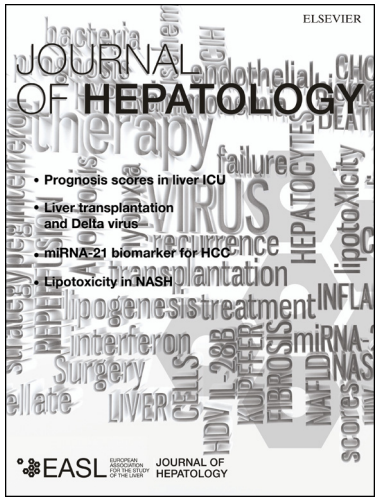
Accepted Manuscript



Fracture risk in hepatitis C virus infected persons: results from the DANVIR cohort study

Ann-Brit Eg Hansen, Lars Haukali Omland, Henrik Krarup, Niels Obel on behalf of the DANVIR cohort study

PII: S0168-8278(14)00151-2
DOI: http://dx.doi.org/10.1016/j.jhep.2014.03.007
Reference: JHEPAT 5068

To appear in: *Journal of Hepatology*

Received Date: 22 December 2013
Revised Date: 6 March 2014
Accepted Date: 7 March 2014

Please cite this article as: Eg Hansen, A-B., Haukali Omland, L., Krarup, H., Obel, N., on behalf of the DANVIR cohort study Fracture risk in hepatitis C virus infected persons: results from the DANVIR cohort study, *Journal of Hepatology* (2014), doi: http://dx.doi.org/10.1016/j.jhep.2014.03.007

Title

Fracture risk in hepatitis C virus infected persons: results from the DANVIR cohort study.

Authors

Ann-Brit Eg Hansen[1*], Lars Haukali Omland[2*] (*joint first authors), Henrik Krarup[3] and Niels Obel[2]; on behalf of the DANVIR cohort study

Affiliations

[1] Department of Pulmonary and Infectious Diseases, Copenhagen University Hospital, Nordsjællands Hospital, Hillerød, Denmark.

[2] Department of Infectious Diseases, Copenhagen University Hospital, Rigshospitalet, Copenhagen, Denmark.

[3] Section of Molecular Diagnostics, Clinical Biochemistry, Aalborg University Hospital, Aalborg Hospital, Aalborg, Denmark

Corresponding author:

Ann-Brit Eg Hansen, Department of Infectious Diseases, Department of Pulmonary and Infectious Diseases, Copenhagen University Hospital, Nordsjællands Hospital. Dyrehavevej 29, 3400 Hillerød. Denmark.

Telephone number: +45 4829 6974, Fax number: +45 4829 3909

e-mail: ann-brit.eg.hansen@regionh.dk

Electronic word count: 4948 (including the abstract, references, tables, and figure legends)

Number of figures and tables: 2 figures and 3 tables

Lists of abbreviations:

HCV: hepatitis C virus; CI: confidence interval; aIRR: adjusted incidence rate ratio; BMD: bone mineral density; CRS: Civil Registration System; DNPR: Danish National Hospital Registry; ICD-8: International Classification of Diseases, revision 8; ICD-10: International Classification of Diseases, revision 8; IDU: intravenous drug use; CCI: Charlsons Comorbidity Index; IRR: incidence rate ratio; PY: person years of observation

Conflicts of interest: '

N Obel received grants from Roche, Bristol-Meyers Squibb, Merck Sharp and Dohme, GlaxoSmithKline, Abbott, Boehringer Ingelheim, Janssen-Cilag, and Swedish-Orphan Drugs. All other authors report no conflict of interest.

Financial support: No financial support was provided for this study

ABSTRACT

Background and aims: The association between Hepatitis C virus (HCV)-infection and fracture risk is not well characterized. We compared fracture risk between HCV-seropositive (HCV-exposed) patients and the general population and between patients with cleared and chronic HCV-infection.

Methods: Outcome measures were time to first fracture at any site, time to first low-energy and first non low-energy (other) fracture in 12,013 HCV-exposed patients from the DANVIR cohort compared with a general population control cohort (n = 60,065) matched by sex and age. Within DANVIR, 4500 patients with chronic HCV-infection and 2656 patients with cleared HCV-infection were studied.

Results: Compared with population controls, HCV-exposed patients had increased overall risk of fracture [adjusted incidence rate ratio (aIRR) 2.15, 95% Confidence Interval (CI) 2.03 – 2.28], increased risk of low-energy fracture (aIRR 2.13, 95% CI 1.93 – 2.35) and of other fracture (aIRR 2.18, 95% CI 2.02 – 2.34). Compared with cleared HCV-infection, chronic HCV-infection was not associated with increased risk of fracture at any site (aIRR 1.08, 95% CI 0.97 – 1.20), or other fracture (aIRR 1.04, 95% CI 0.91 – 1.19). The aIRR for low-energy fracture was 1.20 (95% CI 0.99 – 1.44).

Conclusions: HCV-exposed patients had increased risk of all fracture types. In contrast, overall risk of fracture did not differ between patients with chronic versus cleared HCV-infection, although chronic HCV-infection might be associated with a small excess risk of low-energy fractures. Our study suggests that fracture risk in HCV-infected patients is multi-factorial and mainly determined by lifestyle-related factors associated with HCV-exposure.

Key words: fracture risk, chronic HCV, cleared HCV, comorbidity.

INTRODUCTION

It is well known that advanced liver disease caused by hepatitis C virus (HCV) is associated with reduced bone mineral density (BMD) [1]. It remains controversial whether chronic HCV-infection has detrimental effect on bone health in the absence of advanced liver disease or other risk factors. Some studies have reported low BMD among non-cirrhotic HCV-infected patients [2, 3] while other studies have indicated that HCV-infected patients without significant liver disease or alcohol/drug use have BMD values similar to age-matched controls [4, 5].

Osteoporotic fractures have substantial public health impact [6] and it is therefore important to establish whether the reported increased risk of low BMD in HCV-infected individuals translate into increased incidence of fractures. Only few studies have examined fracture risk in HCV-monoinfected patients. In a large cohort study, Lo Re *et al*. [7] observed increased risk of hip fractures among patients with HCV-infection compared with HCV-uninfected persons. This study did not report results for viremic versus nonviremic patients and the mechanisms for the increased fracture risk are not well understood. It remains important to further examine the association between fracture risk in HCV-seropositive individuals and the contribution of chronic viremia versus the contribution of lifestyle related risk factors associated with HCV-exposure such as illicit drug use, alcohol abuse, poor nutrition and increased risk of trauma.

In HIV-infection, it is well established that HCV-coinfection is associated with increased risk of fracture [8-10]. Recently, we extended these findings by demonstrating that HIV/HCV-coinfected patients had increased risk of both low-energy and high-energy fractures compared to both HIV-monoinfected patients and population controls [8]. The increased risk of high-energy fractures suggests that increased risk of trauma contribute to fracture risk in patients with HIV/HCV-coinfection. However HIV/HCV-coinfected patients represent an extremely marginalized group and the results may not be generalizable to HCV-monoinfected persons.

Additional data therefore are needed to clarify the association between HCV-infection and risk of fracture. In the present study, we compare the risks of fracture at any site, low-energy fractures, and other fractures including high-energy fractures in a Danish cohort of HCV-seropositive (HCV-exposed) patients and a population-based comparison cohort matched on age and sex. We further examine the incidence of low-energy and other fractures within the HCV cohort, comparing patients who were viremic (HCV RNA-positive) with those who were nonviremic (HCV RNA-negative) following a positive test for HCV antibodies.

PATIENTS AND METHODS

Study design

We used a cohort design to conduct two sub-studies: In sub-study 1, fracture risk was compared between Danish HCV-exposed patients and a population-based comparison cohort matched on age and sex. In sub-study 2, we used a cohort of HCV-antibody positive patients to compare fracture risk between HCV RNA-positive *(viremic)* and HCV RNA-negative *(nonviremic)* patients. Patients were included in the period 1995 – 2007 and followed until 2010.

Setting

Denmark has a population of 5.4 million [11], with an estimated HCV prevalence of 0.4 % [12]. HCV-infected patients are treated in hospital departments that are specialised in infectious diseases, gastroenterology, or hepatology. Medical care, including antiviral treatment, is provided free-of-charge to all HCV-infected residents. Although with some uncertainty, it has been estimated that only 2% of the Danish HCV-infected population has been treated with interferon [13].

Data sources

We used the unique 10-digit civil registration number assigned to all Danes at birth or immigration [14] to link the following data sources:

Danish HCV cohort (DANVIR): HCV-exposed patients were identified from the DANVIR cohort, which comprises all patients tested for HCV in 14 of the 18 laboratories that perform such testing in Denmark [15, 16]. DANVIR data includes results and dates of HCV antibody tests (from 1991 onward) and HCV RNA tests (from 1995 onward). While HCV antibody tests are performed in all participating DANVIR centres, most HCV RNA measurements are done in one centre (Department of Clinical Biochemistry, Aalborg University Hospital), as described previously [17]. When the DANVIR cohort was established, it was estimated to include more than 90% of all Danish patients tested for HCV RNA and the majority of patients tested for HCV antibodies [15, 16].

Civil Registration System (CRS): Established in 1968, the CRS maintains information on vital status and migration for all Danish residents [14]. We used CRS to select the comparison cohort and to obtain information on dates of death.

Danish National Patient Registry (DNPR): The DNPR, established in 1977, collects information on all non-psychiatric hospital admissions in Denmark. Data from outpatient and emergency department visits have been included since 1995. For each contact, the DNPR records dates of admission and discharge and up to 20 discharge diagnoses, assigned by physicians and coded according to the *International Classification of Diseases*, 8th revision (ICD-8) through 1993 and the 10th version (ICD-10) from 1994 onward [18]. We extracted data from the DNPR on fractures, comorbidity (including liver diseases), HIV-infection, alcohol abuse and drug abuse [15].

The Registry of Drug Abusers Undergoing Treatment has registered all individuals in Denmark assigned to treatment of drug addiction since 1996 [19]. Treatment of drug addiction in Denmark is restricted to referral centres, which must supply data to the registry in order to obtain funding.

Study population

We conducted two sub-studies to analyse the association between HCV-infection and risk of fractures.

- In *sub-study 1* the study groups were the *HCV cohort*, which included all patients > 16 years of age in the DANVIR cohort who after 1 January 1995 and after the age of 16 years tested positive for HCV (a positive test for HCV antibodies and/or HCV RNA), and the *comparison cohort* was extracted at random from CRS and consisted of 5 age- and sex-matched individuals who were alive and not registered with HCV diagnosis at study inclusion. For the HCV-exposed individual and the matched population controls study inclusion was the date of positive test for HCV in the index patient. Rates of fractures in the HCV and comparison cohorts were compared.
- *Sub-study 2* included all patients from the DANVIR cohort with a positive HCV antibody test and at least one test for HCV RNA after the positive HCV antibody test. Study inclusion was defined as the date of HCV RNA measurement. Rates of fractures were compared between HCV RNA-positive and HCV RNA-negative individuals. HCV RNA category was not changed to accord with the results of any subsequent testing [15, 20].

Information on HCV-exposed patients and the comparison cohort

The following information on the HCV-exposed patients and persons in the comparison cohort was ascertained as of the date of study inclusion: comorbidity (+/- liver diseases), alcohol abuse, intravenous drug use (IDU) and HIV.

Information on alcohol abuse, IDU and HIV coinfection was obtained from DNPR and the Registry of Drug Abusers Undergoing Treatment (see Appendix 1 for details). Comorbidity was included in the analyses as a modified Charlson Comorbidity Index (CCI) score derived from diagnoses registered in the Danish National Hospital Registry prior to the HCV diagnosis date [21, 22]. The CCI assigns a score between one and six to a range of diseases, with the sum of individual scores serving as a measure of patients' overall comorbidity. We identified comorbid diseases using the ICD-10 codes provided by Quan *et al.* [23] (matching ICD-8 codes to ICD-10 codes as closely as possible) (please see appendix 2 for details). Liver diseases were not included in the CCI in the present study. We defined three modified comorbidity levels based on the following CCI scores: none (score = 0), low (score = 1 - 2) and high (score ≥ 3).

Outcome

We had three outcomes: Time to fracture at any site, time to low-energy fractures and time to other fractures (please see appendix 1 for details). Low-energy fractures were defined as fractures possibly due to osteoporosis, typically those caused by low-energy trauma [6, 8].

Statistical analysis

Person-years at risk were computed from study inclusion until the first date of any fracture diagnosis, death, emigration or 1 January 2010, whichever came first. To assess the effects of HCV on the risk of fracture, we fitted cause-specific proportional hazards models for each of the three outcomes (any fracture, low-energy fracture and other fracture) and computed hazard ratios with

95% confidence intervals (CI) as an estimate of incidence rate ratios (IRRs). In accordance with the matched design stratified models were used for sub-study 1. Individuals who died were censored on the date of death [24]. For each outcome two main models were used: model 1 in which no adjustments were made and model 2 including adjustment for HIV, IDU, alcohol abuse and comorbidity. Further, we fitted two exploratory models, model 3 and 4 which were as model 2, but also adjusted for liver diseases (model 3) and previous fractures (model 4). In sub-study 2 models 2-4 were further adjusted for age and sex. The analyses were repeated in strata defined by sex, age (< 50 years and ≥ 50 years at study inclusion) and absence of liver disease, and for sub-study 2, also presence of liver disease.

To estimate absolute risks of fractures we used the cumulative incidence function. In the analyses of any fracture, death was handled as competing risks, in the analyses of low-energy fracture, other fracture and death were handled as competing risks and in the analyses of other fractures, low-energy fracture and death were handled as competing risks [25]. SPSS, version 19 (IBM Inc), and R software, version 2.14.2, were used for data analysis.

Ethics

Our study was approved by the Danish Data Protection Agency.

RESULTS

Descriptive data

Sub-study 1 included 12,013 HCV-exposed patients and 60,065 individuals from the comparison cohort with 72,356 and 454,115 person years of observation (PY) (Table 1). 64% were male and the median age at inclusion was 40 years (interquartile range: 31–48). HCV-exposed patients were more likely to have a diagnosis of alcohol abuse, IDU, HIV, other comorbidity and a previous

diagnosis of fracture than persons from the comparison cohort. Sub-study 2 included 7,156 HCV antibody-positive patients with 42,390 PY, of whom 4,500 (63%) were viremic and 2,656 (37%) were non-viremic at the time of the HCV RNA test. Viremic patients were more likely to be male and to have a diagnosis of IDU and a previous diagnosis of fracture than nonviremic patients (Table 1).

Risk of fracture in HCV-exposed patients versus the comparison cohort

HCV-exposed patients had an increased risk of fracture, low-energy fracture and other fractures compared to the comparison cohort (Figure 1). Unadjusted IRRs (model 1) were 2.83 (95% CI: 2.70-2.97) for all fractures, 2.96 (95% CI: 2.73–3.22) for low-energy fracture, and 2.77 (95% CI: 2.61–2.93) for other fracture. Adjusted IRRs (model 2) were 2.15 (95% CI: 2.03-2.28) for all fractures, 2.13 (95% CI: 1.93-2.35) for low-energy fracture, and 2.18 (95% CI: 2.02-2.34) for other fracture. The estimates obtained in model 3 and 4 did not diverge substantially from that of model 2 (appendix 3). As shown in table 2, stratified analyses revealed that fracture risk was equally increased in the examined subgroups. The 10-year risk of low-energy fracture and other fractures for HCV-infected patients were 9.0% (95% CI: 8.4%-9.7%) and 17.4% (95% CI: 16.6%-18.3%) whereas the corresponding figures for the comparison cohort were 4.2% (95% CI: 4.0% - 4.4%) and 7.9% (95% CI: 7.7% - 8.2%) (Figure 1).

Risk of fracture in viremic versus nonviremic patients

In unadjusted analyses, HCV RNA-positive patients had a slightly increased risk of fracture compared to HCV RNA-negative patients: unadjusted IRRs were 1.18 (95% CI: 1.07-1.32) for all fractures, 1.28 (95% CI: 1.06-1.53) for low-energy fracture, and 1.14 (95% CI: 1.00-1.30) for other fracture. In adjusted analyses fracture risk did not differ significantly between the two groups:

adjusted IRRs (model 2) were 1.08 (95% CI: 0.97-1.20) for all fractures, 1.20 (95% CI: 0.99-1.44) for low-energy fracture and 1.04 (95% CI: 0.91-1.19) for other fracture. The estimates obtained in model 3 and 4 did not diverge substantially from that of model 2 (appendix 3). In the adjusted analyses we found no statistically significant increased fracture risk in any of the examined subgroups when comparing chronic versus cleared infection (Table 3). The 10-year risk of low-energy fracture and other fractures for HCV RNA-positive patients were 9.5% (95% CI: 8.5%-10.6%) and 18.0% (95% CI: 16.6%-19.4%), whereas the corresponding figures for HCV RNA-negative patients were 7.7% (95% CI: 6.5% - 9.0%) and 16.6% (95% CI: 14.9% - 18.4%) (Figure 2).

DISCUSSION

In this large population-based, nationwide cohort study we observed a more than two fold increased risk of fracture in HCV-exposed patients compared to the general population and the risk was equally raised for low-energy and other fracture types. Furthermore the overall risk of fracture did not differ between patients with chronic versus cleared HCV-infection.

Both impaired bone strength and increased risk of traumas may contribute to the increased risk of fracture among HCV-exposed patients. The equally increased risk of all fracture types including non low-energy fractures suggests that increased risk of trauma contributes substantially to the increased fracture risk. HCV-infection is a marker of past or on-going intravenous drug use and risk-taking behaviour [26, 27], and alcohol overuse and HCV-infection often coexist [28]. Thus a number of factors may increase risk of falls, violence and traffic accidents in the HCV population [29].

Low BMD may be an effect of HCV-infection induced through inflammation or HCV induced liver disease [30]. Patients with cirrhosis, including HCV related cirrhosis, have a high prevalence of osteoporosis [1, 31]. In our study, adjusting for liver disease did not change the risk estimates substantially indicating that advanced liver disease is not a major driver of fracture risk in our populations. Low BMD may also be induced by risk factors associated with being exposed to HCV-infection including a lifestyle associated with malnutrition [32] and negative effects on bone metabolism associated with alcohol consumption [33-35]. Adjusting for HIV, IDU, alcohol abuse and comorbidity diminished the fracture risk associated with being HCV-seropositive which implies that part of fracture risk can be attributed to these risk factors.

The finding of an increased risk of fracture in HCV-exposed patients was extended by comparing risk of fracture between patients with chronic and cleared HCV-infection. Of interest, fracture risk did not differ significantly between patients with cleared versus chronic HCV-infection in adjusted

analyses. Accordingly, factors associated with HCV-infection rather than a direct effect of HCV-infection on bone health, seem to be main determinants of fracture risk. Albeit not a significant finding in adjusted analyses, viremic patients had approximately 20% higher risk of low-energy fracture than non-viremic patients, thus our study cannot discard that chronic HCV-infection per se also exerts negative effects on bone metabolism and thereby fracture risk, especially, when taking into account the rather few patients at the end of follow-up.

Recently, another large cohort study examined fracture risk in HCV-infected patients; this study found that HCV-infected persons had increased risk of hip fracture with a hazard ratio of 2.69 (adjusted hazard ratio of 1.47) compared with uninfected individuals [7]. The risk estimates are comparable with our results as we found a 2.96 fold increased risk of all low-energy fractures in HCV-infected patients (adjusted IRR of 2.13). Lo Re and co-authors only reported data for hip fractures, and they did not report result according to HCV RNA status. Only two small studies in selected groups of post-menopausal women have previously compared fracture rates in patients with chronic versus cleared HCV-infection. In contrast to our results, these studies found significantly increased fracture risk in HCV RNA-positive women compared with HCV RNA-negative women. The results may have been hampered by small number of endpoints [4] or may be confounded by shared risk factors for non-responding to interferon and being prone to fractures [36]. We did not specifically study postmenopausal women, but in stratified analyses, we found approximately the same relative risk estimates among persons older than 50 years or in women when comparing chronic versus cleared HCV-infection.

To our knowledge, this is the first large-scale cohort study designed to compare fracture risk in viremic versus nonviremic HCV-infected patients. The strengths of our study also include the population-based design with unique linkage to validated national registries, which allowed for long and complete follow-up. Our study also had limitations. We estimated fracture risk starting from

time of diagnosis of HCV-infection as we were not able to determine the date of infection. Regular testing for HCV RNA subsequent to an initial diagnosis is not performed systematically in Denmark. Therefore, we could not model HCV viremia as a time updated variable and our analyses did not account for spontaneous or treatment-related viral clearance nor reinfection during follow-up. However, spontaneous clearance of HCV-infection mainly happens shortly after the initial acute phase of the disease and occurs infrequently in later phases of the disease [37]. In addition, during the study period, only a minority of Danish HCV-patients received antiviral treatment [13]. We did not exclude patients with a diagnosis of fracture before study inclusion. Fracture data were incomplete before 1995 as the DNHR only included data on outpatient and emergency patient visits since 1995. The interpretation of our results should also take into considerations that fracture before the age of 50 years rarely reflects osteoporosis [6]. Another potential shortcoming is inaccuracies in fracture diagnoses reported to DNHR. However, the positive predictive value of the diagnoses registered in DNHR is generally high (70% to 99%) [38], and has for hip fractures been shown to be as high as 93% [39]. Another limitation is the lack of data on known risk factors for osteoporosis, such as menopause, medication and smoking. If these factors were distributed unequally between study groups, which is likely to be the case in sub study 1, but less likely in sub study 2, they might have affected the results.

We conclude that HCV-exposed patients have substantially increased risk of all fracture types, while clearance of HCV only leads to a minor risk reduction. Thus our study suggests that fracture risk is multi-factorial and indicate that a direct metabolic effect of HCV-infection on bone mineral density is not the major determinant of fracture risk among HCV-infected individuals. As the risk of low-energy and other fracture types were equally increased in the HCV-exposed population and the risk estimates were reduced when adjusting for alcohol and drug abuse, we suggest that lifestyle

related factors have a substantial impact on the increased risk of fracture in the HCV-infected population. The main preventive measures to decrease risk of fractures in the HCV population should therefore focus on modification of life-style related factors.

Acknowledgments

The authors thank Centers and members of the DANVIR Cohort Study (Appendix 4)

References

[1] Gonzalez-Calvin JL, Gallego-Rojo F, Fernandez-Perez R, Casado-Caballero F, Ruiz-Escolano E, Olivares EG. Osteoporosis, mineral metabolism, and serum soluble tumor necrosis factor receptor p55 in viral cirrhosis. J Clin Endocrinol Metab 2004;89:4325-4330.

[2] Lin JC, Hsieh TY, Wu CC, Chen PJ, Chueh TH, Chang WK, et al. Association between chronic hepatitis C virus infection and bone mineral density. Calcif Tissue Int 2012 ;91:423-429.

[3] Schiefke I, Fach A, Wiedmann M, Aretin AV, Schenker E, Borte G, et al. Reduced bone mineral density and altered bone turnover markers in patients with non-cirrhotic chronic hepatitis B or C infection. World J Gastroenterol 2005;11:1843-1847.

[4] Nanda KS, Ryan EJ, Murray BF, Brady JJ, McKenna MJ, Nolan N, et al. Effect of chronic hepatitis C virus infection on bone disease in postmenopausal women. Clin Gastroenterol Hepatol 2009;7:894-899.

[5] Pelazas-Gonzalez R, Gonzalez-Reimers E, eman-Valls MR, Santolaria-Fernandez F, Lopez-Prieto J, Gonzalez-Diaz A, et al. Bone alterations in hepatitis C virus infected patients. Eur J Intern Med 2013;24:92-96.

[6] Lippuner K, Golder M, Greiner R. Epidemiology and direct medical costs of osteoporotic fractures in men and women in Switzerland. Osteoporos Int 2005;16 Suppl 2:S8-S17.

[7] Lo RV, III, Volk J, Newcomb CW, Yang YX, Freeman CP, Hennessy S, et al. Risk of hip fracture associated with hepatitis C virus infection and hepatitis C/HIV Coinfection. Hepatology 2012;56:1688-98.

[8] Hansen AB, Gerstoft J, Kronborg G, Larsen CS, Pedersen C, Pedersen G, et al. Incidence of low and high-energy fractures in persons with and without HIV infection: a Danish population-based cohort study. AIDS 2012;26:285-293.

[9] Yin MT, Shi Q, Hoover DR, Anastos K, Sharma A, Young M, et al. Fracture incidence in HIV-infected women: results from the Women's Interagency HIV Study. AIDS 2010;24:2679-2686.

[10] Young B, Dao CN, Buchacz K, Baker R, Brooks JT. Increased rates of bone fracture among HIV-infected persons in the HIV Outpatient Study (HOPS) compared with the US general population, 2000-2006. Clin Infect Dis 2011;52:1061-1068.

[11] Statistics Denmark. Population and election. Population and population forecasts. Population at the first day of the quarter by municipality, sex, age, marital status, ancestry, country of origin and citizenship (FOLK1). Available from: http://www.statistikbanken.dk. Accessed November 10, 2013

[12] Christensen PB, Hay G, Jepsen P, Omland LH, Just SA, Krarup HB, et al. Hepatitis C prevalence in Denmark -an estimate based on multiple national registers. BMC Infect Dis 2012;12:178.

[13] Lettmeier B, Muhlberger N, Schwarzer R, Sroczynski G, Wright D, Zeuzem S, et al. Market uptake of new antiviral drugs for the treatment of hepatitis C. J Hepatol 2008;49:528-536.

[14] Pedersen CB, Gotzsche H, Moller JO, Mortensen PB. The Danish Civil Registration System. A cohort of eight million persons. Dan Med Bull 2006;53:441-449.

[15] Omland LH, Krarup H, Jepsen P, Georgsen J, Harritshoj LH, Riisom K, et al. Mortality in patients with chronic and cleared hepatitis C viral infection: a nationwide cohort study. J Hepatol 2010;53:36-42.

[16] Omland LH, Jepsen P, Krarup H, Schonning K, Lind B, Kromann-Andersen H, et al. Increased mortality among persons infected with hepatitis C virus. Clin Gastroenterol Hepatol 2011;9:71-78.

[17] Krarup HB, Drewes AM, Madsen PH. A quantitative HCV-PCR test for routine diagnostics. Scand J Clin Lab Invest 1998;58:415-422.

[18] Andersen TF, Madsen M, Jørgensen J, Mellemkjær L, Olsen JH. The Danish Hospital Register: a valuable source of data for modern health sciences. Dan Med Bull 1999;46:263-268.

[19] Statens Serum Institut. The Registry of Drug Abusers Undergoing Treatment (Register over stofmisbrugere i behandling). Available from: http://www.ssi.dk/Sundhedsdataogit/Registre/Register%20over%20stofmisbrugere%20i%20behandling.aspx. Accessed October 16, 2013. Danish.

[20] Omland LH, Jepsen P, Weis N, Christensen PB, Laursen AL, Nielsen H, et al. Mortality in HIV-infected injection drug users with active vs cleared hepatitis C virus-infection: a population-based cohort study. J Viral Hepat 2010;17:261-268.

[21] Charlson ME, Pompei P, Ales KL, MacKenzie CR. A new method of classifying prognostic comorbidity in longitudinal studies: development and validation. J Chronic Dis 1987;40:373-383.

[22] Sundararajan V, Henderson T, Perry C, Muggivan A, Quan H, Ghali WA. New ICD-10 version of the Charlson comorbidity index predicted in-hospital mortality. J Clin Epidemiol 2004;57:1288-1294.

[23] Quan H, Sundararajan V, Halfon P, Fong A, Burnand B, Luthi JC, et al. Coding algorithms for defining comorbidities in ICD-9-CM and ICD-10 administrative data. Med Care 2005;43:1130-1139.

[24] Lau B, Cole SR, Gange SJ. Competing risk regression models for epidemiologic data. Am J Epidemiol 2009;170:244-256.

[25] Pintille M. Competing Risks: A Practical Perspective, 1st ed John Wiley & Sons Ltd.; 2006.

[26] Hansen AB, Gerstoft J, Kronborg G, Pedersen C, Sorensen HT, Obel N. Mortality in Siblings of Patients Coinfected with HIV and Hepatitis C Virus. J Infect Dis 2007;195:230-235.

[27] Hansen AB, Lohse N, Gerstoft J, Kronborg G, Laursen A, Pedersen C, et al. Cause-specific excess mortality in siblings of patients co-infected with HIV and hepatitis C virus. PLoS One 2007;2:e738.

[28] Mueller S, Millonig G, Seitz HK. Alcoholic liver disease and hepatitis C: a frequently underestimated combination. World J Gastroenterol 2009;15:3462-3471.

[29] Oppenheim WL. The "battered alcoholic syndrome". J Trauma 1977;17:850-856.

[30] Rouillard S, Lane NE. Hepatic osteodystrophy. Hepatology 2001;33:301-307.

[31] Gallego-Rojo FJ, Gonzalez-Calvin JL, Munoz-Torres M, Mundi JL, Fernandez-Perez R, Rodrigo-Moreno D. Bone mineral density, serum insulin-like growth factor I, and bone turnover markers in viral cirrhosis. Hepatology 1998;28:695-699.

[32] Santolaria F, Gonzalez-Reimers E, Perez-Manzano JL, Milena A, Gomez-Rodriguez MA, Gonzalez-Diaz A, et al. Osteopenia assessed by body composition analysis is related to malnutrition in alcoholic patients. Alcohol 2000;22:147-157.

[33] Diamond T, Stiel D, Lunzer M, Wilkinson M, Posen S. Ethanol reduces bone formation and may cause osteoporosis. Am J Med 1989;86:282-288.

[34] Farley JR, Fitzsimmons R, Taylor AK, Jorch UM, Lau KH. Direct effects of ethanol on bone resorption and formation in vitro. Arch Biochem Biophys 1985;238:305-314.

[35] Spencer H, Rubio N, Rubio E, Indreika M, Seitam A. Chronic alcoholism. Frequently overlooked cause of osteoporosis in men. Am J Med 1986;80:393-397.

[36] Arase Y, Suzuki F, Suzuki Y, Akuta N, Kobayashi M, Sezaki H, et al. Virus clearance reduces bone fracture in postmenopausal women with osteoporosis and chronic liver disease caused by hepatitis C virus. J Med Virol 2010;82:390-395.

[37] Lauer GM, Walker BD. Hepatitis C virus infection. N Engl J Med 2001;345:41-52.

[38] Nickelsen TN. [Data validity and coverage in the Danish National Health Registry. A literature review]. Ugeskr Laeger 2001;164:33-37.

[39] Hoidrup S, Gronbaek M, Gottschau A, Lauritzen JB, Schroll M. Alcohol intake, beverage preference, and risk of hip fracture in men and women. Copenhagen Centre for Prospective Population Studies. Am J Epidemiol 1999;149:993-1001.

Table 1. Characteristics of the study populations

	Sub-study 1		Sub-study 2	
	HCV-exposed patients	Comparison cohort	HCV RNA + patients	HCV RNA - patients
Number	12,013	60,065	4500	2656
Person years of observation	72,356	454,115	25,991	16,399
Outcome				
Censured	7,169	51,935	2,728	1,789
Death	2,133	2,042	766	335
Other fracture	1,774	4,001	659	362
Low-energy fracture	937	2,087	347	170
Male	7,687 (64%)	38,435 (64%)	3,085 (69%)	1,481 (56%)
Age, years (IQR*)	39.55 (31.38-47.89)	39.55 (31.38-47.88)	39.85 (32.01-47.42)	37.60 (29.78-47.07)
Modified CCI**				
0	9,499 (79%)	54,270 (90%)	3,509 (78%)	2,138 (80%)
1	1,826 (15%)	5,002 (9%)	698 (16%)	364 (14%)
2	688 (6%)	793 (1%)	293 (7%)	154 (6%)
Previous fracture	4,282 (36%)	9,868 (16%)	1,843 (41%)	879 (33%)
HIV	286 (2%)	31 (0%)	177 (4%)	81 (3%)
Drug	3,225 (27%)	242 (0%)	1,489 (35%)	716 (27%)
Alcohol	1,451 (12%)	660 (1%)	684 (15%)	297 (11%)

Note: *IQR: interquartile range, CCI: Charlson Comorbidity Index

Table 2. Risk of fracture in HCV-exposed patients compared to the comparison cohort.

		Model 1	Model 2
	Fracture type	IRR (95% CI)	IRR (95% CI)
All	All	2.83 (2,70-2,97)	2.15 (2.03-2.28)
	Low-energy	2.96 (2.73-3.22)	2.13 (1.93-2.35)
	Other	2.77 (2.61-2.93)	2.18 (2.02-2.34)
Subgroups			
< 50 years	All	3.08 (2.92-3.25)	2.40 (2.24-2.56)
	Low-energy	3.40 (3.08-3.74)	2.51 (2.22-2.83)
	Other	2.96 (2.78-3.15)	2.36 (2.18-2.55)
≥ 50 years	All	1.89 (1.68-2.13)	1.42 (1.24-1.63)
	Low-energy	2.11 (1.81-2.48)	1.56 (1.31-1.87)
	Other	1.64 (1.36-1.97)	1.26 (1.02-1.56)
Women	All	2.43 (2.23-2.65)	1.84 (1.65-2.05)
	Low-energy	2.49 (2.18-2.85)	1.86 (1.59-2.18)
	Other	2.39 (2.13-2.68)	1.83 (1.58-2.11)
Men	All	3.03 (2.86-3.21)	2.32 (2.16-2.48)
	Low-energy	3.30 (2.97-3.67)	2.34 (2.06-2.66)
	Other	2.92 (2.72-3.13)	2.32 (2.13-2.52)
No liver disease	All	2.76 (2.62-2.90)	2.12 (2.00-2.26)
	Low-energy	2.90 (2.65-3.16)	2.11 (1.90-2.34)
	Other	2.69 (2.53-2.86)	2.14 (1.98-2.30)

Model 1: Unadjusted IRRs.

Model 2: IRRs adjusted for HIV, IDU, alcohol abuse and modified Charlson Comorbidity Index, age and sex.

Table 3. Risk of fracture in patients with chronic HCV-infection compared to patients with cleared HCV-infection.

		Model 1	Model 2
	Fracture type	IRR (95% CI)	IRR (95% CI)
All	All	1.18 (1.07-1.32)	1.08 (0.97-1.20)
	Low-energy	1.28 (1.06-1.53)	1.20 (0.99-1.44)
	Other	1.14 (1.00-1.30)	1.04 (0.91-1.19)
Subgroups			
< 50 years	All	1.17 (1.04-1.31)	1.07 (0.95-1.20)
	Low-energy	1.38 (1.11-1.72)	1.23 (0.99-1.53)
	Other	1.09 (0.96-1.25)	1.01 (0.88-1.16)
≥ 50 years	All	1.26 (0.95-1.67)	1.18 (0.88-1.59)
	Low-energy	1.08 (0.76-1.54)	1.12 (0.77-1.61)
	Other	1.63 (1.02-2.60)	1.29 (0.79-2.11)
Women	All	1.14 (0.95-1.37)	1.06 (0.88-1.28)
	Low-energy	1.26 (0.95-1.67)	1.19 (0.90-1.58)
	Other	1.05 (0.82-1.34)	1.00 (0.78-1.28)
Men	All	1.14 (1.00-1.30)	1.09 (0.95-1.24)
	Low-energy	1.31 (1.02-1.67)	1.20 (0.94-1.54)
	Other	1.08 (0.93-1.26)	1.05 (0.90-1.23)
No liver disease	All	1.19 (1.07-1.34)	1.08 (0.97-1.21)
	Low-energy	1.25 (1.03-1.52)	1.20 (0.98-1.47)

	Other	1.17 (1.02-1.34)	1.05 (0.92-1.21)
With liver disease	All	1.02 (0.74- 1.41)	0.99 (0.71-1.37)
	Low-energy	1.29 (0.77- 2.15)	1.11 (0.66-1.87)
	Other	0.87 (0.58- 1.32)	0.91 (0.60-1.39)

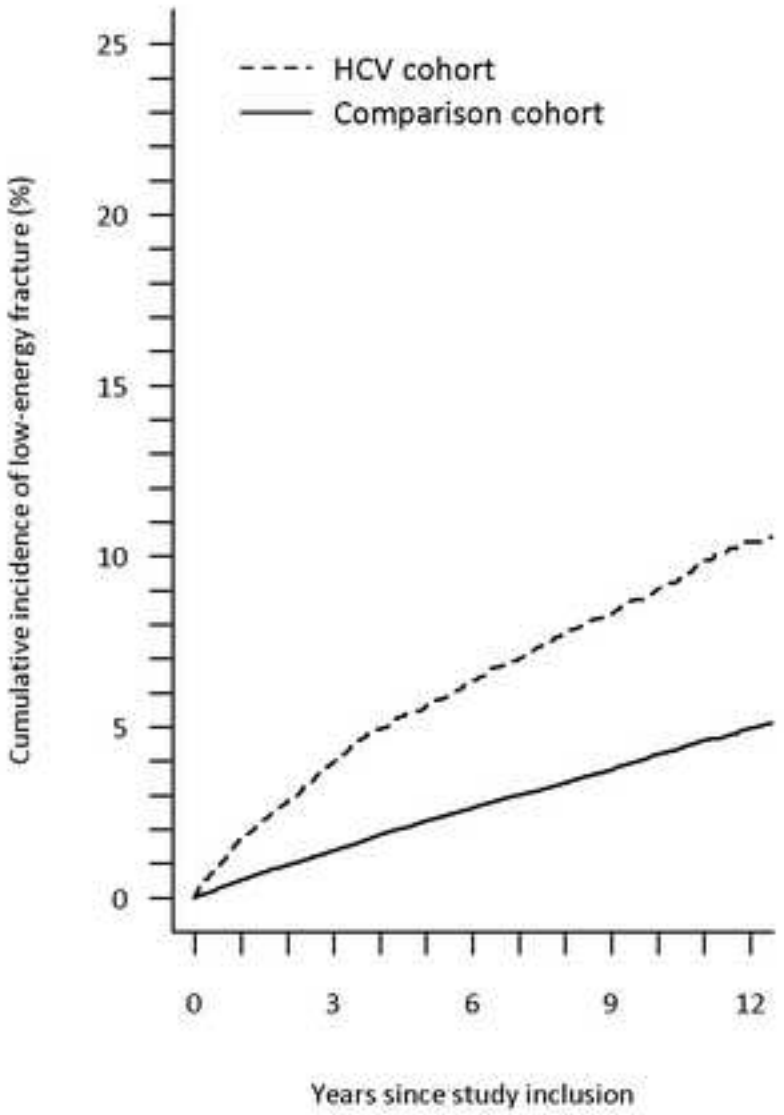
Model 1: Unadjusted IRRs.

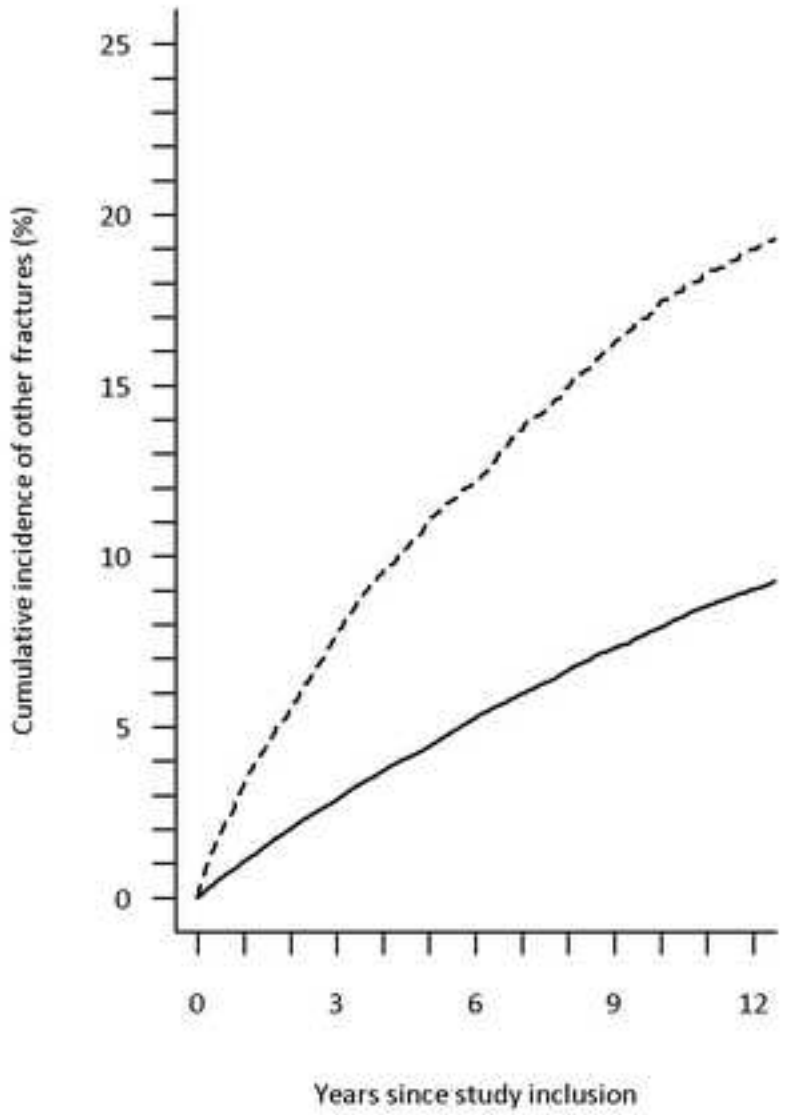
Model 2: IRRs adjusted for HIV, IDU, alcohol abuse and modified Charlson Comorbidity Index, age and sex.

FIGURE LEGENDS.

Fig. 1. Risk of fractures in the HCV cohort (dotted line) and the comparison cohort (full line). The left panel illustrates the risk of low-energy fracture and the right panel illustrates the risk of other fractures.

Fig. 2. Risk of fractures in HCV RNA-positive-patients (dotted line) and HCV RNA-negative patients (full line). The left panel illustrates the risk of low-energy and the right panel illustrates the risk of other fractures.

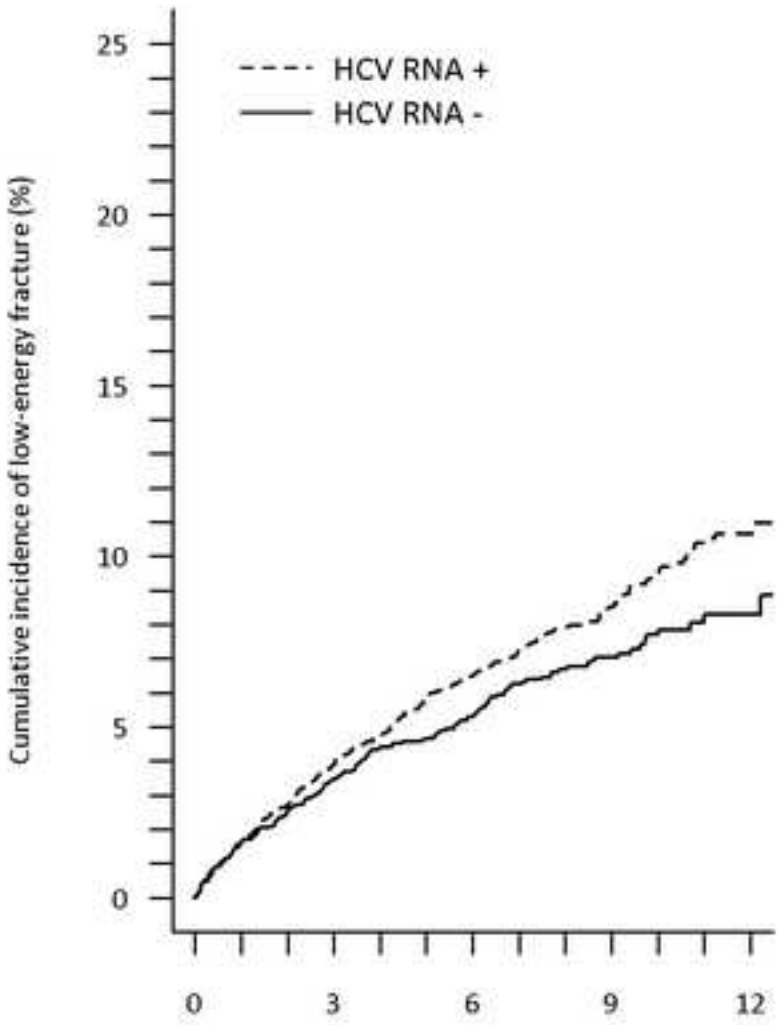


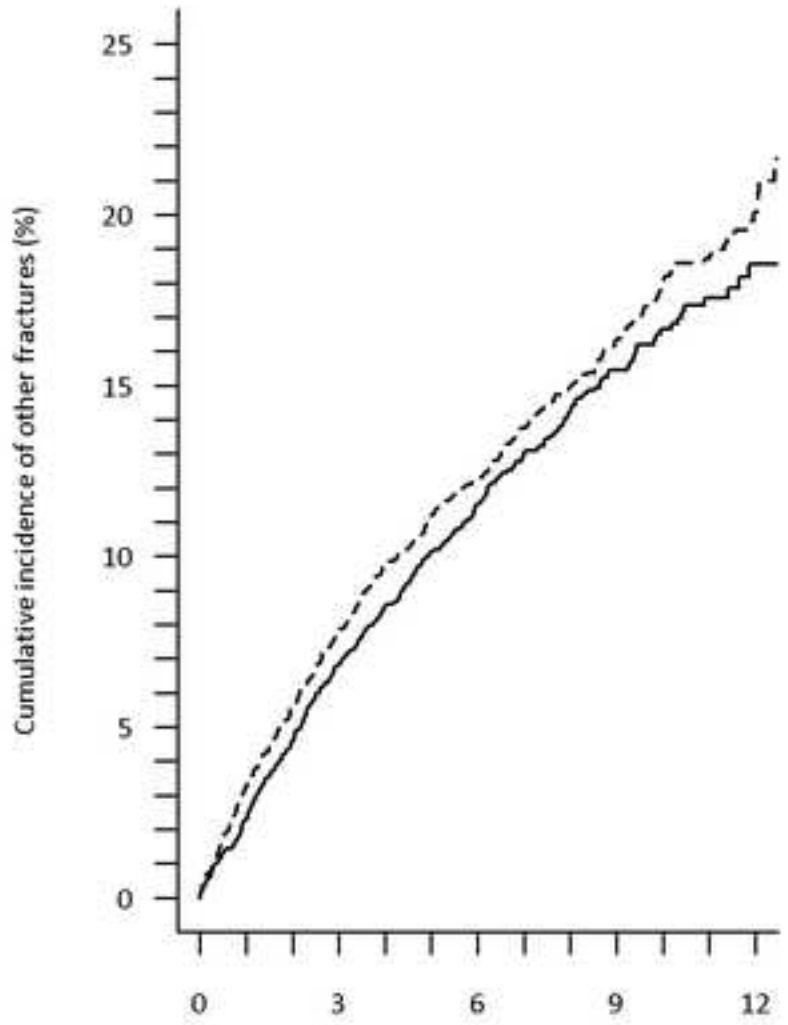


No. at risk

HCV cohort	12013	9295	5622	2521	849
Comparison cohort	60065	55770	38102	19093	7540

	12013	9295	5622	2521	849
	60065	55770	38102	19093	7540





No. at risk					
HCV RNA +	4500	3506	2032	830	160
HCV RNA -	2656	2166	1311	591	130

4500	3506	2032	830	160
2656	2166	1311	591	130