevance and prior literature. These included demographic factors (age, sex at birth, mode of HIV transmission, education level), BL CD4 count, BL CD8 count, duration of viral suppression prior to BL, duration of ART, calendar year of BL, BL body mass index, BL hepatitis C virus status, and a sum of comorbidities (diabetes mellitus, dyslipidemia, cardiovascular disease, chronic kidney disease, and cancer). In addition, we also controlled for confounding by means of covariate adjustment using a propensity score constructed using the same variables. Of note, resistance test results are not routinely collected in the cohort, so none of the models could be controlled for drug resistance. Full details of the definitions and assumptions used to identify the confounder sets used are available in Supplementary Figure 1.

In order to minimize bias due to HIV-RNA measurement errors or underreporting of viral loads test results, we performed 2 main sensitivity analyses: (*i*) in which we used an alternative definition of LLVR insisting on 2 consecutive HIV-RNA values of 51–199 copies/mL (sensitivity 1); and (*ii*) restricting to only ICONA sites without electronic submission of viral load data to the central database (sensitivity 2).

In addition, we also evaluated alternative specific contrasts after slightly modifying the definition of the exposure: (*iii*) including less frequently used 3DR, such as DTG/ABC/3TC and ABC/3TC + RPV (alternative exposure 1); restricting 2DR to only DTG/3TC (alternative exposure 2); restricting 3DR to only BIC/FTC/TAF (alternative exposure 3); or restricting 3DR to only BIC/FTC/TAF and 2DR to only DTG/3TC (alternative exposure 4). Statistical analyses were performed using SAS 9.4 (Cary, NC, USA) and STATA 18.5 (StataCorp LLC College Station, TX, USA) software.

RESULTS

To provide context on the overall incidence of LLVR in our cohort study population, among 6823 PWH, 261 experienced LLVR, yielding a crude incidence rate of 3.8%. The cumulative incidence calculated using the Kaplan–Meier method was 2.7% (95% CI, 2.3%–3.1%) by 2 years considering all single LLVR events and 1.8% (95% CI, 1.4%–2.1%) when limited to 2 consecutive values (Supplementary Figure 2). A description of the characteristics of the PWH enrolled in the whole ICONA cohort is also shown in Supplementary Table 1.

All of these 261 incident LLVR events were included in the case-control study for whom we identified unique matched controls; out of a total of 261 matched sets, 250 had 3 unique controls, while for 11 sets only 2 controls could be obtained, leading to a total of 772 controls and a total sample size of 1033 PWH included in the case-control analysis.

Overall, 215 of 1033 (21%) were females, 78% were born in Italy, the median age was 43 years (IQR, 34–51 years), BL CD4 count was 601 cells/μL (IQR, 379–826 cells/μL), and calendar year at BL was 2017 (IQR, 2015–2020); 849 participants had at least 1 comorbidity, more frequently found in the 3DR group than 2DR (82.7% vs 80.8%; $P = .024$). General characteristics of the case-control study population according to the ART regimen used at the index date are reported in Table 1. A similar table and a figure showing the decomposition of the exposure after stratification by case/control status are reported in Supplementary Table 2 and Supplementary Figure 3.

Regarding our main exposure of interest, 2DR were predominantly INSTI-based (70.4%), with DTG/3TC as the most common used combination (79% of 291); conversely 3DR were characterized by a more equal distribution of anchor drug (INSTI in 36.8%, protease inhibitor in 28%, and nonnucleoside reverse transcriptase inhibitor in 35%), with major use of RPV/TXF/FTC and BIC/FTC/TAF (30.5% and 24.4% of 742 participants on 3DR, respectively) (Figure 1).

The prevalence of 2DR was 25% (66/262) in cases versus 29% (225/772) in controls ($P = .23$). Participants using 2DR were younger (median age, 41 [IQR, 34–50] vs 44 [IQR, 35–52] years in 3DR; $P = .017$]) and more likely to be men who have sex with men (52.9% vs 44.9%), whereas heterosexual transmission was more common in 3DR (41.4% vs 36.8% in 2DR; $P = .09$). 2DR were more commonly used in recent years (median 2018) compared to 3DR (median 2017, $P < .001$). While there was no evidence for a difference in terms of ART duration ($P = .32$), participants receiving 2DR at index date had slightly longer duration of virological suppression than those on 3DR (median, 10.3 [IQR, 5.6–22.8] vs 9.3 [IQR, 4.9–18.1] months, $P = .02$).

In agreement with the crude prevalence data, the unadjusted conditional logistic regression analysis yielded an OR of 0.81 (95% CI, .58–1.13) when comparing the prevalence of 2DR in cases versus controls, thus with a point estimate suggesting a lower risk of LLVR with 2DR. Results were similar after controlling for confounding although with even wider CIs (adjusted OR [aOR], 0.86 [95% CI, .57–1.29]). Results were similar also in a model adjusted for a propensity score covariate (OR, 0.89 [95% CI, .63–1.27]) and after further controlling for the matching factors to minimize the risk of collider bias (aOR, 0.86 [95% CI, .57–1.28]; $P = .45$) [20]. We also looked carefully at specific source of potential confounding by age and by estimated glomerular filtration rate and found little evidence for this. Of note, 2DR appeared to be associated with higher risk in participants aged 18–43 years (OR, 1.15 [95% CI, .68–1.96], interaction $P = .10$). Overall, despite this latter result, plausibility intervals were wide in most analyses, and our study is inconclusive regarding whether the risk of LLVR may be different in PWH currently receiving 2DR versus 3DR.

Results were also similar in most sensitivity analyses (sensitivity analysis from 1 to 2) and across most alternative exposure definitions (alternative exposures 1–4). For example, when the analysis was restricted to the specific contrast of 2DR (DTG/3TC) versus 3DR (BIC/FTC/TAF) (alternative exposure 4), we found an aOR of 0.85 (95% CI, .57–1.27) (Figure 2).

Table 1. General Characteristics of Study Population by Antiretroviral Regimen

Characteristic at Baseline	2DR (n = 291)		3DR (n = 742)		Overall (N = 1033)		*P* Value
Controls	225	(77.3)	547	(73.7)	772	(74.7)	.23
Cases	66	(22.7)	195	(26.3)	261	(25.3)	
Age, y, median (IQR)	41	(34–50)	44	(35–52)	43	(34–51)	.02
Female sex	54	(18.6)	161	(21.7)	215	(20.8)	.26
HIV transmission risk group							.24
PWID	18	(6.2)	53	(7.1)	71	(6.9)	
Heterosexual	107	(36.8)	307	(41.4)	414	(40.1)	
MSM	154	(52.9)	333	(44.9)	487	(47.1)	
Other/unknown	12	(4.1)	49	(6.6)	61	(5.9)	
Italian (vs non-Italian)	222	(76.3)	584	(78.7)	806	(78)	.40
HCV Ab^+ test result							.004
Negative	160	(88.4)	664	(77.9)	824	(79.8)	
Positive	13	(7.2)	88	(10.3)	101	(9.8)	
Unknown	8	(4.4)	100	(11.7)	108	(10.5)	
History of AIDS	35	(12)	109	(14.7)	144	(13.9)	.27
CD4 count, cells/μL, median (IQR) at BL	586	(369–812)	603	(384–832)	601	(379–826)	.42
CD4 nadir, cells/μL, median (IQR)	292	(130–437)	295	(156–447)	294.5	(146–443)	.49
CD8 count, cells/μL, median (IQR) at BL	884	(619–1178)	862	(619–1123)	864	(619–1146)	.42
Peak HIV-RNA, $\log_{10}$ copies/mL, median (IQR)	4.9	(4.3–5.5)	4.9	(4.3–5.5)	4.9	(4.3–5.5)	.55
Anchor drug class							<.001
INSTI	205	(70.4)	273	(36.8)	478	(46.3)	
Mixed	17	(5.8)	0	(0)	17	(1.6)	
NNRTI	28	(9.6)	260	(35)	288	(27.9)	
PI	41	(14.1)	209	(28.2)	250	(24.2)	
Previous gap in care >18 mo	75	(25.8)	171	(23)	246	(23.8)	.36
Duration of most recent HIV-RNA suppression, mo, median (IQR)	9.3	(4.9–18.1)	10.3	(5.6–22.8)	10	(5.5–21.4)	.02
Duration of ART, mo, median (IQR)	40	(24–68)	37.5	(19–73)	38	(21–72)	.32
Year of baseline, median (IQR)	2018	(2016–2020)	2017	(2015–2021)	2017	(2015–2020)	<.001
No. of comorbidities[a]							.02
0	34	(11.7)	48	(6.5)	82	(7.9)	
1	235	(80.8)	614	(82.7)	849	(82.2)	
2	21	(7.2)	76	(10.2)	97	(9.4)	
3	1	(0.3)	4	(0.5)	5	(0.5)	
BMI, kg/m^2							.35
<30	189	(64.9)	508	(68.5)	697	(67.5)	
≥30	16	(5.5)	47	(6.3)	63	(6.1)	
Unknown	86	(29.6)	187	(25.2)	273	(26.4)	
Level of education							.42
Primary school	11	(3.8)	31	(4.2)	42	(4.1)	
Secondary school	138	(47.4)	343	(46.2)	481	(46.6)	
College/university	41	(14.1)	78	(10.5)	119	(11.5)	
Unknown	101	(34.7)	290	(39.1)	391	(37.9)	

Data are presented as No. (%) unless otherwise indicated.

Abbreviations: 2DR, 2-drug regimen; 3DR, 3-drug regimen; ART, antiretroviral therapy; BL, baseline; BMI, body mass index; HCV Ab, hepatitis C virus antibody; HIV, human immunodeficiency virus; INSTI, integrase strand transfer inhibitor; IQR, interquartile range; MSM, men who have sex with men; NNRTI, nonnucleoside reverse transcriptase inhibitor; PI, protease inhibitor; PWID, people who inject drugs.

[a]Includes diabetes mellitus, dyslipidemia, cardiovascular disease, cancer, and chronic kidney disease.

DISCUSSION

In our case-control study nested within a cohort of PWH who had achieved virological suppression ≤50 copies/mL after 11 November 2014, we evaluated whether current use of 2DR compared to 3DR was associated with the risk of developing LLVR.

First, we showed that LLVR (defined as a single viral load of 51–199 copies/mL) following confirmed viral suppression was not as common as previously reported. By 2 years from the date of achieving viral suppression, the incidence of LLVR was approximately 3% when all single episodes were considered and decreasing to 1.8% when restricted to 2 consecutive

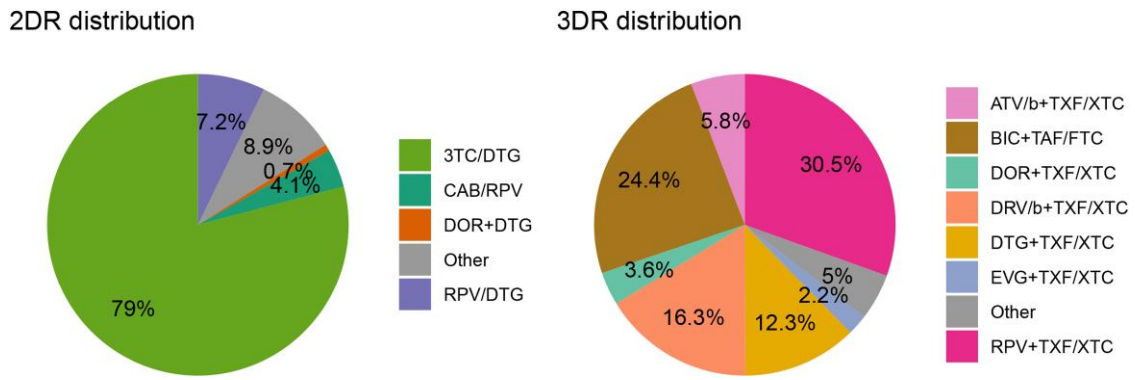

Figure 1. Distribution of regimens in 2-drug and 3-drug groups. Abbreviations: 2DR, 2-drug regimen; 3DR, 3-drug regimen; 3TC, lamivudine; ATV/b, boosted atazanavir; BIC, bictegravir; CAB, cabotegravir; DOR, doravirine; DRV/b, boosted darunavir; DTG, dolutegravir; EVG, elvitegravir; FTC, emtricitabine; RPV, rilpivirine; TAF, tenofovir alafenamide; TXF, tenofovir disoproxil fumarate or tenofovir alafenamide; FTC, emtricitabine or 3TC lamivudine.

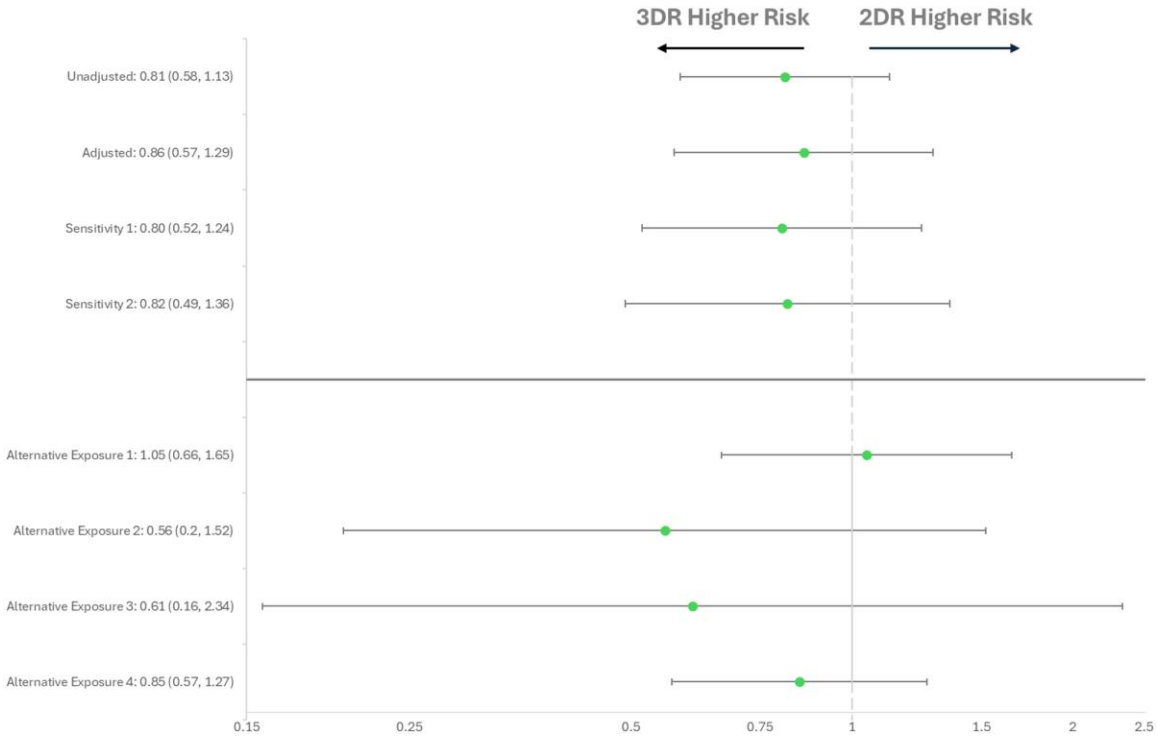

Figure 2. Results of the conditional logistic regression [odds ratios (OR) with 95% confidence intervals (CI)] on the comparison of risk of LLVR between 2- and 3-drug regimens across adjusted models. All models (apart from the unadjusted model) were adjusted for age, sex at birth, country of birth, mode of HIV transmission, education level, baseline CD4 cell count, baseline CD8 cell count, AIDS at baseline, duration of HIV-RNA suppression, duration of ART, year at baseline, baseline BMI, baseline HCV, and a sum of comorbidities (diabetes mellitus, dyslipidemia, cardiovascular diseases, chronic kidney disease, and cancer). Sensitivity 1: an alternative definition of LLVR insisting on 2 consecutive HIV-RNA values of 51–199 copies/mL; Sensitivity 2: restricting to only ICONA sites without electronic submission of viral load data to the central database. Alternative exposure 1: including uncommon 3DR regimens, such as DTG/ABC/3TC and ABC/3TC + RPV. Alternative exposure 2: restricting 2DR to only DTG/3TC. Alternative exposure 3: restricting 3DR to only BIC/FTC/TAF. Alternative exposure 4: restricting 3DR to only BIC/FTC/TAF and 2DR to only DTG/3TC. Abbreviations: 2DR, 2-drug regimen; 3DR, 3-drug regimen; 3TC, lamivudine; ABC, abacavir; ART, antiretroviral therapy; BIC, bictegravir; BMI, body mass index; CAB, cabotegravir; DTG, dolutegravir; FTC, emtricitabine; HCV, hepatitis C virus; HIV, human immunodeficiency virus; LLVR, low-level viral rebound; RPV, rilpivirine; TAF, tenofovir alafenamide.

measurements. This finding suggests that LLVR events are infrequent during long-term suppressive ART. The distinction between isolated and consecutive LLVR episodes likely reflects biological variability or assay fluctuations rather than true viral replication in most cases, supporting the clinical stability of virologically suppressed individuals under modern ART.

Consistent with the literature, LLVR in our cohort was more common among individuals with older age, lower nadir and current CD4 count, a history of AIDS, and higher zenith of HIV viral load. Although these associations were only investigated in univariable analyses, so confounding cannot be ruled out, these findings suggest that the risk of LLVR may depend on factors closely related to individuals' size of the HIV reservoir as well as current viro-immunological status [14, 15]. Indeed, it has been hypothesized that the larger the HIV reservoir, the greater a passive release of virions from latently infected cells leading to the development of LLV, regardless of the type of ART received [11]. The role of age is currently under further examination in an ongoing analysis of the data of the cohort.

Our analysis is inconclusive regarding the association between the use of 2DR versus 3DR and the risk of LLVR. The point estimate suggests a small benefit (10% reduction in risk) in PWH currently receiving 2DR, but the data are also consistent with a 29% higher risk with these regimens versus 3DR. Results were similar across multiple sensitivity analyses using alternative definitions of both the outcome and of the exposure. Overall, our data can rule out a moderate to high risk of LLVR in PWH treated with 2DR versus 3DR and therefore supports optimization/simplification to 2DR as a safe strategy alternative to traditional 3DR. Clinical trials with virological rebound >200 copies/mL showed the noninferiority of 2DR regimens such as DTG/3TC, DTG/RPV, and CAB/RPV [16–18] when compared to 3DR, and real-world evidence studies widely consolidated and supported these results in clinical practice [21–31]; our results are consistent with these data and potentially extend this finding to lower levels of viral rebound.

Our study has some limitations. First, the analysis was underpowered, as indicated by the wide CIs. This was the case even in the main analysis using a single value of HIV-RNA of 51–199 copies/mL. We used a single value not only to maximize the statistical power but also to minimize bias due to underreporting of HIV-RNA values. Indeed, in HIV cohorts typically HIV-RNA values are reported every 3–6 months, so results of repeated HIV-RNA tests conducted over shorter time windows to confirm LLVR may have not been reported to the central ICONA database. On the other hand, this is counterbalanced by the fact that a single altered HIV-RNA value could be due to random fluctuations or technical issues rather than a viral breakthrough on ART. However, results were similar in the sensitivity analysis using 2 consecutive values of 51–199 copies/mL.

Most importantly, we cannot rule out residual or unmeasured confounding. Indeed, PWH receiving 2DR are often inherently different from those treated with 3DR in terms of lifestyle and behavioral factors (ie, active substance use) or adherence (pharmacokinetics profiles and drug resistance), which are unmeasured factors in the ICONA cohort. We have tried to mitigate these possible sources of bias by controlling for mode of HIV transmission as well as history of previous gaps in care as a proxy of poor adherence and more chaotic lifestyles. Finally, most participants in the 2DR group were using the 3TC/DTG combination, which limits the generalizability of the results to other commonly used 2DR strategies. However, our analysis is comprehensive, featuring numerous comparisons among groups within the population who were currently receiving specific 2DR and 3DR regimens.

Despite these limitations, the study also has some strengths. ICONA is one of the largest cohorts of PWH in Europe, collecting longitudinal data from multiple infectious disease centers nationwide and providing a comprehensive and realistic picture of clinical practice in Italy. Although the cohort primary enrolls ART-naive individuals, participants are prospectively followed up beyond the achievement of viral suppression, allowing evaluation of outcomes in the switch setting, preserving the entire history since ART initiation. Moreover, in ICONA a large proportion of participants currently receive modern 2DR regimens, giving a realistic picture of our real-world experience. Finally, another strength of this analysis lies in its methodological richness, characterized by a wide array of contrasts across distinct subgroups of individuals treated with specific 2DR and 3DR regimens, giving more robustness to the results despite a certain margin of uncertainty.

CONCLUSIONS

In conclusion, our findings rule out a risk of LLVR >29% among PWH currently receiving 2DR compared with 3DR, providing some reassurance to clinicians regarding the use of 2DR in virologically suppressed individuals. These results support current HIV treatment guidelines and reinforce the role of 2DR as an effective simplification strategy in appropriately selected patients (those with stable virological suppression, good adherence, and no history of treatment failure or resistance). Further research is warranted to evaluate 2DR regimen performance in more vulnerable populations (eg, those with suboptimal adherence or comorbid conditions), to explore the immunological and inflammatory correlates of LLVR and to investigate its long-term evolution and its potential progression to virological failure or non-AIDS events. The optimal management of LLVR under 2DR remains uncertain; randomized studies are needed to determine whether, for example, an early switch to 3DR vs wait and watch in PWH experiencing LLVR while receiving 2DR regimens may be beneficial for long-term outcomes.

Supplementary Data

Supplementary materials are available at *Open Forum Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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Data availability. The datasets generated during the current study are not publicly available because they contain sensitive data to be treated under data protection laws and regulations. Appropriate agreement of data sharing can be arranged after a reasonable request to the corresponding author.

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