Trouble with Bleeding: Risk Factors for Acute Hepatitis C among HIV-Positive Gay Men from Germany—A Case-Control Study

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Abstract

Objectives: To identify risk factors for hepatitis C among HIV-positive men who have sex with men (MSM), focusing on potential sexual, nosocomial, and other non-sexual determinants.

Background: Outbreaks of hepatitis C virus (HCV) infections among HIV-positive MSM have been reported by clinicians in post-industrialized countries since 2000. The sexual acquisition of HCV by gay men who are HIV positive is not, however, fully understood.

Methods: Between 2006 and 2008, a case-control study was embedded into a behavioural survey of MSM in Germany. Cases were HIV-positive and acutely HCV-co-infected, with no history of injection drug use. HIV-positive MSM without known HCV infection, matched for age group, served as controls. The HCV-serostatus of controls was assessed by serological testing of dried blood specimens. Univariable and multivariable regression analyses were used to identify factors independently associated with HCV-co-infection.

Results: 34 cases and 67 controls were included. Sex-associated rectal bleeding, receptive fisting and snorting cocaine/amphetamines, combined with group sex, were independently associated with case status. Among cases, surgical interventions overlapped with sex-associated rectal bleeding.

Conclusions: Sexual practices leading to rectal bleeding, and snorting drugs in settings of increased HCV-prevalence are risk factors for acute hepatitis C. We suggest that sharing snorting equipment as well as sharing sexual partners might be modes of sexual transmission. Condoms and gloves may not provide adequate protection if they are contaminated with blood. Public health interventions for HIV-positive gay men should address the role of blood in sexual risk behaviour. Further research is needed into the interplay of proctosurgery and sex-associated rectal bleeding.

Introduction

Hepatitis C virus (HCV) outbreaks among HIV-positive men who have sex with men (MSM) have been reported by clinicians in post-industrialized countries since 2000 [1,2]. Outbreaks in this population have been described in Paris [3–9], Amsterdam [10,11], London and Brighton [12,13], New York City [14], Sydney [15], and in major cities in Switzerland [16] and Germany [17]. Data on HIV serostatus are not routinely collected upon German hepatitis C notification [18], but recent epidemiological surveillance data demonstrated MSM to be the only group showing an increase in diagnosed hepatitis C since 2000 [19]. A large cohort of European HIV-positive MSM who deny injection drug use (IDU) showed a HCV prevalence of 6.6% [20].

Transmission of HCV typically requires direct blood exposure, e.g. through sharing needles or paraphernalia, or through contaminated blood products [21]. HCV has also been identified showing an increase in diagnosed hepatitis C since 2000 [19]. A large cohort of European HIV-positive MSM who deny injection drug use (IDU) showed a HCV prevalence of 6.6% [20].
studies of HCV-serodiscordant heterosexual couples have not yielded significant evidence for sexual transmission; so condom use for the prevention of HCV transmission has not been recommended for vaginal intercourse between monogamous HCV-serodiscordant sexual partners [33–36]. Large cohort studies of HIV-negative MSM showed that sexual behaviour, including unprotected anal intercourse (UAI), was not associated with prevalent HCV infections [37]; and those HCV seroconversions rarely observed among MSM without HIV infection could typically be attributed to injection drug use [10,30]. Certain sexual practices involving trauma of the rectal mucosa have been discussed as relevant risk factors among MSM [13,39]. Co-infections with bacterial sexually transmitted infections (STIs), especially ulcerative STIs such as syphilis or lymphogranuloma venereum (LGV) have also been proposed as risk factors for HCV transmission among HIV-positive MSM [7,9,10,24,32,40]. Furthermore, HIV infection has been proposed as risk factors for HCV transmission among STIs such as syphilis or lymphogranuloma venereum (LGV).

Hepatitis C is a major contributor to morbidity and mortality among people living with HIV in post-industrialised countries [42]. Immunosuppression and the HCV genotypes most prevalent among HIV-positive MSM are known to promote treatment failure for HCV-infection [2]. Hepatitis C prevention among HIV-positive MSM is a public health challenge as transmission modes are largely unknown and stigmatization of individuals who are already stigmatized may hamper health-seeking behaviour and disclosure of HCV-seropositivity [43].

The case-control study reported here was conducted to identify risk factors for acute hepatitis C among HIV-positive MSM in Germany, focusing on potential sexual, social, or nosocomial determinants.

**Methods**

**Ethical statement**

Informed consent was obtained from all participants involved. Because of the anonymity of the questionnaire data, no names and signatures were collected. Approval to having anonymous self-reported data stored and analyzed, and – for controls – to have a dried blood specimen tested for HCV antibodies, was declared by sending back a section of the participant’s information paper. Ethic committees of Charité University Clinics in Berlin and of the Medical School of Bonn University approved the study.

**Recruitment of cases and controls**

A case-control study was embedded in a national cross-sectional survey of STI knowledge, attitudes and behaviour among German MSM (KABaSTI study [44]). Participants for the case-control study were recruited between September 2006 and January 2008. No incentives were offered to potential participants.

As cases we included HIV-positive MSM who had been diagnosed with acute hepatitis C infection since 2000. They were recruited from an ongoing study of the treatment of acute hepatitis C among HIV-positive individuals at the University of Bonn, where they had been referred from HIV outpatient departments or practice-based physicians specialised in HIV care across Germany. HIV-positive MSM without a history of HCV infection, recruited by the same referring physicians and matched for age groups (±5 years), served as controls. Injection drug use was an exclusion criterion for cases and controls.

**Biological markers**

Patients were only chosen as controls if their referring physician, according to patient records, had classified them as HCV-negative. In addition, controls were asked to supply a dried blood specimen (DBS) on 903™ specimen collection paper (Whatman® GmbH, Germany), and to post it inside a multi-barrier pouch, along with the questionnaire, in a prepaid envelope. Capillary blood was collected either by the referring physician or by the participant himself, using an enclosed sterile lancet (Haemostilette®, ASID BONZ GmbH, Germany), following the instructions provided. The HCV serostatus of controls was confirmed with an automated chemiluminescent microparticle immunoassay (CMIA, ARCHITECT anti-HCV) from DBS [45].

**Questionnaires**

The KABaSTI study questionnaire has been described in detail elsewhere [46]. Briefly, an anonymous, 66-item self-completion questionnaire was distributed, covering socio-demographic data; sexual behaviour and diagnoses of STIs in the last 12 months; attitudes towards condom use to prevent transmission of STIs other than HIV; attitudes towards condom use with HIV-serodiscordant sexual partners; and HIV serostatus of non-steady sex partners.

Additionally, we developed a 23-item questionnaire to gather data on specific sexual, nosocomial, and other non-sexual exposures potentially associated with transmission of HCV, focusing on the time since 2000 as all cases included in this study seroconverted after 2000. The time frame for cases thus refers to the time frame between 2000 and hepatitis C diagnosis. Variables are described in table 1 and table 2.

**Sample size estimation**

We assumed a prevalence of suspected sexual exposures among controls of ±15%. This is in line with published literature on behavioural surveillance among MSM in Germany (e.g. attending sex parties, snorting cocaine, diagnosed with syphilis, and BDSM [bondage, sadism/masochism] or fisting with non-steady partners) [44,47]. To detect Odds Ratios of ≥3.0 with a power of 80% and an alpha-level of 5%, we aimed at 53 cases and 159 controls, at a case-control ratio of 1:3.

**Statistics**

Due to expected small numbers, two-sided Fisher’s Exact Test was used in univariable analysis to determine if an Odds Ratio (OR) was significantly different from 1. ORs and an approximated 95% confidence interval were calculated with SPSS 16. We looked for confounding factors and effect measure modifiers by stratification. Where effect measure modification was present, we constructed interaction terms that were tested individually for statistical significance.

To identify factors independently associated with acute hepatitis C, variables with ORs significantly different from 1 (p<0.10), were entered into multivariable logistic regression by stepwise forward and backward selection (Wald, cut-off at p<0.10). When effect measure modification was present among the remaining factors, we defined interaction terms that were tested for a statistically significant influence on the outcome parameter. Goodness-of-fit of the models was evaluated with Hosmer-Lemeshow test, and Nagelkerke’s pseudo-R-square to estimate the explanatory power of the models [48]. Venn diagrams were used to visualize and to compare overlap of exposures among cases and controls. The circular areas correspond with the respective presence of exposures.
Table 1. Socio-demographic data.

| Category | Subcategory | Cases (%) | Controls (%) |
|----------|-------------|-----------|--------------|
| N        |             | 34 (100.0)| 67 (100.0)   |
| Age      | 20–29 years | 4 (11.8)  | 9 (13.4)     |
|          | 30–44 years | 24 (70.6) | 48 (71.6)    |
|          | 45–65 years | 6 (17.6)  | 10 (14.9)    |
| Mean ± SD|             | 39 ± 8    | 39 ± 7       |
| Median (range) |          | 41 [22–65] | 41 [20–57]  |
| Age at first sex with a man | Mean ± SD | 17 ± 3 | 17 ± 2 |
|          | Median      | 17        | 17           |
| Year of HIV diagnosis | Mean ± SD | 1999 ± 5 | 1999 ± 7 |
|          | Median (range) | 2000 (1987–2007) | 2001 (1987–2007) |
| Self reported last CD4 count | >350/µL | 22 (64.7) | 44 (65.7) |
|          | 200–350/µL | 4 (11.8) | 13 (19.4) |
|          | <200/µL    | 2 (5.9)  | 3 (4.5)     |
|          | Unknown    | 6 (17.6) | 8 (11.9)    |
| Antiretroviral therapy | Presently taking | 28 (82.4) | 50 (74.6) |
| Screening for HCV | Ever offered | 27 (79.4) | 54 (79.4) |
| Year of HIV diagnosis | Mean ± SD | 2005 ± 1 | —            |
|          | Median (range) | 2005 (2003–2007) | — |
| City size | ≥500,000 | 31 (91.2) | 50 (74.6) |
| Education | General qualification for university | 17 (50.0) | 35 (52.2) |
| Employment status | Retired/unemployed | 13 (41.2) | 26 (38.8) |
| Relationship status | Single | 13 (38.2) | 26 (38.8) |
| Sexual identification | Gay | 33 (97.1) | 67 (100.0) |

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Results

Overall, 39 men were recruited as cases and 78 as controls. Of the cases, five had to be excluded: three had a history of IDU, one became HCV-infected before 2000, and one provided implausible responses. Of the controls, one was excluded because of IDU; eight because they were not HIV-positive, and two due to inconsistent responses. Thus, 34 cases and 67 controls were included in the univariable and multivariable analyses. Of the controls, 43 (64%) provided DBS. All specimens tested negative for HCV antibodies.

Socio-demographic characteristics of study population

The median age of respondents was 41 years, ranging from 20 to 65 years. Of the 34 cases, most were living in Berlin (n = 27), Frankfurt (n = 3), Hamburg (n = 3), or small cities in northern and south-western federal states. Of the 67 controls, most were living in Berlin (n = 37), Frankfurt (n = 10), Cologne/Bonn (n = 9), Mannheim (n = 1), the Ruhr (n = 4), or small cities in south-western federal states (n = 6). Cases and controls did not differ with respect to age, education, employment, sexual debut with a man, relationship status, ever being screened for HCV, year of HIV diagnosis, self-reported last CD4 count, or being on antiretroviral treatment. All participants but one self-identified as gay (table 1).

Univariable analysis: Sexual behaviour and STIs in the last 12 months, and attitudes towards condom use

Some differences were found between cases and controls with respect to sexual behaviour during the twelve months preceding study participation (table 2). More than ten sex partners were reported by 65% of the cases vs. 49% of the controls. Applying higher cut-off values for dichotomisation (more than twenty, or more than fifty sex partners), did not result in a statistically significant association with hepatitis C. Cases reported more anal intercourse with non-steady partners, and were more likely to report episodes of unprotected anal intercourse (UAI) with non-steady partners of unknown HIV serostatus than controls (65% vs. 38%). More cases than controls had non-steady sex partners who were HIV-positive (79% vs. 57%), and reported more diagnoses of syphilis, Chlamydia infection, or gonorrhoea (56% vs. 31%); and also less often considered condoms for STI-prevention (41% vs. 19%).

Among cases as among controls, a majority said they would not use a condom if sex partners were seroconcordant for HIV; but cases were less likely to reject any anal intercourse without a condom (12% vs. 33%).

Univariable analysis: Specific sexual exposures since 2000

As to the time period since the year 2000, 9% of cases reported UAI with a sex partner known to be HCV positive; so did 8% of controls (table 2). Group sex since the year 2000 was reported by 91% of cases vs. 66% of controls (OR = 5.4); and 44% vs. 21% said that, when having sex, this had been frequently or always in a group of men (table 2).

Anorectal trauma with subsequent visible bleeding was a frequent or regular experience for 21% of the cases (vs. 5% of the controls), showing a strong association with acute hepatitis C (OR = 5.5). Frequent or regular use of phosphodiesterase-5 (PDE-5) inhibitors was reported by 44% of cases vs. 18% of controls.

Receptive fisting was even more common: 50% of the cases vs. 22% of the controls reported receptive fisting since 2000; and 35% vs. 9% said they had been fisted ‘frequently or always when having sex’. Only 6 out of all 32 receptive ‘fisters’ (19%) said their partner(s) had mostly worn gloves (to cover hands and forearms) for fisting. Moreover, 12 out of 17 (71%) HCV/HIV co-infected, and 8 out of 15 (53%) HIV mono-infected receptive ‘fisters’ reported using some sort of collective lubricant supply. Cases had a 7.5 times higher odds of reporting a combination of frequent fisting and not using gloves (or sharing them) than controls did; and a 5.2 times higher odds of reporting a combination of frequent fisting and sharing of lubricant.

Univariable analysis: Nosocomial and other non-sexual exposures since 2000

We observed no differences between cases and controls regarding most nosocomial exposures. Blood or plasma transfusions before 1991 (when screening of donors for HCV antibodies was introduced in Germany), were as common among cases as among controls, as were multiple exposures to gastroscopies, colonoscopies, cystoscopies, major dental treatment, or acupuncture since the year 2000. Body piercings (21% vs. 8%), but not tattoos, were more common among cases than among controls. More cases than controls reported a history of multiple episodes of major surgery (32% vs. 16%). Surgery was associated with frequent group sex (OR = 5.69; p < 0.001), more than twenty sex partners in the previous twelve months (OR = 3.70; p = 0.008), frequent receptive (but not insertive) anal intercourse with non-steady sex partners (OR = 3.32; p = 0.015), and particularly with

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Table 2. Exposures and potential risk factors for acute hepatitis C; bold if included in multivariable analysis.

| Category                                      | Subcategory                        | Cases (%) | Controls (%) | Univariable OR (95%-CI) | p    | Multivariable Adj. OR (95%-CI) | p    |
|------------------------------------------------|------------------------------------|-----------|--------------|-------------------------|------|---------------------------------|------|
| **N**                                          |                                    | 34 (100.0)| 67 (100.0)   |                         |      |                                 |      |
| **Sexual behaviour in the previous 12 months**  |                                    |           |              |                         |      |                                 |      |
| Number of sex partners                         | More than 10                       | 22 (64.7)| 33 (49.3)    | 1.89 (0.81–4.42)        | 0.204| 0.939                           |      |
| **Sexual behaviour in the previous 12 months**  | More than 20                       | 17 (50.0)| 27 (40.3)    | 1.48 (0.65–3.40)        | 0.400| 0.641                           |      |
| **Anonymous sex partners**                     | More than 50                       | 7 (20.6) | 13 (19.4)    | 1.08 (0.39–3.01)        | 1.000|                                 |      |
| **Anonymous sex partners**                     | More than 50 per cent             | 16 (47.1)| 30 (44.8)    | 1.10 (0.48–2.51)        | 0.836|                                 |      |
| **Sex frequency**                              | Twice weekly or more               | 9 (26.5) | 17 (25.4)    | 1.06 (0.41–2.71)        | 1.000|                                 |      |
| **Met sex partners through the internet**       | Often or always                    | 18 (52.9)| 26 (38.8)    | 1.77 (0.77–4.08)        | 0.206|                                 |      |
| **Met sex partners in saunas**                 | Often or always                    | 5 (14.7) | 9 (13.4)     | 1.11 (0.34–3.62)        | 1.000|                                 |      |
| **Met sex partners at sex parties**            | Often or always                    | 10 (29.4)| 12 (17.9)    | 1.91 (0.73–5.02)        | 0.209|                                 |      |
| **Met sex partners at leather venues**         | Often or always                    | 8 (23.5) | 11 (16.2)    | 2.27 (0.77–6.70)        | 0.155|                                 |      |
| Insertive AI† with non-steady partners         | Frequently or always               | 21 (61.8)| 24 (35.8)    | 2.89 (1.23–6.79)        | 0.019|                                 | 0.347|
| **Receptive AI† with non-steady partners**     | Frequently or always               | 20 (58.8)| 26 (38.8)    | 2.25 (0.97–5.23)        | 0.062|                                 | 0.759|
| **BDSM**† or fisting with non-steady partners  | Frequently or always               | 10 (29.4)| 9 (13.4)     | 2.69 (0.97–7.44)        | 0.063|                                 | 0.708|
| **UAI with non-steady partners**               | One episode or more                | 22 (64.7)| 25 (37.3)    | 3.08 (1.30–7.28)        | 0.012|                                 | 0.288|
| **UAI with non-steady partners**               | Five episodes or more              | 15 (44.1)| 14 (20.9)    | 2.99 (1.22–7.33)        | 0.020|                                 | 0.653|
| STIs and condom attitudes                      |                                    |           |              |                         |      |                                 |      |
| History of syphilis                            | In the last 12 months              | 10 (29.4)| 9 (13.4)     | 2.69 (0.97–7.44)        | 0.063|                                 | 0.192|
| History of Chlamydia infection                 | In the last 12 months              | 7 (20.6) | 10 (14.9)    | 1.48 (0.51–4.30)        | 0.575|                                 |      |
| History of gonorrhoea                          | In the last 12 months              | 8 (23.5) | 12 (17.9)    | 1.41 (0.51–3.87)        | 0.599|                                 |      |
| History of any of the three above              | In the last 12 months              | 19 (55.9)| 21 (31.3)    | 2.78 (1.18–6.50)        | 0.020|                                 | 0.211|
| Non-steady partners with syphilis              | In the last 12 months              | 5 (14.7) | 10 (14.9)    | 0.98 (0.31–3.14)        | 1.000|                                 |      |
| Non-steady partners with HIV                   | Presently                          | 27 (79.4)| 38 (56.7)    | 2.94 (1.13–7.70)        | 0.029|                                 | 0.986|
| Would engage in UAI if sex partner is seroconcordant for HIV | | | | | | |
| Would never engage in any UAI                  |                                    | 4 (11.8) | 22 (32.8)    | 0.27 (0.09–0.87)        | 0.029|                                 | 0.296|
| Condoms not considered for prevention of STIs (other than HIV) | | | | | | |
| UAI† with person known to be HCV positive      |                                    | 3 (8.8)  | 5 (7.5)      | 1.20 (0.27–5.35)        | 1.000|                                 |      |
| Group sex                                      | Yes                                | 31 (91.2)| 44 (65.7)    | 5.40 (1.49–19.58)       | 0.007| 3.50 (0.84–14.52)                | 0.084|
| PDE-5 inhibitors**                             | Frequently or always†              | 15 (44.1)| 14 (20.9)    | 3.00 (1.22–7.33)        | 0.020|                                 | 0.786|
| Nitrite inhalants or large amounts of alcohol  | Frequently or always†              | 15 (44.1)| 12 (17.9)    | 3.62 (1.44–9.09)        | 0.008|                                 | 0.557|
| Dilative sex toys                              | Usage                              | 27 (79.4)| 37 (55.2)    | 3.13 (1.20–8.17)        | 0.028|                                 | 0.588|
| Frequently sharing dilative sex toys           |                                    | 3 (8.8)  | 2 (3.0)      | 3.15 (0.50–19.80)       | 0.332|                                 |      |
| Anal pain with bloody diarrhoea                | Repeatedly                         | 6 (17.6) | 4 (6.0)      | 3.38 (0.88–12.91)       | 0.082|                                 | 0.384|
| Rectal trauma with bleeding                    | Frequently or always†              | 7 (20.6) | 3 (4.5)      | 5.53 (1.33–23.00)       | 0.029| 6.19 (1.17–32.81)                | 0.032|
| Skin damage (BDSM†) with bleeding              |                                    | 8 (23.5) | 9 (13.4)     | 1.98 (0.69–5.72)        | 0.261|                                 |      |
| Receptive fisting                              | Yes                                | 17 (50.0)| 15 (22.4)    | 3.47 (1.43–8.39)        | 0.007|                                 |      |
| Frequently sharing of lubricant                |                                    | 12 (35.3)| 6 (9.0)      | 5.55 (1.86–16.57)       | 0.002|                                 |      |
| Frequent receptive fisting without gloves (or gloves shared) | | | | | | |
| Frequent receptive fisting and sharing of lubricant | | | | | | |
| Blood or plasma transfusion                    | Any before 1991                    | 2 (5.9)  | 5 (7.5)      | 0.78 (0.14–4.22)        | 1.000|                                 |      |

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having HIV-positive non-steady sex partners (OR = 7.56; p = 0.002), but not with fisting or age.

Among non-sexual exposures, consumption of nasally-administered drugs (NADs) – cocaine, amphetamines, or ketamine – was associated with HCV co-infection. More cases than controls reported that they had consumed NADs since the year 2000 (85% vs. 52%), and more had done so monthly or weekly (21% vs. 5%; OR = 5.5). Both groups reported sharing of snorting equipment (79% vs. 71%). A combination of NAD consumption and sharing of related equipment was especially common among cases (65% vs. 37%).

Multivariable logistic regression analysis

We identified frequent rectal trauma with bleeding, frequent receptive fisting without gloves, group sex, and consumption of NADs as independent risk factors for acute hepatitis C virus infection, with NAGELKERKE’s pseudo-R-square of 32.7% (table 1).

Effect measure modification was present between fisting/bleeding; fisting/consumption of NADs; fisting/group sex; and between group sex/consumption of NADs. By including these interaction terms, consumption of NADs and group sex were replaced by their respective interaction term (OR = 5.91; 95%-CI: 2.04–17.14; p = 0.001). NAGELKERKE’s pseudo-R-square for the remaining three variables was 34.6%. In 32 out of 34 cases (94%), at least one of these three factors was present. The model retained its robustness when restricting the multivariable analysis to controls with confirmed HCV-negative serostatus.

Overlap of exposures

There was an overlap between frequent receptive fisting without gloves and frequent rectal trauma with bleeding among cases (9% reported both exposures), whereas among controls, no such overlap was present (figure 1). All risk factors independently associated with HCV infection showed a higher degree of overlap among cases. Neither fisting nor rectal bleeding was associated with a recent history of bacterial STIs (p = 0.577; p = 0.512), a higher number of sex partners (p = 0.403; p = 1.000), or unprotected anal intercourse with a partner of unknown HIV serostatus (p = 0.278; P = 0.501). Among cases, but not among controls, there was substantial overlap between a history of multiple episodes of major surgery and rectal bleeding, or use of PDE-5-inhibitors (figure 2).

Discussion

This case-control study adds information on risk factors for acute hepatitis C among gay men with HIV infection. Our findings suggest that sexual and sex-associated exposures form distinct patterns that are particularly prevalent in this group. Previous research has shown that outbreaks of hepatitis C among HIV-positive men in Europe were linked to MSM-specific viral clusters of HCV [1,3,13,49], unrelated to injection drug use, suggesting MSM specific – but not necessarily entirely sexual – transmission modes.

Risk patterns identified by this study encompass frequent rectal trauma with bleeding, frequent engagement in receptive fisting without gloves, group sex and consumption of nasally administered drugs (NADs). To our knowledge, this is the first study to specifically collect self-reported data on sex-associated mucosal trauma with subsequent bleeding.

We think it is plausible to assume that the amount of visible bleeding reported by respondents suggests even higher rates of
bleeding that remained unnoticed. Our findings may be corroborated by showing a marked overlap of exposures among cases, e.g. 'receptive fisting and rectal bleeding' or 'history of major surgery and rectal bleeding'. This is consistent with the concept of causal pies in epidemiology, which describes how several causal components can act in concert to produce an effect [50].

Given the strong association of reported surgery with a high number of sex partners, group sex, use of PDE-5 inhibitors (and thus prolonged anal sex), and receptive anal intercourse, we hypothesize that a substantial proportion of the reported surgical interventions had been due to genital or anal condylomata, which are common among HIV-positive gay men with multiple sexual partners [51–53]. All types of common proctosurgical interventions for condylomata result in a traumatized anorectal mucosa with lesions and a considerable likelihood of postoperative bleeding. A history of proctosurgery and fast postoperative re-uptake of anal intercourse could explain the overlap of multiple surgical interventions and frequent rectal bleeding.

Our study revealed a considerable proportion of HIV-seroconcordant non-steady sexual partners, contributing to an increased chance of exposure to HCV. Not using condoms with HIV-seroconcordant partners ('serosorting') has been described as a risk management approach [54–56] that might increase the likelihood of acquiring other STIs than HIV [57–61].

HCV retains viability upon environmental exposure to room temperature and drying for at least sixteen hours [62]. This may affect the risk through sharing of equipment for NADs, sharing of sex toys or lubricant, or sharing of sex partners in a group sex setting.

**Figure 1. Venn diagram for overlap of exposures (frequent receptive fisting without gloves, frequent anal bleeding, group sex & consumption of NADs).** *Unexposed:* Respondents who neither had been frequently fisted without gloves (or with gloves that were shared), nor frequently experienced anal bleeding when having sex, nor reported group sex activities plus consumption of NADs. The circular areas correspond with the respective presence of exposures; intersecting areas reflect only roughly the given percentages.

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**Figure 2. Venn diagram for overlap of exposures (frequent use of PDE-5 inhibitors, frequent anal bleeding, history of major surgery).** *Unexposed:* Respondents who neither had frequently used PDE-5 inhibitors, nor frequently experienced anal bleeding when having sex, and who did not report multiple surgical interventions. The circular areas correspond with the respective presence of exposures; intersecting areas reflect only roughly the given percentages.

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setting. Non-injecting recreational use of amphetamines, cocaine, or ketamine is common among gay men with multiple partners, specifically HIV-positive gay men [47,51,63]. These substances may be administered orally, rectally or intra-nasally.

The equipment for intranasal administration is often shared and may come into contact with mucosal secretions or blood from fellow users. Traumatized nasal mucosa, reactive hyperemia, and epistaxis have been shown to result from snuffing cocaine powder; blood and HCV were found in nasal secretions of intranasal drug users, and transfer of contaminated blood onto shared snuffing implements has been demonstrated [64–66]. A study among female non-injecting drug users found that shared implements are likely to serve as vectors for HCV [67]. Given all the above, a rolled bank note that is being circulated at a commercial sex party might be sufficient to expose consumers to HCV-contaminated blood, even without knowingly sharing utensils. While it is possible that the association with drug use reflects residual confounding as suggested by others [2], we see no reason to believe that this known transmission route is negligible for gay men.

Our observations are in line with other studies, particularly regarding exposures like fisting, group sex, intranasal drug use, or a combination of these factors [10,13,39,43,68]. Other research suggests that organized sex parties of predominantly HIV-positive core groups, together with rectal STIs (Shigella, LGV, or rectal gonorrhoea) constitute drivers of sexual HCV transmission. Core groups represent sexual networks characterized by a high number of sex partners who are interconnected [69]. It has been demonstrated that such core groups exist among gay men (In this study nearly all respondents self-identified as gay. As sexual identity is a key factor for forming and sustaining sexual networks, we speak of ‘gay men’ instead of ‘MSM’ [70]). It has further been shown that STIs – particularly rectal STIs with few or no symptoms – accumulate in such groups [57–61]. Despite the substantial overlap with consumption of NADs and group sex, fisting and sex-associated rectal bleeding were not associated with a recent history of bacterial STIs, with a higher number of sex partners, or unprotected anal intercourse. Fisting and sex-associated rectal bleeding might therefore be regarded as additional causal pies, rather than proxy measures for being part of an STI core group, as suggested by other research groups [13].

HCV transmission has often been conceptualized as a pathogen being transferred from an infected insertive man to his receptive partner through his seminal fluid. However the infectivity of semen-derived HCV is being debated; and the presence of HCV RNA in seminal fluid has been demonstrated to be intermittent, and to be independent from blood HCV RNA concentration, immune status, or – in most studies – from HIV co-infection [22,23,25,29,71–75]. Given the strong association with fisting and sex-related rectal bleeding in our study, we suggest that blood rather than semen is the critical medium. An insertive partner’s fist (or penis), contaminated with blood, might serve as a vector for subsequent receptive partners in a group sex session, when condoms or gloves are either not applied, or not changed for every new partner – particularly when using a collectively shared supply of lubricant. Lesions in the anorectal mucosa – from fisting, prolonged anal intercourse, or rectal STIs – could serve both as a portal of entry and as a source of infection. Such a vector hypothesis might explain why this and other studies [13,28] failed to show an independent association of UAI on HCV/HIV co-infection.

Limitations

A lower than expected response from participating physicians precluded individual matching of study participants. Because of small numbers of cases and controls, the power of the study may not be sufficient to detect less frequent risk factors that may be masked by potential confounders. Despite attempts to minimize recall bias by including only individuals with acute hepatitis C and documented seroconversion, some recall bias – especially regarding the time frame ‘since the year 2000’ – cannot be ruled out. Misclassification might have occurred for controls that did not provide dried blood specimens (DBS) and therefore could not be confirmed to be truly seronegative for HCV. However, it is likely that HCV-positives would have been already identified, as German guidelines recommend frequent routine screening of liver enzymes and HCV antibodies among people with HIV [76], as indicated by the high proportion that had been previously screened for HCV antibodies (table 1). Moreover, as a case-cohort design approximates true relative risks in a better way than Odds Ratios in classical case-control studies do [77,78], this is considered a minor limitation.

It should be underlined that anorectal STIs are profoundly underdiagnosed [79]; hence, the association between hepatitis C and STIs may be underestimated in our study.

While the specific hepatitis C questionnaire addressed the time period prior to seroconversion, the broader KABSTI questionnaire aimed at the twelve months before study participation, a time frame not exclusively preceding HCV co-infection. It is possible that behaviour or attitudes changed after being diagnosed with and being counselled on acute hepatitis C; this might be particularly applicable to the questions on unprotected anal intercourse or attitudes toward condom use, leading to an underestimation of levels of UAI before HCV acquisition among the cases. Given these limitations, and given the fact that all behavioural data is self-reported, causal inferences have to be drawn with caution.

Conclusions and recommendations

There is a need for specific public health interventions targeting HIV-positive gay men, which address the heightened risk of HCV transmission within their sexual networks. Sexual transmission of HCV seems to occur when HCV-contaminated blood is passed on in a context of situationally-increased HCV prevalence (group sex in HIV-positive gay sexual networks), and mucosal integrity is disrupted due to inflammatory or ulcerative STIs and/or prolonged (PDE-5 inhibitors) or traumatising genito- anal contacts. This risk is heightened in a setting of fisting and sexually-induced anal haemorrhage, particularly following anorectal surgical interventions. If condoms are used but not changed for every new partner, they might work as a vector for blood-borne viruses in a context of sex in a group, regardless of whether the insertive partner is infected with HCV. Similar cross-infection could be facilitated by sharing a glove used during fisting, or a rolled bank note used for consuming NADs. This evidence suggests that public health messages addressing sexual HCV transmission among gay men should focus on the avoidance of sexual and sex-associated exposure to blood rather than seminal fluid. Prevention efforts are needed to communicate sexual or sex-associated routes of transmission for HCV that may not be addressed by messages that focus on the use of condoms.

Proprietors of sex venues need to be educated about HCV transmission risks, and thus to avoid offering shared lubricant supplies. Distributing individual non-traumatising snorting tubes alongside condoms to persons visiting respective sex venues could be considered for harm reduction.

The association of nosocomial risk factors among HIV-positive gay men needs to be elucidated – specifically the interplay of proctosurgery, mucosal disintegrity and the timing of restarting...
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References

1. Van de Laar T, Pybus O, Brustein S, Brown D, Nelson M, et al. (2009) Evidence of a large, international network of HCV transmission in HIV-positive men who have sex with men. Gastroenterology 136: 1609–1617.

2. van de Laar T, Reinosky DS, Kottkamp H, et al. (2009) Acute hepatitis C in HIV-infected men who have sex with men: an emerging sexually transmitted infection. Sex Transm Dis 36: S31–S36.

3. Serpaggi J, Chaix ML, Batsis D, Dupont C, Vallet-Pichard A, et al. (2006) Sexually transmitted acute infection with a clade genotype 4 hepatitis C virus in HIV-infected men and inefficacy of early antiviral therapy. AIDS 20: 233–240.

4. Gambili I (2005) Acute hepatitis C infection in HIV positive men who have sex with men. Paris, France, 2001; 2004. Euro Surveill 10: 74.

5. Ghor J, Tibault V, Delaugerre C, Fontaine H, Lortholary O, et al. (2008) Sexually transmitted hepatitis C virus superinfection in HIV-infected men who have sex with men. AIDS 22: 658–661.

6. Ghor J, Deveaux C, Goujard C, Garrigue I, Saïchi N, et al. (2006) Increase in hepatitis C virus incidence in HIV-infected patients followed up since primary infection. Sex Transm Infect 82: 458–460.

7. Ghor J, Pierre-Joncas S, Thibault V, Duvoisin C, Tubiana R, et al. (2004) Acute hepatitis C in HIV-infected men who have sex with men. Med Sci (Paris) 20: 303–306.

8. Bouvet E (2005) Sexual practices and transmission of HAV and HCV. Euro Surveill 10: 74.

9. Ghor J, Larsen C, Piroth L, Duval X, Aupinier I, et al. (2009) Evidence of Ongoing Epidemic Sexual Transmission of HCV (2006–2007) among HIV-1-infected Men Who Have Sex with Men: France. 16th Conference on Retroviruses and Opportunistic Infections. Montreal. Poster abstract 400.

10. Van de Laar T, Van der Bij AK, Prins M, Brustein SM, Brinkman K, et al. (2007) Increase in HIV incidence among men who have sex with men in Amsterdam most likely caused by sexual transmission. J Infect Dis 196: 230–238.

11. Van den Berg G, Blok W, Barrands H, Regez K, Froseen J, et al. (2009) Rapid Rise of Acute HCV Cases Among HIV-infected Men Who Have Sex With Men. Amsterdam. 16th Conference on Retroviruses and Opportunistic Infections. Montreal. Poster abstract 304.

12. Brose W, Rosd K, Gillette J, Atkins M, Mandaliya S, et al. (2004) Increased numbers of acute hepatitis C infections in HIV positive homosexual men: is sexual transmission feeding the increase? Sex Transm Infect 80: 326–327.

13. Danta M, Brown D, Bhagani S, Pybus OG, Sabin CA, et al. (2007) Recent epidemic of acute hepatitis C virus in HIV-positive men who have sex with men linked to high-risk sexual behaviours. AIDS 21: 983–991.

14. Fierer D, Fishmen S, Uriel A, Carriero D, Factor SH, et al. (2009) Increase in HCV incidence among men who have sex with men. Int J STD AIDS 20: 725–729.

15. Jin F, Prestage GP, Matthews G, Zablotska I, Rawstorne P, et al. (2010) Prevalence of Risk Factors for Hepatitis C Virus in HIV-Infected and HIV- Hepatitis C Virus-Coinfected Patients. Sex Transm Dis 37: 367–370.

16. Clarke A, Kajaleegaram R (2006) Hepatitis C transmission – where are we now? Int J STD AIDS 17: 74–90.

17. Minko E, Baldo V, Baldoni T, Trivello R, Faccioni A (2006) Intramuscular transmission of hepatitis C virus infection. Eur J Epidemiol 21: 293–297.

18. Filippini P, Coppola N, Scolastico C, Rossi G, Onofrio M, et al. (2001) Does HIV Infection Favor the Sexual Transmission of Hepatitis C? Sex Transm Dis 28: 723–729.

19. Giaccia M, Cappelli F, Gentili G, Maini A, Lepri AC, et al. (1997) Incidence and determinants of hepatitis C virus infection among individuals at risk of sexually transmitted diseases attending a human immunodeficiency virus type 1 testing program. Sex Transm Infect 73: 333–337.

20. Tahan V, Karaca C, Yildiz B, Reiswich D, Aroustrup K N (2005) Sexual transmission of HCV between spouses. Am J Gastroenterol 100: 821–824.

21. Neumayr G, Probst A, Schwaighofer H, Jodl M, Vogel W (1999) Lack of evidence for the heterosexual transmission of hepatitis C. QJM 92: 305–308.

22. Wyllie R, Robertson JR, Brettle RP, McAllister L, Prescott L, et al. (1997) Absence of hepatitis C virus transmission but frequent transmission of HIV-1 from sexual contact with doubly-infected individuals. J Infect 35: 163–166.

23. Vandelli A, Renzo F, Romano` L, Del Piero AC, et al. (1997) Absence of evidence of sexual and heterosexual transmission of hepatitis C among homosexual couples: results of a 10-year prospective follow-up study. Am J Gastroenterol 99: 853–859.

24. Jin F, Prestage GP, Kippax SC, Kaldor JM, Dore JJ, et al. (2005) Prevalence and risk factors of hepatitis C in homosexual men: data from two cohorts of HIV-negative and HIV-positive men in Sydney, Australia. Sex Transm Infect 81: 25–29.

25. Vu M, Hsu M, Wells C, et al. (2004) Unsafe sex and increased incidence of hepatitis C virus infection among HIV-infected men who have sex with men: the Swiss HIV Cohort Study. Clin Infect Dis 41: 395–402.

26. Vogel M, Detertling K, Wiegand J, Grüner NH, Baumgarten A, et al. (2009) Initial presentation of acute hepatitis C virus (HCV) infection among HIV-negative and HIV-positive individuals-experience from 2 large German networks on the study of acute HCV infection. Clin Infect Dis 49: 317–319.

27. Walter J, Radan D, Clauw H, Hanoumou O, Stark R (2005) [Hepatitis B and C risk factors in Germany-results of the national surveillance]. Gesundheitswesen 67: 441–447 [German].

28. Robert Koch Institute (2009) [Viral Hepatitis B, C, and D in 2008 in Germany]. Epidemiologisches Bulletin 58: 197 [German].

29. Rockstroh JK, Mocroft A, Soriano V, Tural C, Losso MH, et al. (2005) Influence of hepatitis C virus infection on HIV-1 disease progression and response to highly active antiretroviral therapy. J Infect Dis 192: 992–1002.
43. Owen G (2008) An ‘elephant in the room’? Stigma and hepatitis C transmission among HIV-positive ‘serostrating’ gay men. Cult Health Sex 10: 601–610.
44. Schmidt AJ, Marcus U, Hamouda O (2007) [KABaSTI-study - Knowledge, Attitudes, and Behaviour as to Sexually Transmissible Infections. 2nd Generation Surveillance for HIV and other STIs among German Men Who Have Sex With Men. Report to the Federal Ministry of Health]. Berlin: Robert Koch Institute [German].
45. Tuanillon E, Mondain AM, Merosnef F, Ottomani L, Piot MC, et al. (2010) Dried blood spot for hepatitis C virus serology and molecular testing. Hepatology 51: 752–759.
46. Marcus U, Schmidt AJ, Kollan C, Hamouda O (2009) The denominator problem: Estimating MSM-specific incidence of sexually transmitted infections and prevalence of HIV using population sizes of MSM derived from Internet surveys. BMC Public Health 9: 181.
47. Bochow M, Wright MT, Lange M (2004) [Gay Men and AIDS: Risk management in times of social normalization of an infectious disease. A survey on behalf of the Federal Center for Health Education]. Berlin: Deutsche AIDS-Hilfe, AIDS-Forum DAH Vol. 48 [German].
48. Nagelkerke N (1991) A Note on a General Definition of the Coefficient of Determination. Biometrika 78: 691–692.
49. Gotz HM, van Doornum G, Niesters HG, den Hollander JG, Thio HB, et al. (2005) A cluster of acute hepatitis C virus infection among men who have sex with men—results from contact tracing and public health implications. AIDS 19: 969–974.
50. Rothman KJ, Greenland S (2005) Causation and causal inference in epidemiology. Am J Public Health 95 Suppl 1: S144–150.
51. Bochow M, Schmidt AJ, Grote S (2010) [Gay Men and AIDS: Sexual Lifestyles and Risk Management 2007. 8th Follow Up Survey on Behalf of the Federal Center for Health Education]. Berlin: Deutsche AIDS-Hilfe, AIDS-Forum DAH Vol. 55 [German].
52. Wieczorek R, Brockmeyer NH, Kreuter A (2006) [Human papilloma virus-induced disease in HIV-positive patients]. Hautarzt 57: 994–998 [German].
53. Madiroka T, Jaspers C, Elbersbrink P, Hoepelman A (2008) HIV-related anogenital disease and HIV infection: not always ‘ordinary’ condyloma acuminate. Neth J Med 66: 98–102.
54. Van Kesteren NM, Hospers HJ, Kok G (2007) Sexual risk behavior among HIV-positive men who have sex with men: a literature review. Patient Educ Couns 63: 5–20.
55. Parsons JT, Schrimshaw EW, Wolitski RJ, Halkitis PN, Purcell DW, et al. (2005) Sexual harm reduction practices of HIV-seropositive gay and bisexual men: serostrating, strategic positioning, and withdrawal before ejaculation. AIDS 19 Suppl 1: S13–S25.
56. Mao L, Crawford JM, Hospers HJ, Prestage GP, Grulich AE, et al. (2006) ‘Serostrating’ in casual anal sex of HIV-negative gay men is noteworthy and is increasing in Sydney, Australia. AIDS 20: 1204–1206.
57. Jin F, Prestage GP, Zablotska I, Rawstorne P, Kippax SC, et al. (2007) High rates of sexually transmitted infections in HIV positive homosexual men: data from two community based cohorts. Sex Transm Infect 83: 397–399.
58. Jin F, Prestage GP, Zablotska I, Rawstorne P, Kippax SC, et al. (2007) Sexual harm reduction practices of HIV-seropositive gay and bisexual men: serostrating, strategic positioning, and withdrawal before ejaculation. AIDS 19 Suppl 1: S13–S25.
59. Jin F, Prestage GP, Zablotska I, Rawstorne P, Kippax SC, et al. (2007) High rates of sexually transmitted infections in HIV positive homosexual men: data from two community based cohorts. Sex Transm Infect 83: 397–399.
60. Schmidt AJ, Marcus U, Toppich J, Bochow M (2009) Serosorting Among German Men Who Have Sex With Men. Implications for Community