

UCSF

UC San Francisco Previously Published Works

Title

Role of HCV Viremia in Corroborated HCV Transmission Events Within Young Adult Injecting Partnerships

Permalink

<https://escholarship.org/uc/item/9xd896db>

Journal

Open Forum Infectious Diseases, 6(4)

ISSN

2328-8957

Authors

Hahn, Judith A
Tully, Damien C
Evans, Jennifer L
[et al.](#)

Publication Date

2019-04-01

DOI

10.1093/ofid/ofz125

Peer reviewed

Role of HCV Viremia in Corroborated HCV Transmission Events Within Young Adult Injecting Partnerships

Judith A. Hahn,¹ Damien C. Tully,² Jennifer L. Evans,³ Meghan D. Morris,³ Alya Briceno,³ David J. Bean,⁴ Todd M. Allen,⁴ and Kimberly Page⁵

¹Department of Medicine, University of California, San Francisco, San Francisco, California; ²Department of Infectious Disease Epidemiology, London School of Hygiene & Tropical Medicine, London, UK; ³Department of Epidemiology and Biostatistics, University of California, San Francisco, San Francisco, California; ⁴Ragon Institute of MGH, MIT, and Harvard, Cambridge, Massachusetts; ⁵University of New Mexico, Albuquerque, New Mexico

Background. Hepatitis C virus (HCV), a major cause of morbidity and mortality, is common and rising among young persons who inject drugs (PWID). Reducing the level of viremia may be an intervention, yet the impact of viremia on HCV transmission is unknown.

Methods. We conducted a prospective study of injecting partnerships (Partner Study) of young adult (age < 30 years) PWID within the UFO Study, which enrolled those at risk for HCV or with seronegative viremic infection and up to 3 HCV RNA-positive regular injecting partners. We examined the level of HCV viremia and stage of infection in the HCV-positive partner in regression analyses of HCV transmission events that were corroborated via HCV phylogenetic linkage analyses.

Results. We enrolled 69 at-risk/acutely infected PWID. There were 25 new HCV infections (incidence rate, 35.9 per 100 person-years; 95% confidence interval [CI], 24.3–53.2 per 100 person-years); 12/25 (48%) were phylogenetically linked to at least 1 partner. We found no association between the infected partner's quantitative level of HCV viremia and likely transmission in multivariate analyses (adjusted odds ratio [AOR], 0.90; 95% confidence interval [CI], 0.55–1.46); however, seronegative viremic infection in the infected partner was associated with increased transmission (AOR, 28.02; 95% CI, 5.61–139.95).

Conclusions. The HCV viremia level was not associated with increased odds of transmission, yet acute HCV infection (seronegative viremic) was. Explanations include high-risk behavior during acute infection or missed fluctuations in viremia during acute infection. Both point to the need for frequent testing to detect new infection and attempt to prevent onward transmission.

Keywords. acute hepatitis C infection; hepatitis C virus; injecting partnerships; phylogenetic linkage.

Hepatitis C virus (HCV) transmission is common and rising among young adult people who inject drugs (PWID) [1, 2]. Despite the implementation of services designed to prevent transmission of blood-borne infections in urban areas [3], the HCV incidence in PWID in several North American cities has remained high over the past 15 years; it is 25 per 100 persons-years in young adult PWID in San Francisco [4]. In nonurban areas in the United States, HCV incidence is on the rise among young adult PWID [5, 6], in tandem with the national opioid epidemic.

Strategies to combat the HCV epidemic among PWID include providing sterile injecting equipment and medication-assisted therapy [1, 7, 8]. In addition, as direct-acting antiviral agents (DAAs) that have high cure rates (>90%), reduced treatment time, and few side effects [9–12] have become available, treatment as prevention (TasP) has been proposed as an important

step toward HCV elimination [13, 14]. Another vital approach to combat the HCV epidemic is the development of a vaccine. Vaccines may provide protection against infection, or more likely, in the case of HCV, prevent chronic infection or decrease the level of viremia during acute infection [15–17]. However, it is unknown whether the level of HCV viremia is associated with injecting-related transmission of HCV, although mother-to-child transmission of HCV is associated with level of maternal HCV viremia [18]. HCV viremia is highest during acute infection, most notably during the period of infection when HCV antibodies (anti-HCV) are not detectable [19]. Thus, we aimed to determine the role of HCV viremia on HCV transmission.

We conducted a longitudinal study to detect new HCV infections within at-risk young adult PWID in injecting partnerships. We previously found that young PWID have a large number of injecting partners [20]; therefore, we used viral sequencing and phylogenetic analyses to provide further evidence for suspected HCV transmission events within injecting partnerships. We aimed to determine whether the level of viremia in the source partners was associated with increased HCV transmission to the at-risk partners and examined other partnership characteristics previously associated with HCV transmission, including the infection phase of the infected partner (HCV-seronegative viremic vs HCV-seropositive viremic). We controlled for partnership characteristics previously associated

Received 3 December 2018; editorial decision 2 March 2019; accepted 5 April 2019.

Correspondence: J. Hahn, PhD, MA, Division of HIV, Infectious Diseases & Global Medicine, Department of Medicine, University of California, San Francisco, 550 16th St., 3rd Floor, San Francisco, CA 94158 (judy.hahn@ucsf.edu).

Open Forum Infectious Diseases®

© The Author(s) 2019. Published by Oxford University Press on behalf of Infectious Diseases Society of America. This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivs licence (<http://creativecommons.org/licenses/by-nc-nd/4.0/>), which permits non-commercial reproduction and distribution of the work, in any medium, provided the original work is not altered or transformed in any way, and that the work is properly cited. For commercial re-use, please contact journals.permissions@oup.com DOI: 10.1093/ofid/ofz125

with HCV transmission such as partnership type (sexual activity yes vs no), age difference between partners, and injecting behaviors such as frequency of injecting together and needle/syringe and injecting equipment sharing within partnerships.

METHODS

Study Participants

The Partner Study is a substudy of the UFO Study, a prospective cohort study of incident HCV and natural history of early infection [21]. Participants were enrolled in the Partner Study from 2006 through 2017; enrollment was not continuous due to funding breaks. Eligibility for the UFO Study included self-reported current injecting (past 30 days, confirmed by demonstrating knowledge of injecting procedures), being under age 30 years, having the ability to provide informed consent, being fluent in English, having no plans to leave San Francisco, and self-reporting not being HCV RNA positive at baseline. UFO Study participants who reported injecting drugs with another person during study interviews were invited to bring their current injecting partner(s) for Partner Study screening. Eligibility criteria for the Partner Study included reporting regularly injecting together in the same space in the prior month (≥ 5 times in the prior month for pre-2015 enrollees, ≥ 3 times in the prior month for enrollees in 2015 or after). Partnerships were eligible for the UFO Partner Study if they were discordant on HCV RNA or if they were HCV RNA concordant (both positive), with at least 1 of the partners recruited in the HCV-seronegative viremic phase of HCV infection (Table 1). The partner who was HCV RNA negative or in the seronegative viremic phase is referred to as the “at-risk partner.” For the purposes of data analysis, when both partners presented with early acute infection ($n = 3$), the partner interviewed first was labeled the at-risk partner. In addition, because the at-risk partner was, by definition, either HCV RNA negative or within the seronegative viremic phase, we enrolled several (16) at-risk partners who were HCV RNA negative but HCV antibody (anti-HCV) positive, suggestive of previous spontaneous clearance of HCV infection. The HCV RNA-positive partner was called the “index partner.” At-risk participants were allowed to be enrolled with up to 3 index partners at 1 time, and new partners could be enrolled at any time (with a maximum of 3 concurrent partners

per at-risk participant). All participants were asked to individually complete monthly visits for 6 months, with active partnerships re-enrolled for another 6 months.

Laboratory Measures

Incident HCV infection in at-risk partners was detected at baseline (HCV-seronegative viremia) and subsequently (via HCV RNA and/or anti-HCV) via quarterly antibody and RNA testing. Plasma samples were tested for HCV RNA using a nucleic acid amplification test (Proclix HIV-1/HCV assay, Gen-Probe Inc., San Diego, CA, and marketed by Novartis Vaccines & Diagnostics, Emeryville, CA). Anti-HCV was tested at a commercial lab on samples obtained by venipuncture (HCV version 3.0 ELISA test system; Ortho Clinical Diagnostics) before 2012, and with anti-HCV rapid testing (OraSure Technologies, Bethlehem, PA) starting in 2012. These tests have been shown to be highly concordant [22]. Quantitative HCV viremia was measured for the index partners at Partner Study entry and approximately yearly thereafter using the Bayer HCV RNA branched DNA assay (Versant HCV 3.0; Bayer Diagnostics, Tarrytown, NY) before 2011, and the Hepatitis C Virus RNA Quantitative Real-Time PCR (ARUP Laboratories, Salt Lake City, UT) starting in 2011; the latter detects higher levels of virus in some genotypes [23, 24]. HCV genotypes for newly identified infections and their index partners were determined by Hepatitis C Viral RNA Genotype LiPA (Quest Diagnostics, San Jose, CA).

HCV Amplification, Sequencing, and Phylogenetic Analysis

HCV RNA was isolated from plasma using the QIAamp Viral RNA Mini Kit (Qiagen) for both partners, collected closest to the first visit where HCV viremia was detected, in pairs in which the genotype matched. Viral RNA was reverse-transcribed and amplified using up to 3 different primer sets. An amplicon approximately 3.2 kb in length covering Core to NS2 was attempted, as was a 389-bp fragment of the NS5B region. If amplification was unsuccessful, a smaller region of the HCV genome encompassing the HVR1 was attempted. Complete details on primers and PCR conditions used for Core to NS2 and HVR1 have been described by Tully et al. [23], and complete details on NS5B have been described by Murphy et al. [25]. Samples that successfully amplified were

Table 1. HCV Status of Participant and Recruited Partner (n = 101 Partnerships) at the Time of UFO Partner Study Recruitment

UFO Participant HCV Status	HCV Naïve (Anti-HCV- and HCV RNA-)	Cleared HCV (Anti-HCV+ and HCV RNA-)	Chronic HCV (Anti-HCV+ and HCV RNA+)	Acute HCV (Anti-HCV- and HCV RNA+)	Total
HCV naïve (anti-HCV- and HCV RNA-)	Not eligible	Not eligible	n = 29	n = 1	n = 30
Cleared HCV (anti-HCV+ and HCV RNA-)	Not eligible	Not eligible	n = 13	n = 3	n = 16
Chronic HCV (anti-HCV+ and HCV RNA+)	n = 38	n = 7	Not eligible	n = 3	n = 48
Acute HCV (anti-HCV- and HCV RNA+)	n = 0	n = 0	n = 4	n = 3	n = 7
Total	n = 38	n = 7	n = 46	n = 10	n = 101

Abbreviation: HCV, hepatitis C virus.

multiplexed and sequenced on an Illumina MiSeq platform, using a 2×250-bp V2 reagent kit. Paired-end reads obtained from Illumina MiSeq were subjected to stringent quality control and filtering and assembled into an HCV consensus sequence using the VICUNA de novo assembler software [26] and finished with V-FAT, as previously described [27, 28]. Maximum likelihood phylogenetic trees were constructed with the robustness of the resulting tree, assessed by bootstrapping with 1000 replicates, and clusters were identified with a bootstrap threshold of 90% and a maximum genetic distance threshold of 0.05.

Study Variables

The UFO Study conducted quarterly structured interviews with all participants. Variables included person-level characteristics (gender, age, race). Partnership-level structured interviews were conducted monthly for each partner separately and included past month partnership injecting behaviors (receptive syringe sharing, ancillary injecting equipment sharing, number of days injected together, number of other injecting partners) and sexual exposures. For behavioral variables, we used the responses of the at-risk partner.

Statistical Analyses

We calculated the overall incidence of HCV infection within at-risk partners using person-time methods. We categorized HCV transmission within partnerships as follows: group 1: “no transmission event” for partnerships in which no new HCV infection occurred in the at-risk partner, that is, no HCV transmission; group 2: “corroborated HCV transmission event” for partnerships in which the at-risk partner became infected with HCV or was recruited during the HCV-seronegative viremic phase, *and* the viral sequences from both partners were phylogenetically linked; group 3: “new HCV infection, transmission not corroborated” for partnerships in which the at-risk partner become infected during the study or was in the early acute infection phase at recruitment, but the viral sequence of the new HCV infection was *not* phylogenetically linked with that of the enrolled partner or the HCV genotypes differed between the partners; and group 4: “new HCV infection, sequence not determined” for the small number (4) of partnerships in which a new HCV infection occurred during the study, but phylogenetic linkage could not be determined due to insufficient materials available for sequencing for 1 or both of the partners. We present proportions and medians (with interquartile ranges [IQRs]) for at-risk partner characteristics, partnership characteristics, and partnership behaviors (at Partner Study enrollment), overall and by transmission category.

To examine the characteristics associated with corroborated HCV transmission events within partnerships, we conducted generalized estimating equation logistic regressions with robust standard errors, to account for multiple visits per partnership,

to approximate a Cox model. We used all available at-risk partner visits, with the outcome variable coded as 0 for visits occurring before new HCV infection for all outcome groups and 1 for the first visit for which the HCV transmission event was detected. All visits for which the at-risk partner was HCV RNA negative were included, whereas we dropped the visits occurring after new HCV infection was identified and those visits for which and after new HCV infections were identified for which phylogenetic linkage was not determined (ie, group 4) or there was no match to the putative partner (ie, group 3). Demographic and partnership characteristic variables (index race/ethnicity and age, age difference between partners, gender composition of partnership, duration of partnership, whether the index partner was in the HCV-seronegative viremic phase) were time invariant, whereas partnership injecting behavior variables (frequency of injecting, receptive needle sharing, ancillary equipment sharing, sex with partner, number of other injecting partners) were updated for each at-risk partner visit in the regression analyses.

Because the index partner’s HCV viremia level (analyzed as a log₁₀ transformation) was measured approximately yearly, rather than at each study visit, and because the study visits for partners were not required to be on the same day, we used the level of viremia at the most recent index partner visit if the visit occurred within 2.5 months of the at-risk partner visit. To fill in for missing levels of viremia, we conducted the analysis using a last value carried forward approach for the level of index partner viremia, thus allowing all visits described above to be included (n = 509 visits). We conducted sensitivity analyses that included *only* visits for which the partner HCV viremia level was available within 2.5 months of the at-risk partner visit.

We further constructed a multivariable regression model of phylogenetically corroborated HCV transmission events, including HCV viremia, the main exposure of interest, as well as variables that were associated with HCV transmission in the bivariate analyses ($P < .10$), as above. Partner viremia was included using last-value carried forward, as above, and we additionally conducted a sensitivity analysis using only visits for which the partner level of viremia was measured within 2.5 months of the at-risk partner visit. In addition, because early acute HCV infection status of the partner was likely to be correlated with HCV viremia [19], we constructed additional models removing the former variable. Lastly, we ran the multivariable model removing the partnerships in which both partners were enrolled during the HCV-seronegative viremic phase to determine the impact of including these participants on the parameter estimates.

RESULTS

Sixty-nine persons enrolled as at-risk participants, with a median of 1 HCV-infected index partner per at-risk partner

Table 2. Demographic and Behavioral Characteristics of 101 Prospectively Followed Injecting Partnerships, Overall and by HCV Transmission Category

Characteristic	All (n = 101)	Group 1: No Transmission Event (n = 61)	Group 2: Phylogenetically Corroborated HCV Transmission Event (n = 12)	Group 3: New HCV Infection, Transmission Not Phylogenetically Corroborated (n = 24)	Group 4: New Infection, Sequence Not Determined (n = 4)
		No. (%) or Median (IQR)			
Demographics (baseline)					
Race of at-risk ^a partner = nonwhite	33 (33)	19 (31)	5 (42)	9 (38)	0 (0)
Median age of at-risk ^a partner (IQR)	25 (23 to 27)	25 (23 to 27)	25 (24 to 26)	23 (22 to 26)	24 (24 to 26)
Median age difference of partnership (index ^b – at-risk ^a partner) (IQR), y	2 (-2 to 5)	1 (-2 to 5)	0.5 (-3.5 to 4.5)	3 (-1 to 5.5)	4 (1.5 to 5.5)
Gender composition of partnership					
Male/male	38 (38)	25 (41)	3 (25)	10 (42)	0 (0)
Male at-risk ^a /female index ^b	11 (11)	4 (7)	4 (33)	3 (13)	0 (0)
Female at-risk ^a /male index ^b	45 (44)	26 (43)	5 (42)	10 (42)	4 (100)
Female/female	7 (7)	6 (10)	0 (0)	1 (4)	0 (0)
Median duration of partnership (IQR), mo	12 (6 to 24)	12 (5 to 24)	14.5 (5 to 24)	11 (6 to 14)	36 (24 to 60)
Injecting and sexual behaviors, reported at partnership enrollment					
Median No. of days partners injected together, past month (IQR)	15 (8 to 21)	12 (6 to 17)	26.5 (20 to 30)	15 (10 to 22)	20 (17.5 to 25)
Any receptive needle sharing by at-risk ^a partner with index ^b partner, prior month = yes	12 (12)	9 (15)	2 (17)	0 (0)	1 (25)
Ancillary equipment sharing at all instances of the partners injecting together, prior month = yes	40 (40)	24 (39)	6 (55)	7 (58)	2 (50)
Median No. of other injecting partners, prior month	4 (2 to 10)	3.5 (2 to 6.5)	3 (1 to 10)	5 (3 to 15)	6 (2.5 to 14.5)
Partners had sexual relationship, prior month = yes	33 (33)	14 (23)	10 (83)	5 (21)	4 (100)
HCV characteristics of index partner					
Index partner ^b in HCV-seronegative viremic phase ^c at partnership enrollment = yes	7 (7)	1 (2)	4 (33)	2 (8)	0 (0)
Median HCV level of viremia (log ₁₀) of index partner ^b at partnership enrollment (IQR)	5.5 (4.6 to 6.3)	5.4 (4.5 to 6.1)	5.8 (2.4 to 6.5)	5.8 (4.9 to 6.4)	6.5 (5.6 to 7.0)

Abbreviations: HCV, hepatitis C virus; IQR, interquartile range.

^aAt-risk partner: enrolled as either HCV RNA negative or HCV-seronegative viremic.

^bIndex partner: enrolled as HCV RNA positive.

^cHCV-seronegative viremic phase: indicated by anti-HCV-negative and HCV RNA-positive test results.

(range, 1–9), for a total of 101 partnerships; 62 were HCV RNA negative, and 7 were in the HCV-seronegative viremic phase. Of those who were RNA negative, 14 were anti-HCV positive, indicating prior cleared infection.

Among the 69 at-risk participants, there were 25 new HCV infections (including the 7 persons who enrolled in the HCV-seronegative viremic phase and 6 of the 14 who enrolled as cleared infections), for an overall HCV incidence rate of 35.9 per 100 person-years (95% confidence interval [CI], 24.3–53.2). Among these 25, 20 (80%) were matched by genotype to at least 1 of their HCV-infected partners, whereas 5 did not match any partner's genotype and thus did not undergo sequencing and were grouped with those with no phylogenetic linkage. Of the 20 with at least 1 genotype-matched partner, 12 were phylogenetically linked, and thus likely transmissions. Thus, the proportion of newly identified infections with a phylogenetically linked HCV transmission event was 12/25 (48%).

Partnerships (n = 101) were followed for a median (IQR) of 6 (2–11) months; partnership characteristics are described overall and by group in Table 2. The 25 new HCV infections occurred among 40 partnerships (allowing for more than 1 partner per at-risk participant), and there were 61 partnerships with no new infection (group 1). Of the 40 partnerships with new infections, HCV transmission was corroborated in 12 (group 2), 24 were not phylogenetically matched (group 3), and 4 were not determined (group 4). Of note, 3 new infections occurred in

partnerships in which both partners were in the seronegative viremic phase, and all 3 of these were corroborated transmissions. The proportion of partnerships with a phylogenetically linked HCV transmission was 11.7% (12/101).

In regression analyses, we found no difference in the level of HCV viremia on the odds of having a corroborated transmission event (OR, 1.03 per log10 difference; 95% CI, 0.69–1.53) (Table 3). However, having an index partner who was in the HCV-seronegative viremic (acute infection) phase was associated with significantly higher odds of a transmission event (OR, 14.84; 95% CI, 2.77–79.48), as was injecting more days together in the past month (per injecting day OR, 1.11; 95% CI, 1.05–1.18), always sharing ancillary injection equipment (OR, 4.87; 95% CI, 1.45–16.35), being male in a partnership with a female (OR, 7.12; 95% CI, 1.14–44.44), and being in a sexual relationship with one's partner (OR, 9.74; 95% CI, 2.40–39.48). In multivariable analyses, index partners in the HCV-seronegative viremic phase (AOR, 28.02; 95% CI, 5.61–139.95), injecting more days together in the past month (AOR, 1.07; 95% CI, 1.01–1.14) per day of injecting together), and always sharing injection equipment (AOR, 5.32; 95% CI, 1.25–22.70) were independently associated with increased odds of corroborated transmission events. In sensitivity analyses, we found no substantive differences in the results using viral load measurements within a window of 3 months (compared with last value carried forward), nor when the HCV infection phase of the index

Table 3. Unadjusted and Adjusted ORs for Phylogenetically Corroborated Transmission Events vs no Transmission (GEE Analyses) Among 101 Prospectively Followed Young PWID Partnerships

Characteristic	Unadjusted OR (95% CI)	Adjusted OR (95% CI)
Demographics (at enrollment)		
Race of at-risk ^a partner = nonwhite	0.52 (0.15–1.84)	
Age of at-risk ^a partner	1.01 (0.86–1.19)	
Age difference of partnership, y	0.93 (0.84–1.03)	
Gender composition of partnership		
Male/male or female/female	Ref	Ref
Male at-risk ^a /female index ^b	7.12 (1.14–44.44)	1.91 (0.17–21.10)
Female at-risk ^a /male index ^b	1.97 (0.50–7.84)	0.29 (0.01–6.33)
Duration of partnership, mo	0.97 (0.95–1.01)	
Injecting and sexual behaviors (time updated)		
No. of days the partners injected together, past month	1.11 (1.05–1.17)	1.07 (1.01–1.14)
Any receptive needle sharing by at-risk ^a partner with index ^b partner, prior month = yes	1.44 (0.14–14.82)	
Ancillary equipment sharing at all instances of the partners injecting together, prior month = yes	4.87 (1.45–16.35)	5.32 (1.25–22.70)
Median No. of other injecting partners of the at-risk ^a partner	1.01 (1.00–1.03)	
Partners had sexual relationship, prior month = yes	9.74 (2.40–39.48)	7.77 (0.58–103.52)
HCV characteristics of index ^b partner (time updated)		
Index ^b partner in HCV-seronegative viremic phase ^c	14.84 (2.77–79.48)	28.02 (5.61–139.95)
HCV viremia of index ^b partner nearest visit (log10)	1.03 (0.69–1.53) ^d	0.90 (0.55–1.46) ^d

Bold indicates statistical significance ($P < .05$).

Abbreviations: CI, confidence interval; GEE, generalized estimating equation; HCV, hepatitis C virus; OR, odds ratio; PWID, people who inject drugs.

^aAt-risk partner: enrolled as either HCV RNA negative or HCV-seronegative viremic.

^bIndex partner: enrolled as HCV RNA positive.

^cHCV-seronegative viremic phase: indicated by anti-HCV-negative and HCV RNA-positive test results, as well as RNA-positive test results.

^dLast value carried forward.

partner variable (HCV-seronegative viremic phase yes/no) was removed. Results for when both partners enrolled in the early acute infection phase were excluded also yielded similar results, although the confidence intervals were wide.

CONCLUSIONS

We conducted a prospective study that followed young adult PWID injecting partnerships for new HCV infection, leveraging phylogenetic sequencing to provide evidence for the transmission within the partnerships. We did not observe increased odds of transmission among partnerships by the viremia level of the index partner, but instead found higher odds of transmission if the index partner was in the HCV-seronegative viremic phase (acute infection).

We had hypothesized that the HCV viral load of the index partner would be associated with increased risk of transmission to the at-risk partner, as has been shown for HIV [29, 30]. Although we found no increase in odds of transmission in association with quantitative level of viremia, having an index partner in the acute seronegative viremic phase was associated with significantly increased odds of transmission. This finding suggests that HCV transmissions occur in rapid succession, in which multiple PWID become infected at once, possibly before HCV infection is detectable via anti-HCV testing. It is possible that HCV infectivity is higher during this period due to the lack of neutralizing antibody response very early in infection, as is the case for HIV [31]. This suggests that while biomedical approaches such as TasP [13, 14, 32] and vaccines [33] have the potential to reduce HCV transmission by reducing community viral load, behavioral and testing interventions to reduce exposure to HCV during early infection are needed as well [3]. The lack of an association with HCV viral load, despite this positive finding in association with acute infection period, may be related to the low frequency of viremia testing in the positive partners [34] and the dynamic nature of HCV viremia in early infection [35], which hindered our ability to measure actual viremia near the time of transmission. Previous studies have shown that the frequency of viremia testing can vastly impact study findings, leading to underestimates of HCV transmission [34].

In either case, the results suggest that scaling up HCV testing of young adult PWID is urgently needed, including testing that includes the detection of viremia during the seronegative acute phase of infection [1]. Upstream approaches to reduce injecting of drugs, including with widespread provision of medication-assisted therapy [8], and overall injecting risk may also be useful, given that we found that within partnerships, the number of days injecting together and engaging in ancillary equipment sharing were additional independent risk factors for corroborated transmission events.

This study has strengths and limitations to note. Although other studies have examined HCV sequences and phylogenetic

clustering among PWID with HCV [36–39], and 1 study compared the overlap of phylogenetic clustering and PWID social networks [40], we not aware of any studies that have prospectively detected and corroborated transmission events within injecting partnerships identified a priori. The study is limited by the small number of partnerships, averaging 1 per month of active study time. The small number may be due to high levels of HCV concordance within drug-using partnerships, as reported in another study of PWID in San Francisco [41]. We included people who had previously cleared HCV (ie, anti-HCV positive but HCV RNA negative) and became re-infected, which may have impacted our incidence rate. However, it is unlikely that the HCV incidence rate was elevated by including those participants, as studies have shown that reinfection incidence is lower than primary infection incidence [42, 43]. In addition, we were not able to corroborate transmission or conclude no transmission within 4 partnerships with new HCV infection detected, because the level of viremia in the available sample was too low to conduct genotyping or sequencing, further limiting the sample size. Due to expense, we were able to measure the viral load in the index partner only approximately 1 time per year. As noted above, this limitation means that the level of viremia used in the analysis may not have reflected the level of viremia in the index at the time of transmission, masking a possibly real association. Lastly, all the behavioral variables were self-reported, and we used the responses of the at-risk partner in the analyses. In a previous analysis of Partner Study data, we found high concordance between injecting partners in reporting partnership characteristics including being in a sexual relationship, but low concordance on injecting risk behavior questions [44], which may have biased our results.

In summary, the odds of transmission were not related to level of HCV viremia but were greatly increased for those whose partners were in the seronegative viremic infection phase. These findings suggest that HCV transmission may happen in bursts of high risk, perhaps with partners other than regular injecting partners, that are difficult to pinpoint even with careful longitudinal study. The implications are that interventions to end the HCV epidemic in PWID will need to be broad enough to reduce community viral load and injecting in general, because sporadic periods of behavioral risk that co-occur with new infection are likely to escape more targeted interventions.

Acknowledgments

The authors thank the participants of this study and the UFO Study field staff for their support and assistance during data collection.

Financial support. This work was supported by the National Institutes of Health (R01 DA016017 to K.P.; U19 AI082630 to T.M.A.; K24 AA022586 to J.A.H.).

Potential conflicts of interest. The authors have no conflicts of interest.

References

1. Page K, Morris MD, Hahn JA, et al. Injection drug use and hepatitis C virus infection in young adult injectors: using evidence to inform comprehensive prevention. *Clin Infect Dis* 2013; 57(Suppl 2):S32–8.

2. Hagan H, Pouget ER, Des Jarlais DC, Lelutiu-Weinberger C. Meta-regression of hepatitis C virus infection in relation to time since onset of illicit drug injection: the influence of time and place. *Am J Epidemiol* **2008**; 168:1099–109.
3. Coffin PO, Rowe C, Santos GM. Novel interventions to prevent HIV and HCV among persons who inject drugs. *Curr HIV/AIDS Rep* **2015**; 12:145–63.
4. Morris MD, Shiboski S, Bruneau J, et al. Geographic differences in temporal incidence trends of hepatitis C virus infection among people who inject drugs: the InC3 collaboration. *Clin Infect Dis* **2017**; 64:860–9.
5. Suryaprasad AG, White JZ, Xu F, et al. Emerging epidemic of hepatitis C virus infections among young nonurban persons who inject drugs in the United States, 2006–2012. *Clin Infect Dis* **2014**; 59:1411–9.
6. Zibbell JE, Iqbal K, Patel RC, et al. Increases in hepatitis C virus infection related to injection drug use among persons aged ≤ 30 years - Kentucky, Tennessee, Virginia, and West Virginia, 2006–2012. *MMWR Morb Mortal Wkly Rep* **2015**; 64:453–8.
7. MacArthur GJ, van Velzen E, Palmateer N, et al. Interventions to prevent HIV and hepatitis C in people who inject drugs: a review of reviews to assess evidence of effectiveness. *Int J Drug Policy* **2014**; 25:34–52.
8. Tsui JI, Evans JL, Lum PJ, et al. Association of opioid agonist therapy with lower incidence of hepatitis C virus infection in young adult injection drug users. *JAMA Intern Med* **2014**; 174:1974–81.
9. Conteduca V, Sansonno D, Russi S, et al. Therapy of chronic hepatitis C virus infection in the era of direct-acting and host-targeting antiviral agents. *J Infect* **2014**; 68:1–20.
10. Feld JJ. Direct-acting antivirals for hepatitis C virus (HCV): the progress continues. *Curr Drug Targets* **2017**; 18:851–62.
11. Muir AJ. The rapid evolution of treatment strategies for hepatitis C. *Am J Gastroenterol* **2014**; 109:628–35; quiz 36.
12. Feld JJ, Jacobson IM, Hézode C, et al; ASTRAL-1 Investigators. Sofosbuvir and velpatasvir for HCV genotype 1, 2, 4, 5, and 6 infection. *N Engl J Med* **2015**; 373:2599–607.
13. Martin NK, Vickerman P, Dore GJ, et al; STOP-HCV Consortium. Prioritization of HCV treatment in the direct-acting antiviral era: an economic evaluation. *J Hepatol* **2016**; 65:17–25.
14. Hajarizadeh B, Grebely J, Martinello M, et al. Hepatitis C treatment as prevention: evidence, feasibility, and challenges. *Lancet Gastroenterol Hepatol* **2016**; 1:317–27.
15. Liang TJ. Current progress in development of hepatitis C virus vaccines. *Nat Med* **2013**; 19:869–78.
16. Walker CM. Designing an HCV vaccine: a unique convergence of prevention and therapy? *Curr Opin Virol* **2017**; 23:113–9.
17. Major M, Gutfraind A, Cui Q, et al. Model-based analysis of patient immunity profiles indicates that vaccination could reduce hepatitis C transmission via syringe sharing. *Sci Transl Med* **2018**; 10:eaa04496.
18. Garcia-Tejedor A, Maiques-Montesinos V, Diago-Almela VJ, et al. Risk factors for vertical transmission of hepatitis C virus: a single center experience with 710 HCV-infected mothers. *Eur J Obstet Gynecol Reprod Biol* **2015**; 194:173–7.
19. Hajarizadeh B, Grady B, Page K, et al; InC3 Study Group. Patterns of hepatitis C virus RNA levels during acute infection: the InC3 study. *PLoS One* **2015**; 10:e0122232.
20. Morris MD, Evans J, Montgomery M, et al. Intimate injection partnerships are at elevated risk of high-risk injecting: a multi-level longitudinal study of HCV-serodiscordant injection partnerships in San Francisco, CA. *PLoS One* **2014**; 9:e109282.
21. Page K, Hahn JA, Evans J, et al. Acute hepatitis C virus infection in young adult injection drug users: a prospective study of incident infection, resolution, and reinfection. *J Infect Dis* **2009**; 200:1216–26.
22. Tang W, Chen W, Amini A, et al. Diagnostic accuracy of tests to detect hepatitis C antibody: a meta-analysis and review of the literature. *BMC Infect Dis* **2017**; 17:695.
23. Sarrazin C, Gärtner BC, Sizmann D, et al. Comparison of conventional PCR with real-time PCR and branched DNA-based assays for hepatitis C virus RNA quantification and clinical significance for genotypes 1 to 5. *J Clin Microbiol* **2006**; 44:729–37.
24. Vermehren J, Kau A, Gärtner BC, et al. Differences between two real-time PCR-based hepatitis C virus (HCV) assays (RealTime HCV and Cobas AmpliPrep/Cobas TaqMan) and one signal amplification assay (Versant HCV RNA 3.0) for RNA detection and quantification. *J Clin Microbiol* **2008**; 46:3880–91.
25. Murphy DG, Willems B, Deschênes M, et al. Use of sequence analysis of the NS5B region for routine genotyping of hepatitis C virus with reference to C/E1 and 5' untranslated region sequences. *J Clin Microbiol* **2007**; 45:1102–12.
26. Yang X, Charlebois P, Gnerre S, et al. De novo assembly of highly diverse viral populations. *BMC Genomics* **2012**; 13:475.
27. Henn MR, Boutwell CL, Charlebois P, et al. Whole genome deep sequencing of HIV-1 reveals the impact of early minor variants upon immune recognition during acute infection. *PLoS Pathog* **2012**; 8:e1002529.
28. Tully DC, Ogilvie CB, Batorsky RE, et al. Differences in the selection bottleneck between modes of sexual transmission influence the genetic composition of the HIV-1 founder virus. *PLoS Pathog* **2016**; 12:e1005619.
29. Eshleman SH, Hudelson SE, Redd AD, et al. Treatment as prevention: characterization of partner infections in the HIV prevention trials network 052 trial. *J Acquir Immune Defic Syndr* **2017**; 74:112–6.
30. Wawer MJ, Gray RH, Sewankambo NK, et al. Rates of HIV-1 transmission per coital act, by stage of HIV-1 infection, in Rakai, Uganda. *J Infect Dis* **2005**; 191:1403–9.
31. Vaidya NK, Ribeiro RM, Liu P, et al. Correlation between anti-gp41 antibodies and virus infectivity decay during primary HIV-1 infection. *Front Microbiol* **2018**; 9:1326.
32. Hickman M, De Angelis D, Vickerman P, et al. Hepatitis C virus treatment as prevention in people who inject drugs: testing the evidence. *Curr Opin Infect Dis* **2015**; 28:576–82.
33. Scott N, McBryde E, Vickerman P, et al. The role of a hepatitis C virus vaccine: modelling the benefits alongside direct-acting antiviral treatments. *BMC Med* **2015**; 13:198.
34. Vickerman P, Grebely J, Dore GJ, et al; International Collaboration of Incident HIV and Hepatitis C in Injecting Cohorts (InC3); InC3 Collaborative Group. The more you look, the more you find: effects of hepatitis C virus testing interval on reinfection incidence and clearance and implications for future vaccine study design. *J Infect Dis* **2012**; 205:1342–50.
35. Hajarizadeh B, Grebely J, Applegate T, et al; ATAHc Study Group. Dynamics of HCV RNA levels during acute hepatitis C virus infection. *J Med Virol* **2014**; 86:1722–9.
36. Jacka B, Applegate T, Poon AF, et al. Transmission of hepatitis C virus infection among younger and older people who inject drugs in Vancouver, Canada. *J Hepatol* **2016**; 64:1247–55.
37. Cunningham EB, Jacka B, DeBeck K, et al. Methamphetamine injecting is associated with phylogenetic clustering of hepatitis C virus infection among street-involved youth in Vancouver, Canada. *Drug Alcohol Depend* **2015**; 152:272–6.
38. Jacka B, Applegate T, Krajdén M, et al. Phylogenetic clustering of hepatitis C virus among people who inject drugs in Vancouver, Canada. *Hepatology* **2014**; 60:1571–80.
39. Rodrigo C, Eltahla AA, Bull RA, et al; InC3 Collaborative. Phylogenetic analysis of full-length, early infection, hepatitis C virus genomes among people with intravenous drug use: the InC3 study. *J Viral Hepat* **2017**; 24:43–52.
40. Sacks-Davis R, Daraganova G, Aitken C, et al. Hepatitis C virus phylogenetic clustering is associated with the social-injecting network in a cohort of people who inject drugs. *PLoS One* **2012**; 7:e47335.
41. Rowe C, Santos GM, Raymond HF, Coffin PO. Social mixing and correlates of injection frequency among opioid use partnerships. *Int J Drug Policy* **2017**; 41:80–8.
42. Mehta SH, Cox A, Hoover DR, et al. Protection against persistence of hepatitis C. *Lancet* **2002**; 359:1478–83.
43. Page K, Osburn W, Evans J, et al. Frequent longitudinal sampling of hepatitis C virus infection in injection drug users reveals intermittently detectable viremia and reinfection. *Clin Infect Dis* **2013**; 56:405–13.
44. Evans JL, Morris MD, Yu M, et al. Concordance of risk behavior reporting within HCV serodiscordant injecting partnerships of young injection drug users in San Francisco, CA. *Drug Alcohol Depend* **2014**; 142:239–44.