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Meta-analysis of randomized trials on the association of prophylactic acyclovir and HIV-1 viral load in individuals coinfecting with herpes simplex virus-2

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Abstract

Objective—To summarize the randomized evidence regarding the association between acyclovir use and HIV-1 replication as measured by plasma HIV-1 RNA viral load among individuals coinfecting with herpes simplex virus (HSV)-2.

Design—Meta-analysis of seven randomized trials conducted between 2000 and 2009. Inclusion criteria composed of acyclovir or valacyclovir use as prophylaxis among individuals coinfecting with HIV-1 and HSV-2 who were ineligible for highly active antiretroviral therapy. HIV-1 viral load was the outcome.

Methods—Random-effects summarization was used to combine treatment effect estimates. Stratified and meta-regression analyses were used to compare estimated treatment effects by characteristics of trials and participants.

Results—The summary treatment effect estimate was -0.33 (95% confidence interval: -0.56 , -0.10 , 95% population effects interval: -0.74 , 0.08) \log_{10} copies, an approximate halving of plasma viral load. However, there was marked heterogeneity ($P < 0.001$). Older median age, valacyclovir, higher compliance, earlier publication, and shorter study length were associated with a larger decrease in viral load as compared with their counterparts.

Conclusion—Current evidence suggests a range of favorable effects of acyclovir on plasma HIV-1 viral load among persons coinfecting with HSV-2.

Keywords

acyclovir; herpes simplex virus; HIV; meta-analysis

Introduction

Recent randomized clinical trials suggest that prophylactic treatment with acyclovir may decrease HIV-1 replication among individuals coinfecting with herpes simplex virus

(HSV)-2 [1–6]. In this context, the effect on HIV-1 replication is measured by a change in HIV-1 RNA plasma viral load (henceforth, viral load), which concurrent with antiretroviral therapy has been shown to correlate with a delay in disease progression [7]. Although there have been trials assessing this relationship between acyclovir use and viral load [1–6,8], to our knowledge, no quantitative, systematic review of this literature has been undertaken. Below we review and synthesize the published evidence from randomized trials assessing the association of acyclovir and viral load with a focus on describing trial characteristics.

Methods

Search strategy and inclusion criteria

We sought published articles that met the following criteria. Studies were randomized, placebo-controlled trials of the use of acyclovir as prophylaxis, not as primary use for treatment of genital ulcers. Individuals in the trials were coinfecting with HIV-1 and HSV-2. Participants were not eligible to be on highly active antiretroviral therapy. Finally, trials were required to have nonoverlapping participants; when the same participants were part of multiple publications, the publication reporting the effect of acyclovir on plasma viral load was used. In some cases, trial characteristics from reports not included in the analyses were used when the report that met the inclusion criteria did not report those characteristics.

The search terms ‘HIV’ or ‘AIDS’, ‘HSV’ or ‘herpes simplex virus’, and ‘acyclovir’ or ‘valacyclovir’ were used to identify articles in PubMed, Excerpta Medica database, ClinicalTrials.gov, and National Library of Medicine Gateway. The reference lists of found articles were searched for additional trials. Searches were conducted in May and June of 2010.

Statistical analysis

The linear regression coefficient representing the intent-to-treat, mean difference in \log_{10} HIV-1 plasma viral load at the primary endpoint between treatment and placebo groups and 95% confidence intervals (CIs) were abstracted from each trial. Standard error estimates for each measure were obtained as $\sigma_j = (a_j - b_j) = (2 \times 1.96)$, where a_j and b_j are the reported upper and lower 95% CI, respectively, and $j = 1$ to J indexes the trials.

Covariates abstracted were trial country, trial type (crossover or standard), publication year, trial length, average patient age, drug type (acyclovir or valacyclovir) and dose, compliance, percentage male, covariate adjustment, treatment protocol for genital ulcers, completion rates, and analysis type. Most trials reported median age, though two reported means [3,5]. In one trial [2], where age was reported categorically, the average age was estimated by taking a weighted average using the midrange of each category. Median compliance was measured by pill count in all trials. When compliance was reported only stratified by treatment arm [2,4,8], weighted averages were calculated to attain an overall trial summary. Baseline viral load should be balanced across treatment groups by randomization, and because the outcome measure is not a change score, adjustment for baseline viral load in a randomized trial should not alter the point estimate, but should shorten the CI due to a reduction in residual variation.

Overall heterogeneity was assessed by calculating a P value for the Cochran Q statistic, which is defined as

$$\sum_{j=1}^J (1/\sigma_j^2) [y_j - \bar{y}]^2$$

where j indexes the trials, \bar{y} is the average viral load difference, that is $\bar{y} = J^{-1} \sum_{j=1}^J y_j$. Under the null hypothesis of no heterogeneity, the Q statistic is distributed as χ^2 with $J-1$ degrees of freedom. A funnel plot was examined visually for asymmetry (i.e., a plot of $1/\sigma_j^2$ by y_j), and statistically by the method of Egger *et al.* [9] (i.e., the P value for the test of $\alpha_0 = 0$ from the linear regression equation $E[y_j | \sigma_j] = \alpha_0 + \alpha_1 \sigma_j^{-1}$), using a rank symmetry test of the trial-specific difference estimate and the variance of Begg and Mazumdar [10], as well as by the trim and fill method of Duval and Tweedie [11].

Summary measures and 95% CI were calculated by meta-regression using a random intercept for trial and restricted maximum likelihood to estimate the among-population variance (τ^2) [12,13]. This random effects model assumes a distribution of true effects among different populations (i.e., different trials). Therefore, we also calculated a 95% population effects interval using the among-population variance (τ^2); this is the interval between which 95% of populations are estimated to have their means. Meta-regression models, defined for a covariate X , as $y_j = (\beta_0 + v) \beta_1 X_j + \varepsilon_j$, where v and ε_j are independently distributed as $v \sim N(0, \tau^2)$, and $\varepsilon_j \sim N(0, \sigma_j^2)$, were also fit with one trial characteristic at a time as the independent variable. The small number of trials and clustering of trial characteristics prevented the fitting of multivariable meta-regression models. Analyses were carried out using SAS version 9.2 (SAS Institute Inc., Cary, North Carolina, USA) and STATA version 10.1 (StataCorp LP, College Station, Texas, USA).

Results

Seventeen trials were identified for potential inclusion in the study, all double blinded. Four trials overlapped with included trials, three trials were of short-term treatment for genital ulcer management, one did not include the outcome under study, one trial was not randomized, and in one trial participants were on highly active antiretroviral therapy. In one included trial, 8% of participants ($n = 41$) started antiretroviral therapy during follow-up (C. Tanton, personal communication). Seven trials remained for this study and are described in Table 1 [1–6,8].

The average age of the trial participants ranged from 28 to 33. Five trials were among women only, with one trial among men [6], and one that included both sexes [2]. Four trials were conducted in Africa, two in Peru, and one in Thailand. Four trials used acyclovir as the trial medication; the other three used its prodrug valacyclovir. All trials treated genital ulcers using a short course of high-dose acyclovir or valacyclovir. Completion rates were at or above 84% for all trials. All trials reported balanced completion across treatment arms,

except one [4] crossover trial. Five of the trials reported an effect measure unadjusted for baseline viral load; two reported adjusted estimates. Four of the trials analyzed their data using a linear mixed-effects model; three trials used linear regression.

There was no visual appearance of asymmetry in the sparsely populated funnel plot (Supplementary Fig 1, <http://links.lww.com/QAD/A144>). Egger's and Begg's test for small-study effects yielded *P* values of 0.3 and 0.3, respectively.

Results of stratification by trial characteristics are provided in Table 2. As expected, the more bioavailable preparation, valacyclovir, was seen to have a greater association with a decrease in viral load. Older median age, higher compliance, and shorter trial length were associated with a larger decrease in viral load as compared with their counterparts (Supplementary Fig. 2, <http://links.lww.com/QAD/A144>). Recent publication was associated with a lesser decrease in viral load.

The estimated viral load mean difference was $-0.33 \log_{10}$ copies/ml (95% CI: $-0.56, -0.10$), assuming a normal distribution for the random trial effect. The estimated variance of the random-effects distribution was $0.0440 (\log_{10} \text{ copies/ml})^2$. Thus, 95% of patient populations were estimated to have mean differences in the interval -0.74 – $-0.08 \log_{10}$ copies/ml [i.e., $-0.33 \pm 1.96(0.0440)^{0.5}$]. From the *z*-score of $-1.58 (= -0.33/0.0440^{0.5})$, 6% of populations were estimated to experience adverse effects on mean viral load (i.e., mean differences greater than zero). The Cochran *Q* statistic was 32.2, with six degrees of freedom ($P < 0.001$). This *Q* statistic measures heterogeneity among the trial-specific effect estimates. Therefore, there was notable evidence of heterogeneity. A trim-and-fill analysis, which is often used to explore the impact of publication bias, imputed two possibly missing trial results, reducing the random effects summary estimate from $-0.33 \log_{10}$ co-to $-0.25 \log_{10}$ copies/ml.

Discussion

Acyclovir is an inexpensive and readily available drug that is effective at decreasing HSV-2 shedding, ulcer formation, and HSV-2 transmission in HIV-1-infected and uninfected persons [16]. The pooled estimate of $-0.33 \log_{10}$ copies/ml represents a clinically meaningful decrease in viral load. In the context of HIV-1 therapy, decreases in plasma viral load exhibit a linear relationship with lower risk of AIDS progression and death [17], and viral load has subsequently been used as a surrogate marker for these endpoints in clinical trials. Associations seen between a larger decrease in viral load with greater compliance and shorter trial length in this meta-analysis indicate potential problems for replicating trial results in the general population. Further, studies to assess the impact of acyclovir in the general HIV-infected population are needed as the observed effect on HIV viral load may be mediated by its suppressive effect on HSV, or may be due to direct antiretroviral effects [18]. The latter would presumably be independent of HSV infection status. In addition, trials with median ages outside the narrow 28–33 year range or that report results stratified by age would improve generalizability.

Caution should be exercised when interpreting the pooled results of this meta-analysis. First, meta-analyses are subject to metaconfounding just as observational studies are subject to confounding. Second, randomized trials provide internally valid effect estimates assuming full compliance and no loss to follow-up. Although most of the loss to follow-up in these studies was balanced across treatment arms, there is still potential for selection bias: the individuals remaining under study may not be representative of the randomized sample. Even internally valid trial comparisons may not generalize to a HIV-infected target population that differs in composition from trial populations [19,20], which are often highly selective samples. Use of additional antiherpes drugs could impact results. However, in these trials a majority of the episodes of herpes that required treatment occurred in the placebo group, potentially attenuating the association between acyclovir and viral load. Third, results from the trim-and-fill analysis indicate potential publication bias in this literature.

Fourth, meta-analyses on aggregate data are vulnerable to ecologic biases as the exposure, outcome, and covariates are reported at the group level rather than the individual level. We are interested in the individual, biologic effect of acyclovir on HIV-1 plasma viral load, rather than the group level effect of acyclovir. For instance, we observed a lesser acyclovir association in trials conducted in younger populations. However, given individual data, we may see that, within each trial, the acyclovir association is stronger at younger ages. Such a finding would be an example of the ecologic fallacy [21].

Random effects estimates assume a distribution of population effects rather than a single true effect. The 6% of patient populations estimated to have detrimental association of acyclovir with viral load are not consistent with our understanding of possible acyclovir mechanisms. Estimating a distribution of effects of acyclovir may not be representative of the true effect. Alternatively, the detrimental proportion could be due to chance.

In addition to trials of acyclovir on HIV-1 viral load in plasma, trials have explored acyclovir's effect in genital [3,5], cervical [1,4], cervico-vaginal [8,22], rectal [6], and seminal [14] fluids. Generally, a decrease in viral load has been found in these sites associated with acyclovir treatment. Although a majority of this literature is among participants who were not eligible to be on highly active antiretroviral therapy, the pooled estimate from the present meta-analysis is similar to the estimate from the only published trial examining HIV-1 plasma viral load among participants on highly active antiretroviral therapy (yet poorly controlled with median viral load at randomization of 3.70 log₁₀ copies/ml), which was -0.41 log₁₀ copies/ml (95% CI: -1.35, 0.53) [23].

This is the first meta-analysis of prophylactic acyclovir and HIV-1 plasma viral load in participants coinfecting with HSV-2. Although a recent study in HIV-1 and HSV-2 coinfecting persons has shown that despite lowering HIV-1 plasma levels, acyclovir may not prevent HIV-1 transmission [2], a separate analysis from that study suggested that acyclovir may slow HIV disease progression as measured by a combined outcome of time to CD4 cell counts less than 200, antiretroviral therapy initiation, or death [15]. This influential study has led some investigators to cautiously propose using acyclovir to slow progression of HIV thereby delaying the need for antiretroviral therapy [15,24]. If acyclovir prolongs the time to

effective antiretroviral therapy, yet has no effect on reducing transmission, using acyclovir in this manner could increase HIV-1 transmission. As antiretroviral therapy becomes increasingly affordable and available in the developing world, the modest benefit of acyclovir use may become less important. Additional long-term trials, particularly those that would assess benefits including time to AIDS and death could help to elucidate the mechanism of acyclovir's effect on HIV-1. Understanding whether the association of acyclovir with slowed HIV replication is observed among a broader array of HIV-infected populations will help decide how best to use acyclovir in HIV disease management.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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References

1. Baeten JM, Strick LB, Lucchetti A, Whittington WL, Sanchez J, Coombs RW, et al. Herpes simplex virus (HSV)-suppressive therapy decreases plasma and genital HIV-1 levels in HSV-2/HIV-1 coinfecting women: a randomized, placebo-controlled, cross-over trial. *J Infect Dis.* 2008; 198:1804–1808. [PubMed: 18928378]
2. Celum C, Wald A, Lingappa JR, Magaret AS, Wang RS, Mugo N, et al. Acyclovir and transmission of HIV-1 from persons infected with HIV-1 and HSV-2. *N Engl J Med.* 2010; 362:427–439. [PubMed: 20089951]
3. Delany S, Mlaba N, Clayton T, Akpomiemie G, Capovilla A, Legoff J, et al. Impact of aciclovir on genital and plasma HIV-1 RNA in HSV-2/HIV-1 co-infected women: a randomized placebo-controlled trial in South Africa. *AIDS.* 2009; 23:461–469. [PubMed: 19155993]
4. Dunne EF, Whitehead S, Sternberg M, Thepamnuay S, Leelawiwat W, McNicholl JM, et al. Suppressive acyclovir therapy reduces HIV cervicovaginal shedding in HIV- and HSV-2-infected women, Chiang Rai, Thailand. *J Acquir Immune Defic Syndr.* 2008; 49:77–83. [PubMed: 18667923]
5. Nagot N, Ouedraogo A, Foulongne V, Konate I, Weiss HA, Vergne L, et al. Reduction of HIV-1 RNA levels with therapy to suppress herpes simplex virus. *N Engl J Med.* 2007; 356:790–799. [PubMed: 17314338]
6. Zuckerman RA, Lucchetti A, Whittington WL, Sanchez J, Coombs RW, Zuniga R, et al. Herpes simplex virus (HSV) suppression with valacyclovir reduces rectal and blood plasma HIV-1 levels in HIV-1/HSV-2-seropositive men: a randomized, double-blind, placebo-controlled crossover trial. *J Infect Dis.* 2007; 196:1500–1508. [PubMed: 18008230]
7. O'Brien WA, Hartigan PM, Martin D, Esinhart J, Hill A, Benoit S, et al. Changes in plasma HIV-1 RNA and CD4+ lymphocyte counts and the risk of progression to AIDS. Veterans Affairs Cooperative Study Group on AIDS. *N Engl J Med.* 1996; 334:426–431. [PubMed: 8552144]
8. Tanton C, Weiss HA, Rusizoka M, Legoff J, Chagalucha J, Baisley K, et al. Long-term impact of acyclovir suppressive therapy on genital and plasma HIV RNA in Tanzanian women: a randomized controlled trial. *J Infect Dis.* 2010; 201:1285–1297. [PubMed: 20235836]
9. Egger M, Davey Smith G, Schneider M, Minder C. Bias in meta-analysis detected by a simple, graphical test. *BMJ.* 1997; 315:629–634. [PubMed: 9310563]

10. Begg CB, Mazumdar M. Operating characteristics of a rank correlation test for publication bias. *Biometrics*. 1994; 50:1088–1101. [PubMed: 7786990]
11. Duval S, Tweedie R. Trim and fill: a simple funnel-plot-based method of testing and adjusting for publication bias in meta-analysis. *Biometrics*. 2000; 56:455–463. [PubMed: 10877304]
12. DerSimonian R, Laird N. Meta-analysis in clinical trials. *Control Clin Trials*. 1986; 7:177–188. [PubMed: 3802833]
13. Thompson SG, Sharp SJ. Explaining heterogeneity in meta-analysis: a comparison of methods. *Stat Med*. 1999; 18:2693–2708. [PubMed: 10521860]
14. Zuckerman RA, Lucchetti A, Whittington WL, Sanchez J, Coombs RW, Magaret A, et al. HSV suppression reduces seminal HIV-1 levels in HIV-1/HSV-2 co-infected men who have sex with men. *AIDS*. 2009; 23:479–483. [PubMed: 19169140]
15. Lingappa JR, Baeten JM, Wald A, Hughes JP, Thomas KK, Mujugira A, et al. Daily acyclovir for HIV-1 disease progression in people dually infected with HIV-1 and herpes simplex virus type 2: a randomised placebo-controlled trial. *Lancet*. 2010; 375:824–833. [PubMed: 20153888]
16. Whitley RJ, Gnann JW Jr. Acyclovir: a decade later. *N Engl J Med*. 1992; 327:782–789. [PubMed: 1288525]
17. Marschner IC, Collier AC, Coombs RW, D'Aquila RT, DeGruttola V, Fischl MA, et al. Use of changes in plasma levels of human immunodeficiency virus type 1 RNA to assess the clinical benefit of antiretroviral therapy. *J Infect Dis*. 1998; 177:40–47. [PubMed: 9419168]
18. McMahon MA, Siliciano JD, Lai J, Liu JO, Stivers JT, Siliciano RF, et al. The antiherpetic drug acyclovir inhibits HIV replication and selects the V75I reverse transcriptase multidrug resistance mutation. *J Biol Chem*. 2008; 283:31289–31293. [PubMed: 18818198]
19. Cole SR, Stuart EA. Generalizing evidence from randomized clinical trials to target populations: The ACTG 320 trial. *Am J Epidemiol*. 2010; 172:107–115. [PubMed: 20547574]
20. Greenhouse JB, Kaizar EE, Kelleher K, Seltman H, Gardner W. Generalizing from clinical trial data: a case study. The risk of suicidality among pediatric antidepressant users. *Stat Med*. 2008; 27:1801–1813. [PubMed: 18381709]
21. Morgenstern H. Ecologic studies in epidemiology: concepts, principles, and methods. *Annu Rev Public Health*. 1995; 16:61–81. [PubMed: 7639884]
22. Cowan FM, Pascoe SJ, Barlow KL, Langhaug LF, Jaffar S, Hargrove JW, et al. A randomised placebo-controlled trial to explore the effect of suppressive therapy with acyclovir on genital shedding of HIV-1 and herpes simplex virus type 2 among Zimbabwean sex workers. *Sex Transm Infect*. 2008; 84:548–553. [PubMed: 18684855]
23. Ouedraogo A, Nagot N, Vergne L, Konate I, Weiss HA, Defer MC, et al. Impact of suppressive herpes therapy on genital HIV-1 RNA among women taking antiretroviral therapy: a randomized controlled trial. *AIDS*. 2006; 20:2305–2313. [PubMed: 17117016]
24. Vanpouille C, Lisco A, Margolis L. Acyclovir: a new use for an old drug. *Curr Opin Infect Dis*. 2009; 22:583–587. [PubMed: 19726982]

Table 1
 Characteristics of randomized trials of acyclovir on HIV-1 plasma viral load among HIV-1/HSV-2 coinfecting adults.

Author	N	Trial Length ^a	Treatment	Plasma collection	Estimate (95% CI) ^b	Weight (%) ^c
Nagot <i>et al.</i> [5]	140	12 weeks	Valacyclovir, 500 mg twice daily	Monthly	0.87 (-1.19, -0.55) ^f	6.11
Zuckerman <i>et al.</i> [6]	20	8 weeks, 2-week washout	Valacyclovir, 500 mg twice daily	Weekly	-0.33 (-0.42, -0.23)	17.69
Dunne <i>et al.</i> [4]	67	1 month, 1-month washout	Acyclovir, 800 mg twice daily	Monthly	-0.46 (-0.61, -0.31)	13.89
Delany <i>et al.</i> [3]	300	3 months	Acyclovir, 400 mg twice daily	Monthly	-0.34 (-0.54, -0.15) ^{e,f}	11.13
Baeten <i>et al.</i> [1]	20	8 weeks, 2-week washout	Valacyclovir, 500 mg twice daily	Weekly	-0.26 (-0.33, -0.19)	19.30
Tanton <i>et al.</i> [8]	484	6 months ^d	Acyclovir, 400 mg twice daily	6, 12, and 24 months	0.04 (-0.16, 0.24) ^f	10.86
Celum <i>et al.</i> [2]	3408	2 years	Acyclovir, 400 mg twice daily	Quarterly	-0.25 (-0.29, -0.22) ^e	21.01

CI, confidence interval; HSV, herpes simplex virus.

^aWhen a washout length is given, the design was a crossover trial.

^bMeasured at the end of follow-up.

^cWeights are random-effects weights: the inverse of the sum of each estimated within-trial variance (σ^2_i) and the estimated among-trial variance (τ^2).

^dFull trial length was 2 years, but authors reported primary results at the 6-month time point.

^eAdjusted for baseline HIV-1 concentrations.

^fDifference in HIV-1 viral load calculated by linear regression, other studies used linear mixed effects models.

Table 2

Stratified and meta-regression results of randomized control trials of the association between acyclovir and HIV-1 plasma viral load.

Study characteristic	<i>N</i>	Heterogeneity test <i>P</i>	Random-effects vl difference (95% CI)	Meta-regression (difference of vl difference)
Overall	7	<0.001	-0.33 (-0.56, -0.10)	
Study design				
RCT	4	<0.001	-0.34 (-0.92, 0.27)	0.02 (-0.52, 0.56)
Crossover RCT	3	0.05	-0.33 (-0.57, -0.10)	0
Drug preparation				
Valcyclovir	3	<0.001	-0.46 (-1.25, 0.33)	-0.18 (-0.68, 0.31)
Acyclovir	4	<0.001	-0.26 (-0.59, 0.07)	0
Median age				
>31.05	3	0.02	-0.53 (-1.18, 0.12)	-0.30 (-0.68, 0.89)
31.05	4	0.01	-0.22 (-0.45, 0.00)	0
Compliance				
>96%	3	<0.001	-0.50 (-1.24, 0.24)	-0.25 (-0.70, 0.19)
96%	4	0.009	-0.23 (-0.48, 0.03)	0
Publication year				
2009–2010	3	0.01	-0.19 (-0.66, 0.28)	0.25 (-0.19, 0.69)
2007–2008	4	<0.001	-0.44 (-0.84, -0.05)	0
Trial length				
>12 weeks	2	0.005	-0.12 (-1.95, 1.71)	0.29 (-0.16, 0.73)
12 weeks	5	0.001	-0.41 (-0.67, -0.15)	0
Location				
Africa	4	<0.001	-0.34 (-0.92, 0.25)	0.02 (-0.52, 0.56)
Other	3	0.05	-0.33 (-0.57, -0.10)	0
Adjustment for baseline vl				
Unadjusted	5	0.38	-0.25 (-0.48, -0.03)	0.06 (-0.52, 0.65)
Adjusted	2	<0.001	-0.36 (-0.75, 0.03)	0

CI, confidence interval; RCT, randomized controlled trial; vl, viral load.