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Chronic Liver Disease in Homeless Individuals and Performance of Non-Invasive Liver Fibrosis and Injury Markers: VALID Study

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List of abbreviations
AUD alcohol use disorder
BBV blood borne virus
BMI body mass index
ccCK caspase-cleaved cytokeratin
CLD chronic liver disease
CSHF clinically significant hepatic fibrosis
CI confidence intervals
ELF Enhanced Liver fibrosis
HCV hepatitis C virus
HA Hyaluronic acid
IDU injecting drug use
ITT intention to treat
IFN-γ interferon-gamma
IL interleukin
LSM liver stiffness measurements
MMP-2 Matrix metalloproteinase-2
ODN Operator Delivery Network
PWAH people who are homeless
PWID people who inject drugs
P111NP Procollagen III amino terminal peptide
SVR sustained virological response
T-helper Th
TIMP-1 Tissue inhibitor of metalloproteinase-1
TE transient elastography
TNF-α TNF tumour necrosis factor alpha

Conflict of interest
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Abstract

**Background/aims:** Community-based assessment and management of chronic liver disease (CLD) in people who are homeless (PWAH) remains poorly described. We aimed to determine prevalence/predictors of chronic liver disease (CLD) in PWAH and assess performance of non-invasive hepatocyte fibrosis and injury markers.

**Methods:** The Vulnerable Adult Liver Disease (VALID) study provided a “one-stop” liver service based at homeless hostels. Our primary outcome was the prevalence of clinically significant hepatic fibrosis (CSHF) (liver stiffness measurement (LSM) > 8kPa).

**Results:** Total individuals recruited were 127, mean±SD age 47±9.4 years, 50% (95% CI 41%-59%) and 39% (95% CI 31%- 48%) having alcohol dependence and a positive HCV RNA respectively. CSHF was detected in 26% (95% CI 17%-35%), independent predictors being total alcohol unit/week (OR 1.01, 95% CI 1.00-1.02, p=0.002) and HCV RNA positivity (OR 2.93, 95% CI 1.12-7.66, p=0.029). There was moderate agreement between LSM and Enhanced Liver Fibrosis (ELF) score (kappa 0.536, p<0.001) for CSHF as assessed by LSM ≥8kPa. Those with CSHF had significantly higher levels of IFN-γ (p=0.002), IL-6 (p=0.001), MMP-2 (p=0.006), ccCK-18 (p<0.001) and ELF biomarkers (p<0.001), compared to those without CSHF. Service uptake was >95%. Direct acting antiviral (DAA) treatment completion was 93% (95% CI 77%-99%), sustained virological response (SVR) being 83% (95% CI 64%-94%).

**Conclusion:** There is a significant liver disease burden from HCV and alcohol in PWAH. Non-invasive hepatocyte fibrosis and injury markers can help in identifying such individuals in the community. Despite a challenging cohort, excellent service uptake and high DAA-based SVRs can be achieved.

Key words: homeless people, community health services, fibrosis, hepatitis C, cytokines
Lay Summary

1. The extent of liver disease amongst homeless individuals remains unclear.
2. We describe a novel service based at homeless hostels using a painless liver scan (fibroscan).
3. Based on the fibroscan, about one in five homeless individuals had liver scarring. This was due to high prevalence of both alcohol use and hepatitis C. Despite being vulnerable, service uptake and hepatitis C cure rates were high.
4. We found that special blood tests (ELF, cytokines) could also help identify homeless individuals with liver scarring in the community.
5. Future community-based studies need to address how best to improve liver health of homeless individuals.
Introduction

In England, homelessness and rough sleeping have increased 42% and 134% respectively in the last 7 years (1), consistent with the trends in Europe (2). In 2018, 1:200 individuals in England were estimated to be homeless (3). Though hepatitis C virus (HCV) infection disproportionately affects people who are homeless (PWAH) (4-8), a significant proportion still remains undiagnosed and consequently untreated (9). The advent of direct-acting antivirals (DAAs) (10) provides an unprecedented opportunity to treat individuals in the community, an important step in achieving HCV elimination by 2030 (11). There is also high prevalence of alcohol use, both in PWAH (12) and in those with HCV infection (13-14).

Studies from the UK and USA show that substance/alcohol use and liver disease account for about 20% - 40% of all deaths amongst PWAH in England and Wales (15-18).

Despite the potential for significant liver disease in PWAH, prevalence and predictors of chronic liver disease (CLD) remains largely uncharacterised in this disenfranchised cohort. A recent systematic review indicates that community-based holistic models of care that include point of care testing can facilitate CLD management amongst the homeless (4). Transient elastography, a quick, non-invasive scan for hepatic fibrosis assessment has good reproducibility (19), is increasingly being used in community settings (4) and can also enhance engagement (20). Other non-invasive fibrosis markers such as the Enhanced Liver Fibrosis (ELF) and AST:Platelet Ratio Index (APRI) score shows good performance and considerable diagnostic value for predicting histological fibrosis stage across a spectrum of liver diseases (21-23). However, they have largely been assessed in secondary care settings (21-23).

To further characterise the CLD burden amongst PWAH, we set up the Vulnerable Adult Liver Disease (VALID) study, a “one-stop” comprehensive liver service based at two
homeless hostels in southeast England. Our primary objectives were to determine prevalence and predictors of chronic liver disease (CLD) in PWAH. Our secondary objectives included assessing performance of non-invasive hepatic fibrosis (TE, ELF score and APRI) and injury markers in identifying CLD in a community setting. In addition to TE, we assessed ELF and APRI score as there is limited data in vulnerable adults in a community setting. We studied the following non-invasive hepatocyte injury markers as they are the key players driving hepatic inflammation and fibrosis: tumour necrosis factor alpha (TNF-α), interferon gamma (IFN-γ), T-helper (Th) 17 cytokines, serum caspase cleaved cytokeratin-18 (ccCK-18) and matrix metalloproteinase 2 (MMP-2) (24-32). As these markers have largely been assessed in secondary care (24-32), we aimed to assess their performance in a vulnerable cohort in the community.

Methods

The VALID study was a three-year prospective cohort study that commenced in Sept 2015. It was based at two homeless hostels and their affiliated primary care practices in southeast England. The process of service set up was similar to our earlier ITTREAT model (8, 33). Inclusion criteria were consecutive adults attending the two homeless hostels and or the affiliated homeless primary care practice and willing and able to give informed consent. We required that the individuals be registered at either of the two primary care practices. Initially, the service was offered to individuals aged ≥ 50 years (as per funder remit), but after negotiations with the funder/obtaining additional funding, this was amended to include those aged ≥ 18 years. Although those unwilling to give informed consent were offered the service, their data were not collected.

Outcomes
Our primary outcome was prevalence and predictors of CSHF as assessed by LSM \( \geq 8 \text{kPa} \).

Our secondary outcomes were

- comparing performance of non-invasive fibrosis markers (TE, ELF score and APRI) in identifying CLD in the community (as assessed by LSM \( \geq 8 \text{kPa} \))
- assessing performance of non-invasive hepatocyte injury markers in identifying CLD in the community (as assessed by LSM \( \geq 8 \text{kPa} \))
- service uptake (BBV screening, fibroscan, HCV treatment), prevalence of HCV, IDU, alcohol dependence and cirrhosis and HCV treatment outcomes (intention to treat ITT).

**Services offered**

We provided a “one-stop” service via “drop-in” clinics two to three times a week that were run by AH under SV supervision. After receiving informed consent, each participant was offered the following: routine clinical bloods including blood borne virus (BBV) screening, assessment of alcohol use (AUDIT questionnaire and alcohol breath test analysis - AlcoDigital Life Guard breathalyser); substance misuse (self-reported); and hepatic fibrosis (transient elastography (TE) - FibroScan®402 Echosens). Blood was also collected for hepatocyte fibrosis and injury markers on the same day as TE (see below).

All individuals with a positive HCV PCR and/or clinically significant hepatic fibrosis (CSHF) as assessed by TE (liver stiffness measurements (LSM) \( \geq 8 \text{kPa} \)) were recalled to offer HCV treatment and or liver health promotion. A LSM \( \geq 8 \text{kPa} \) has previously been shown to predict hepatic fibrosis during community screening with about 50% having F2 fibrosis, the remainder having F1 fibrosis (34). The primary care physicians were informed of any cases with alcohol dependence and/or going injecting drug use (IDU) so that appropriate onwards referrals to addiction centres could be arranged.
In England, HCV treatment is provided via 22 regional centres (Operational Delivery Networks - ODNs). DAA regimen is determined and funded by the National Health Service England (NHSE), after approval at a weekly regional ODN meeting. All individuals with a positive HCV RNA were deemed treatment eligible (irrespective of ongoing drug/alcohol use), as long as they were motivated and willing to engage. DAAs were delivered to the homeless hostels and HCV treatment monitored by AH in the community under SV supervision. Supplementary Fig 1 shows the participant pathway.

**Data collection and analysis**

The following anonymised clinical data were prospectively collected onto the study database: demographics, alcohol use, body mass index (BMI), waist/hip ratio and mid-arm circumference, micronutrients (serum calcium, magnesium and phosphate), service uptake, LSM, and HCV treatment outcomes. For biomarker assays, blood samples were transported to the laboratory for processing and storage at -80°C within 4 hours. Serum was obtained following centrifugation (1500g, 15 min) of clotted blood collected in serum separator tubes (BD Ltd). Biomarkers assessed were:

- Ten key cytokines, including markers of the Th17 pathway (Interferon gamma (IFN-γ), Interleukin (IL)-1β, IL-6, IL-10, IL-17A, IL-17E/IL-25, IL-17F, IL-21, IL-22 and tumour necrosis factor alpha (TNF-α)) were measured using the U-PLEX Th17 Combo 2 (human) kit (Mesoscale Discovery), following the manufacturer instructions. Only cytokines with 33% of the cohort having detectable levels were included in the analysis. Undetectable cytokine values were assigned half the lower detection limit of the assay. The detection limit of each assay was 2 pg/ml for IL-17A, 0.5 pg/ml for IL-
Cytokine assays were performed by the NIHR Cambridge BRC Core Biochemical Assay Laboratory.

- Senescence biomarkers: caspase cleave cytokeratin 18 (ccCK18), which was determined in serum using the M30-Apoptosense ELISA kits, (Peviva, Sweden) and tissue inhibitor of metalloproteinase-1 (TIMP-1) and Matrix metalloproteinase-2 (MMP-2) which were quantified by Luminex using a Human Premixed multi-analyte kit (R&D systems). Data were acquired on a validated and calibrated Bio-Plex 200 system (Bio-Rad) and analysed with Bio-Plex Manager 6.0 software with detection target of 50 beads per region and standard curve fitted using five parameter logistic regression. Senescence biomarkers were analyzed at Nottingham Biomedical Research Centre.

- Enhanced Liver Fibrosis (ELF) biomarkers (Hyaluronic acid (HA), Procollagen III amino terminal peptide (PIIINP) and TIMP-1 were determined in serum samples by iQur Ltd (London, UK). The results from the three assays were entered into the manufacturer's algorithm to derive an ELF score.

Further details of how research blood samples were collected, processed, stored and analysed are provided in supplementary file 1. Researchers were blinded to clinical information and all samples were assayed in duplicate and freeze-thaw cycles were limited to two for all measurements.

**Study definitions**

Currently homeless: street homeless or in temporary accommodation at initial assessment
Currently in stable accommodation: in stable accommodation at initial assessment but still attending services at homeless hostels; prior history of homelessness.

Elderly: aged ≥ 50 years.

Current smoking, injecting, non-injecting drug and alcohol use: smoking, drug and alcohol use at initial assessment.

Alcohol dependence: AUDIT questionnaire score ≥ 20.

Stability for HCV treatment: willingness and motivation to engage and be adherent with HCV treatment. This was assessed by AH with input from SV if needed.

Sustained virological response (SVR12): absence of detectable virus (at any level) 12 weeks after end of treatment (EOT).

Successful fibroscan: >10 successful readings, ≥60% success rate, and an interquartile range (IQR) to median ratio of ≤0.30 (as per manufacturer recommendations Echosens, Paris, France).

Clinically significant hepatic fibrosis (CSHF) and cirrhosis: liver stiffness measurement ≥ 8 kPa and ≥13kPa (35-36); AST:Platelet Ratio Index (APRI) score: 1.5-2 and > 2 (23) and ELF score: ≥ 9.8 and ≥10.51 (21-22)

Intention to treat (ITT): includes all individuals who commenced HCV treatment

**Sample size**

Based on our prior work (8), we estimated a) 35% of PWAH will have CSHF (LSM ≥ 8 kPa); b) 40% HCV seroprevalence. The number of patients we aimed to recruit was 300. Of this we expected 100 patients would be aged ≥50 years allowing us to construct a 95% confidence interval (CI) width ±10% (CI 40% to 60%) around an estimated prevalence of CSHF of 50% in this group. We aimed to recruit 200 non-elderly patients allowing us to calculate a 95% CI width ±6% (24% to 37%) around a prevalence of CSHF of 30% in this group. A total of n=300 would allow us to construct a 95% CI of approximate width ±5% (CI 32% to 43%)
around the combined prevalence of CSHF of 37% in the 2 groups. A prevalence of 111/300 would suffice for fitting a logistic regression model with 11 independent variables.

**Statistical analysis**

Data are summarised using counts, means ± standard deviations (for normally distributed variables), medians (interquartile ranges (IQR) for skewed variables, or frequencies and percentages (for categorical variables). Mann-Whitney U and chi-square tests were utilised for continuous and categorical variables respectively. Logistic regression analysis was used to model the relationship between the binary dependent outcomes (Yes vs. No) (HCV RNA positive vs negative, CSHF vs. no CSHF, cirrhosis vs. no cirrhosis) and key independent factors. A multifactorial logistic regression model was then derived to look at the relationship between the key factors and the dependent outcome. To build the model, the statistically significant key factors (p<0.1) from the unifactorial analyses were added to the null model using forward selection where the factor with the highest significant p-value, based on the likelihood ratio test, was added next. Factors were removed from the model if p >0.05. Data were analysed using Stata v16 (Texas, USA) (37) and SPSS v26 (Armonk, NY, USA: IBM Corp). We also assessed correlation (Spearmans correlation) between LSM and APRI/ELF scores for CSHF and cirrhosis.

Ethical approval for the study was obtained from NRES Committee South Central - Hampshire B (REC ref 15/SC/0112), all participants signing an informed consent.

**Results**

During the study period we recruited 127 individuals. Fig 1a shows how the study cohort was selected. Of a total of 1875 vulnerable adults registered at the two primary care practices, 131 (7%) attended the drop in clinics of whom 127 (97%) were recruited. Therefore, we achieved
42% of our target sample size. Of the 131 individuals that attended the “drop-in” clinics, 125 (95%) underwent BBV screening and 127/131 (97%) accepted community-based fibroscan. Of those offered HCV treatment (n=29), all accepted.

Table 1 shows the baseline demographic and clinical data. This was a predominantly Caucasian male cohort, 76% being currently homeless. Comparing currently homeless vs. those currently in stable accommodation (supplementary Table 1), the former were more likely to be males, 78/96 (81%) vs. 19/31 (61%), p=0.023) and be younger (45.7 ± 9.4 vs. 53.5 ± 6.6, p<0.001).

**Hepatic fibrosis data**

All 127 individuals underwent a successful non-fasting fibroscan examination (in two this was at second attempt). Prevalence of CSHF (LSM ≥8kPa) and cirrhosis (LSM ≥13kPa) were 26% (95% CI 17%-35%) and 17% (95% CI 11%-24%) respectively (Table 1). The mean ELF score among participants (n=101) was 9.1±1.4. Twenty-eight (28%, 95% CI 19%-38%) had CSHF (ELF ≥9.8), while 14 (14%, 95% CI 8%-22%) had cirrhosis (ELF > 10.51) (Table 1). Of those with CSHF (LSM ≥ 8) (n=33), six (18%) were HCV RNA positive, n=13 (39%) had alcohol dependence (AUDIT score≥20), n=11 (33%) had both a positive HCV RNA and alcohol dependence and three (9%) had neither risk factor. Supplementary Table 2 shows the demographic and clinical factors associated with CSHF (LSM ≥8kPa). There were no statistically significant differences in prevalence of CSHF in those currently homeless vs. those currently in stable accommodation (26/96 (27%) vs. 7/31 (23%), p=0.619). Of those aged ≥ 50 years, 16/60 (27%) had CSHF (LSM ≥8kPa) vs. 17/67 (25%) aged < 50 years (p=0.868) (supplementary table 1). Table 2 shows the unifactorial and multifactorial regression analysis of predictors of CSHF (LSM ≥8kPa). A variable can be included either as
a continuous or categorical variable in a regression analysis. Since a continuous variable has greater value and provides a more statistically powerful analysis, AUDIT score rather than AUDIT score ≥ 20 was entered into the logistic regression. Independent predictors of CSHF were total alcohol unit/week (OR 1.01, 95% CI 1.00-1.02, p=0.002) and being HCV RNA positive (OR 2.93, 95% CI 1.12-7.66, p=0.029) (Table 2). Fig 1b shows area under the curve (AUC) analysis for weekly alcohol units in predicting CSHF (LSM ≥8kPa). With a cut-off of 41 units or more/week, sensitivity and specificity for predicting CSHF were 81% & 76% respectively.

Supplementary Table 3 shows the baseline demographic and clinical factors associated with cirrhosis (LSM ≥13kPa). There were no statistically significant differences in prevalence of cirrhosis in those currently homeless vs. those currently in stable accommodation (16/96 (17%) vs. 5/31 (16%), p=0.944). Prevalence of cirrhosis (LSM ≥13kPa) in those aged ≥ 50 years vs. <50 years was 10/60 (17%) vs. 11/67 (16%), p=0.97 (supplementary Table 3).

Supplementary Table 4 shows the unifactorial and multifactorial regression analysis of predictors of cirrhosis respectively. Independent predictors of cirrhosis (LSM ≥13kPa) were alcohol units/week (OR 1.014, 95% CI 1.009-1.020, p<0.001). Of the 21 individuals with cirrhosis (LSM ≥13kPa), 10 (48%) had alcohol dependence, nine (43%) had both alcohol dependence and a positive HCV RNA and two (9%) had neither risk factor.

**Correlation between liver stiffness measurements (LSM) and ELF and APRI scores**

There was moderate correlation between LSM and ELF score (Spearman correlation 0.553, p<0.001 (Fig 1c). There was moderate agreement between LSM (≥8kPa) and ELF score (≥ 9.8) for CSHF (kappa 0.536, p<0.001), an ELF score ≥ 9.8 correctly identifying 19/29 (65%) with CSHF (LSM ≥8kPa) (supplementary Table 5). There was also good agreement between
LSM ($\geq 13$kPa) and ELF score ($\geq 10.51$) for cirrhosis (kappa 0.734, p<0.001), an ELF score $\geq 10.51$ correctly identifying 12/17 (71%) with cirrhosis (LSM $\geq 13$kPa) (supplementary Table 6). The correlation between LSM and APRI score was also moderate (Spearman correlation 0.558, p<0.001 (Fig 1d). In comparison to ELF, APRI had lower degree of agreement with LSM for both CSHF (kappa 0.452, p<0.001) and cirrhosis (kappa 0.510, p<0.001). An APRI score between 1.5-2 correctly identifying 13/28 (46%) with CSHF (LSM $\geq 8$kPa) (supplementary Table 7) with an APRI score $> 2$ correctly identifying 10/18 (56%) with cirrhosis (LSM $\geq 13$kPa) (supplementary Table 8).

Non-invasive hepatocyte injury and fibrosis markers

The number of individuals who had cytokine panel, MMP-2, CK-18, and ELF biomarkers analysed were 97, 79, 99 and 101 respectively. Of the 97 patients with cytokine data available, IL-10 was detectable in 42 (43%), IL-17A in 37 (38%), IL-22 in 52 (54%) and IL-6 76 (78%). Only two patients had undetectable levels of IFN-γ, while all had detectable TNF-α levels. Serum ccCK18, MMP-2 and ELF biomarkers were detectable in all patients included in the analysis.

Non-invasive hepatocyte injury and fibrosis markers in those with a positive versus negative HCV RNA

Those with a positive HCV RNA had significantly higher median (IQR) levels of Th17 cytokines (IL-10, IL-22), TNF-α, ccCK18 and ELF biomarkers (PIINP, TIMP-1) (Table 3). Seventy-six percent with a positive HCV RNA had detectable IL-10 levels vs. 22% with negative HCV RNA (p<0.001), there being no statistically significant differences in prevalence of IL-6, IL-17A and IL-22 in those with a positive vs. negative HCV RNA (supplementary Table 9).
Non-invasive hepatocyte injury and fibrosis markers in those with clinically significant hepatic fibrosis (LSM >8kPa) vs. no clinically significant hepatic fibrosis (LSM <8kPa)

Table 4 shows median (IQR) levels of hepatocyte injury and fibrosis markers in those with and without CSHF. The former had significantly higher levels of IFN-γ, IL-6, MMP2, ccCK18 and ELF biomarkers (Table 4). Of those with CSHF, 60% had detectable IL-10 vs. 36% without CSHF (p=0.027), and 93% with CSHF had detectable IL-6 vs 73% without CSHF (p=0.027). There were no statistically significant differences in the prevalence of detectable IL-17A or IL-22 in those with and without CSHF (supplementary Table 10).

Non-invasive hepatocyte injury and fibrosis markers in those with cirrhosis (LSM ≥13kPa) versus no cirrhosis (LSM <13kPa)

Those with cirrhosis had significantly higher levels of IFN-γ, IL-6, MMP2, ccCK-18 and ELF biomarker) (supplementary Table 11). There were no statistically significant differences in the prevalence of detectable cytokines in those with and without cirrhosis (supplementary Table 12).

Hepatitis C virus, injecting drug use (IDU) and alcohol dependence prevalence

HCV seroprevalence was 47% (95% CI 38%-56%), 39% (95% CI 31%-48%) being HCV PCR positive (table 1). There was high prevalence of current IDU (28%, 95% CI 21%-37%), alcohol dependence (AUDIT score ≥ 20) (50%, 95% CI 41%-59%) with 47% (95% CI 38%-56%) being on treatment for mental health conditions (Table 1). Comparing currently homeless vs. those currently in stable accommodation, the former were more likely to be current IDU, 36/96 (38%) vs. 0/32 (0%), p<0.001; current non-IDU, 51/96 (53%) vs. 8/31 (26%), p=0.008; current smokers, 87/96 (91%) vs. 17/31 (55%), p< 0.001; be HCV RNA
positive, 44/96 (46%) vs. 5/31 (16%), p=0.002 and have a detectable breathalyzer test at initial assessment, 28/88 (32%) vs. 1/22 (5%), p=0.009 (supplementary Table 1).

Supplementary Table 13 shows demographic and clinical data in those with and without a positive HCV RNA with supplementary Table 14 showing unifactorial and multifactorial regression analysis of predictors of a positive HCV RNA. Independent predictors of a positive HCV RNA were the AUDIT questionnaire score (OR 0.94, 95% CI 0.90-0.98, p=0.001); current IDU (OR 3.33, 95% CI 1.07-10.39, p=0.038); current non-IDU (OR 4.05, 95% CI 1.49-11.01, p=0.006); and LSM ≥ 8kPa (OR 6.80, 95% CI 2.04-22.72, p=0.002) (supplementary Table 14).

**HCV treatment data and outcomes**

Table 5 shows DAA-based treatment outcomes in 29 individuals, of whom four (14%) had cirrhosis, with 12 (41%) and 16 (55%) having current IDU and alcohol use respectively (i.e. use at initial assessment). ITT SVR rates were 24/29 (83%, 95% CI 64%-94%), treatment completion being 27/29 (93%, 95% CI 77%-99%).

**Discussion**

The VALID study provided a novel service based at homeless hostels. We found a high burden from CLD in PWAH, with just over 25% having CSHF (as assessed by LSM ≥8kPa), both alcohol dependence and HCV RNA positivity being independent predictors. We observed moderate agreement between LSM and ELF score for CSHF in a community setting. Compared to those without CSHF, those with CSHF had significantly higher levels of IFN-γ, IL-6, IL-10, MMP-2, CK-18 and ELF biomarkers, another novel finding. Contrary to widely held perception, our vulnerable homeless population were highly engaged with services offered, with 95% accepting the BBV screening, 97% undergoing fibroscan and all
accepting DAA therapy with excellent SVR rates. A recent systematic review (which includes the current study), has addressed community-based liver care for PWAH (4). Our model is however unique as it was an integrated service (BBV screening, IDU/alcohol assessment, HCV treatment) and the only one to assess the performance of multiple non-invasive liver fibrosis and injury markers in a homeless population. This study is therefore a valuable addition to the current literature.

HCV prevalence is higher amongst those with alcohol excess and the homeless compared with the general population (4, 38) and vice versa, a recent systematic review identified about 1 in 5 with HCV having alcohol use disorder (AUD) (39). We observed a three-fold higher HCV seroprevalence in those currently homeless vs. those currently in stable accommodation, confirming that PWAH remain the most vulnerable of people who inject drugs (PWIDS) (40-41). Though studies show that the highest absolute rate of disease in homeless individuals is for hepatitis C (7, 42-43), it is imperative that alcohol use be concurrently addressed. Alcohol acts synergistically with HCV resulting in a more than three-fold higher risk for liver disease progression (39), maybe by enhancing HCV replication (44). In our study, alcohol use was an independent predictor of both CSHF and cirrhosis as assessed by LSM, consistent with a large American study involving homeless individuals (45). In fact, in about 75% and 90% of our cohort with CSHF and cirrhosis respectively, alcohol, either alone or in conjunction with HCV was the underlying aetiology for CLD. Additionally, ongoing alcohol can negate the beneficial effects of successful DAA therapy with an almost six times higher liver-related morbidity compared to the general population (46). A recent systematic review concluded that integration of HCV and substance use services might improve engagement along the continuum of HCV care amongst PWIDs (47). Despite being one of the most marginalised populations in accessing healthcare, high
SVR rates can also be achieved in PWAH if delivered as part of a holistic service (4, 8, 48), also corroborated by the current study.

While TE is increasingly being used in community settings (4), there is limited data on other non-invasive tests. An American study assessed the FIB4 test in PWAH and reported a 16% prevalence of advanced fibrosis (45) consistent with our study. We were unable to identify any other study specifically assessing ELF or APRI in homeless adults. In a non-community setting, TE might be superior to performance of APRI (49). In our study, we observed a higher agreement between LSM and ELF for CSHF as compared to agreement between LSM and APRI. Another recent community-based study also reported that ELF score more accurately identified individuals with advanced fibrosis due to alcohol-related liver disease than the APRI and FIB4 test (50). These data provide preliminary evidence supporting the use of the ELF score as a non-invasive hepatic fibrosis marker amongst vulnerable adults in the community, though this need confirmation in larger prospective studies.

TNF-α and IFN-γ are pro-inflammatory cytokines produced primarily as part of the T-helper 1 (Th1) response and are known to mediate cytotoxicity and liver injury in the early stages of viral hepatitis (24-25). Interleukin (IL)-6, another pro-inflammatory cytokine, is secreted in the acute phase response to infection and tissue injury and is implicated in the process of hepatic regeneration (26-27). Similarly, IL-17 and IL-22 are important Th17 cytokines, IL-22 tending to ameliorate and IL-17 worsening liver fibrosis (28-29). ccCK-18 and MMP-2 are two hepatic senescence markers associated with hepatic inflammation and fibrosis (30-32). An earlier study has shown suppressed immune responses in the homeless population, which may explain their increased susceptibility to infections (51). We were however unable to find any study assessing hepatocyte injury markers in PWAH. Our preliminary data shows that
IFN-γ, IL-6, IL-10, MMP-2 and ccCK-18 could help identify vulnerable adults with CSHF in a community setting (as assessed by LSM). While levels of Th17 cytokines IL-10 and IL-22 were significantly elevated in patients with HCV, this was not correlated with fibrosis progression. This has also been reported earlier by Chang et al (52) in an Asian cohort with chronic HCV infection. Our results indicate that even amongst vulnerable adults in the community, the role of Th17 cytokines in the pathogenesis of hepatic fibrosis, particularly in HCV infection, are not well established. While we accept that these data are not directly related to service delivery, adding these tests to our existing repertoire, could help identify and clarify the mechanistic paradigm for CLD in vulnerable adults and merits further research.

This study did have limitations. We recruited 42% of our target sample size resulting in less precise prevalence estimates. Margin of error for 26% CSHF prevalence was 8% with our achieved sample size of n=127, whereas it would have been 5.6% with the original sample size of n=300 and expected 37% CSHF prevalence. It also resulted in the opportunity to include fewer variables in our multivariable model for CSHF (approximately 3 as opposed to 11 independent variables). Contributory factors for the slow recruitment were the initial age restrictions, and that participants had to be registered with the affiliated primary care practices as a safety net. As the majority of HCV treatment in our region is now community-based, registration with primary care is on longer necessary. Though we recruited < 10% of the eligible cohort, it must be remembered that PWAH are a transient population. However, of those attending the “drop-in” clinics, almost all (97%) consented to participate. Additional study limitation were absence of liver biopsies, so we lacked a “gold-standard” comparator. Earlier studies nonetheless have indicated that a LSM ≥8kPa can predict CLD in the community (32). We also accept that the high prevalence of alcohol use and the non-fasting
fibroscans could have reduced the accuracy of TE. Finally, due to the small sample size we did not stratify liver fibrosis by underlying disease aetiology.

In conclusion, the VALID study demonstrates the high prevalence of both HCV and alcohol-related liver disease in PWAH, the latter often ignored due to emphasis on HCV elimination. We also provide preliminary data on identifying homeless individuals with CLD in the community using non-invasive liver fibrosis and injury marker. Future studies need to focus on developing community-based services to improve liver health of PWAH including novel test to identify CLD in a community setting.

References


22. Non-alcoholic fatty liver disease (NAFLD): assessment and management. NICE guideline [NG49] Published date: 06 July 2016 https://www.nice.org.uk/guidance/ng49/chapter/recommendations#assessment-for-nafld. Accessed 05.05.21


37. StataCorp. 2019. Stata Statistical Software: Release 16. College Station, TX: StataCorp LLC.


Table 1 Baseline demographic and clinical data in study cohort (n=127)

<table>
<thead>
<tr>
<th>Variable</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yrs)</td>
<td>47±9.4</td>
</tr>
<tr>
<td>Age ≥50</td>
<td>60 (47%)</td>
</tr>
<tr>
<td>Ethnicity</td>
<td>Caucasian 122 (96%)</td>
</tr>
<tr>
<td>Males</td>
<td>97 (76%)</td>
</tr>
<tr>
<td>Recruitment Site</td>
<td></td>
</tr>
<tr>
<td>Homeless hostel 1</td>
<td>88 (69%)</td>
</tr>
<tr>
<td>Primary care practice 1</td>
<td>13 (10%)</td>
</tr>
<tr>
<td>Homeless hostel 2</td>
<td>3 (2.4%)</td>
</tr>
<tr>
<td>Primary care practice 2</td>
<td>23 (18%)</td>
</tr>
<tr>
<td><strong>Currently homeless</strong></td>
<td></td>
</tr>
<tr>
<td>Living in hostel</td>
<td>79 (62%)</td>
</tr>
<tr>
<td>Supported/temporary accommodation</td>
<td>15 (12%)</td>
</tr>
<tr>
<td>Street homeless</td>
<td>2 (2%)</td>
</tr>
<tr>
<td><strong>Currently in stable accommodation</strong></td>
<td></td>
</tr>
<tr>
<td>Alcohol units/week</td>
<td>40 (98)</td>
</tr>
<tr>
<td>Alcohol &gt; weekly recommended (&gt;14 units)</td>
<td>83 (65%)</td>
</tr>
<tr>
<td>Breathalyzer reading (µg/dL) (n=110)</td>
<td>0.01±0.025</td>
</tr>
<tr>
<td>Detectable Alcohol on Breathalyzer (n=110)</td>
<td>29 (26%)</td>
</tr>
<tr>
<td>AUDIT questionnaire score (0-40)</td>
<td>17±13.8</td>
</tr>
<tr>
<td>AUDIT questionnaire ≥ 20 (alcohol dependence)</td>
<td>63 (50%)</td>
</tr>
<tr>
<td>Major comorbidities*</td>
<td>23 (18%)</td>
</tr>
<tr>
<td>Smoking</td>
<td></td>
</tr>
<tr>
<td>Current</td>
<td>104 (82%)</td>
</tr>
<tr>
<td>Ex-Smoker</td>
<td>13 (10%)</td>
</tr>
<tr>
<td>Never</td>
<td>10 (8%)</td>
</tr>
<tr>
<td>Injecting drug use (IDU)</td>
<td></td>
</tr>
<tr>
<td>Current</td>
<td>36 (28%)</td>
</tr>
<tr>
<td>Daily</td>
<td>15 (12%)</td>
</tr>
<tr>
<td>Weekly</td>
<td>15 (12%)</td>
</tr>
<tr>
<td>Less than weekly</td>
<td>5 (4%)</td>
</tr>
<tr>
<td>Missing</td>
<td>1 (1%)</td>
</tr>
<tr>
<td>Past</td>
<td>32 (25%)</td>
</tr>
<tr>
<td>Never</td>
<td>59 (47%)</td>
</tr>
<tr>
<td>Non IDU</td>
<td></td>
</tr>
<tr>
<td>Current</td>
<td>59 (47%)</td>
</tr>
<tr>
<td>Past</td>
<td>29 (23%)</td>
</tr>
<tr>
<td>Never</td>
<td>39 (31%)</td>
</tr>
<tr>
<td>Ever had mental health diagnosis</td>
<td>92 (72%)</td>
</tr>
<tr>
<td>On treatment for mental health conditions</td>
<td>59 (47%)</td>
</tr>
<tr>
<td>Sexual Orientation</td>
<td></td>
</tr>
<tr>
<td>Heterosexual</td>
<td>115 (91%)</td>
</tr>
<tr>
<td>Homosexual</td>
<td>4 (3%)</td>
</tr>
<tr>
<td>Bisexual</td>
<td>7 (6%)</td>
</tr>
<tr>
<td>Transgender</td>
<td>1 (1%)</td>
</tr>
<tr>
<td>Bilirubin (µmol/L)</td>
<td>9±8</td>
</tr>
<tr>
<td>ALT (iu/L) (n=106)</td>
<td>28.5 (38)</td>
</tr>
<tr>
<td>AST (iu/L)</td>
<td>30 (42)</td>
</tr>
<tr>
<td>Albumin (g/L)</td>
<td>45±4</td>
</tr>
<tr>
<td>Platelet count (10^9/L)</td>
<td>245±97</td>
</tr>
<tr>
<td>INR (n=114)</td>
<td>1±0.1</td>
</tr>
<tr>
<td>Magnesium (n=98)</td>
<td>0.86±0.08</td>
</tr>
<tr>
<td>Calcium (n=101)</td>
<td>2.24±0.08</td>
</tr>
<tr>
<td>Phosphate (n=99)</td>
<td>1.07±0.19</td>
</tr>
<tr>
<td>Any micronutrient deficiency (n=101)</td>
<td>17 (17%)</td>
</tr>
</tbody>
</table>
### Hepatic fibrosis

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Median LSM (kPa) (n=127)</th>
<th>Median LSM ≥ 8kPa (CSHF)</th>
<th>Median LSM ≥ 13kPa (cirrhosis)</th>
<th>ELF score (n=101)</th>
<th>ELF &gt; 9.8</th>
<th>ELF &gt; 10.51</th>
<th>APRI score (n =106)</th>
<th>APRI between 1.5-2</th>
<th>APRI &gt;2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>5.4 (4.3-8.0)</td>
<td>33 (26%)</td>
<td>21 (17%)</td>
<td>9.1 +1.4</td>
<td>28 (28%)</td>
<td>14 (14%)</td>
<td>0.4 (0.25-0.97)</td>
<td>18 (17%)</td>
<td>16 (15%)</td>
</tr>
</tbody>
</table>

### Anthropometric measurements

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMI (kg/m²)</td>
<td>24.3 ±4.2</td>
</tr>
<tr>
<td>Mid-Arm circumference (cms)</td>
<td>29.42 ±4</td>
</tr>
<tr>
<td>Waist:Hip ratio</td>
<td>0.93 ±0.08</td>
</tr>
</tbody>
</table>

### Blood borne virus screening (n=125, except HBcAb (n=124))

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>HIV antibody positive</td>
<td>3 (2%)</td>
</tr>
<tr>
<td>HbsAg positive</td>
<td>0 (0%)</td>
</tr>
<tr>
<td>HbcAb positive</td>
<td>21 (17%)</td>
</tr>
<tr>
<td>HCV antibody positive</td>
<td>59 (47%)</td>
</tr>
<tr>
<td>HCV RNA positive</td>
<td>49 (39%)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>21 (43%)</td>
</tr>
<tr>
<td>1b</td>
<td>1 (2%)</td>
</tr>
<tr>
<td>3</td>
<td>24 (49%)</td>
</tr>
<tr>
<td>2b</td>
<td>1 (2%)</td>
</tr>
</tbody>
</table>

| Could not be determined | 2 (4%) |

Continuous data shown as median (IQR) or mean ± SD and categorical data as percentage (%)

Brackets () indicate number with data available

LSM liver stiffness measurement; CSHF clinically significant hepatic fibrosis

Normal values: bilirubin 0-21 µmol/L, ALT 0-41 iu/L, AST 0-40 iu/L, albumin 35-52g/L, INR 0.8-1.2, platelets 150-450x10⁹/L, magnesium 0.66-1.07 mmol/L, calcium 2.15-2.5 mmol/L, phosphate 0.81-1.45 mmol/L

*Major comorbidities were defined as the presence of significant chronic physical illnesses other than liver disease including chronic lung/cardiac disease, cerebrovascular disease, active malignancy, chronic kidney disease and complicated diabetes mellitus. Controlled hypertension, HIV and mental illnesses were excluded
Table 2 Unifactorial and multifactorial regression analysis of baseline demographic and clinical variables predicting clinically significant hepatic fibrosis (LSM >8 kPa)

<table>
<thead>
<tr>
<th>CSHF</th>
<th>Unifactorial analysis</th>
<th>Multifactorial analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Odds ratio</td>
<td>95% CI</td>
</tr>
<tr>
<td>Age ≥ 50</td>
<td>1.07</td>
<td>0.48 – 2.37</td>
</tr>
<tr>
<td>Male Gender</td>
<td>2.03</td>
<td>0.71 – 5.83</td>
</tr>
<tr>
<td>Alcohol AUDIT questionnaire score*</td>
<td>1.04</td>
<td>1.01 – 1.07</td>
</tr>
<tr>
<td>Total alcohol units/week*</td>
<td>1.01</td>
<td>1.005 – 1.014</td>
</tr>
<tr>
<td>Current injecting drug use (IDU)</td>
<td>0.47</td>
<td>0.18 – 1.27</td>
</tr>
<tr>
<td>Current non-IDU</td>
<td>0.95</td>
<td>0.43 – 2.10</td>
</tr>
<tr>
<td>Current Smoker</td>
<td>0.76</td>
<td>0.28 – 2.06</td>
</tr>
<tr>
<td>Currently homeless</td>
<td>1.27</td>
<td>0.49 – 3.31</td>
</tr>
<tr>
<td>Major Comorbidities</td>
<td>1.69</td>
<td>0.64 – 4.44</td>
</tr>
<tr>
<td>Mental Health</td>
<td>1.26</td>
<td>0.51 – 3.14</td>
</tr>
<tr>
<td>Micronutrient deficiency</td>
<td>2.24</td>
<td>0.75 – 6.65</td>
</tr>
<tr>
<td>HCV RNA positive</td>
<td><strong>1.99</strong></td>
<td><strong>1.05 – 5.20</strong></td>
</tr>
</tbody>
</table>

* Indicate continuous variable, the remainder been categorical variables
Table 3 Median (IQR) levels of biomarkers in those with and without a positive HCV RNA

<table>
<thead>
<tr>
<th>Cytokine/Biomarker</th>
<th>HCV RNA positive n=38</th>
<th>HCV RNA negative n=59</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Th17 cytokines</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-10 (pg/ml)</td>
<td>0.7 (1.2)</td>
<td>0.25 (0.0)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>IL-17A (pg/ml)</td>
<td>2 (1.5)</td>
<td>1 (1.4)</td>
<td>0.171</td>
</tr>
<tr>
<td>IL-22 (pg/ml)</td>
<td>0.5 (0.45)</td>
<td>0.6 (0.75)</td>
<td>0.026</td>
</tr>
<tr>
<td>Other cytokines</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IFN-γ (pg/ml)</td>
<td>13.3 (13.2)</td>
<td>11.3 (7.4)</td>
<td>0.176</td>
</tr>
<tr>
<td>TNF-α (pg/ml)</td>
<td>3.7 (2.2)</td>
<td>2.7 (0.8)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>1.5 (1.9)</td>
<td>1.4 (2.1)</td>
<td>0.911</td>
</tr>
<tr>
<td>Senescence biomarkers</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MMP-2 (ng/ml)</td>
<td>229.2 (109.9)</td>
<td>217.7 (123.7)</td>
<td>0.489</td>
</tr>
<tr>
<td>ccCK18 (u/L)</td>
<td>182.8 (388.6)</td>
<td>25 (86.8)</td>
<td>0.005</td>
</tr>
<tr>
<td>ELF biomarkers</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HA (ng/ml)</td>
<td>55.6 (64.8)</td>
<td>27.3 (35.0)</td>
<td>0.103</td>
</tr>
<tr>
<td>PIINP (ng/ml)</td>
<td>8.7 (10.3)</td>
<td>6.6 (4.3)</td>
<td>0.001</td>
</tr>
<tr>
<td>TIMP-1 (ng/ml)</td>
<td>215 (98.8)</td>
<td>171.8 (71)</td>
<td>0.001</td>
</tr>
</tbody>
</table>

IL interleukin, IFN-γ interferon-gamma, TNF-α tumour necrosis factor alpha, MMP-2 Matrix metalloproteinase-2, ccCK caspase-cleaved cytokeratin, HA Hyaluronic acid, PIINP Procollagen III amino terminal peptide, TIMP-1 Tissue inhibitor of metalloproteinase-1
Table 4 Median (IQR) levels of biomarkers in those with (LSM >8kPa) and without (LSM <8kPa) clinically significant hepatic fibrosis (CSHF)

<table>
<thead>
<tr>
<th>Cytokine/Biomarkers</th>
<th>CSHF (LSM &gt;8kPa)</th>
<th>No CSHF (LSM &lt;8kPa)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Th17 Cytokines</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IL-10 (pg/ml)</td>
<td>0.6 (0.73)</td>
<td>0.25 (0.45)</td>
<td>0.049</td>
</tr>
<tr>
<td>IL-17A (pg/ml)</td>
<td>1.5 (1.78)</td>
<td>1 (1.2)</td>
<td>0.106</td>
</tr>
<tr>
<td>IL-22 (pg/ml)</td>
<td>0.65 (0.65)</td>
<td>0.5 (0.45)</td>
<td>0.169</td>
</tr>
<tr>
<td><strong>Other cytokines</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IFN-γ (pg/ml)</td>
<td>16 (10.7)</td>
<td>10.8 (6.4)</td>
<td>0.002</td>
</tr>
<tr>
<td>TNF-α (pg/ml)</td>
<td>3.2 (1)</td>
<td>2.7 (1.15)</td>
<td>0.05</td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>2 (1.48)</td>
<td>1.3 (1.60)</td>
<td>0.001</td>
</tr>
<tr>
<td><strong>Senescence biomarkers</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MMP-2 (ng/ml)</td>
<td>259.8 (171.3)</td>
<td>205.7 (107.6)</td>
<td>0.006</td>
</tr>
<tr>
<td>ccCK18 (U/L)</td>
<td>347.7 (552.2)</td>
<td>25 (47.1)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td><strong>ELF biomarkers</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HA (ng/ml)</td>
<td>76.5 (82.5)</td>
<td>25.8 (35.9)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>PIIINP (ng/ml)</td>
<td>10.2 (11.6)</td>
<td>6.6 (3.2)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>TIMP-1 (ng/ml)</td>
<td>271 (154.3)</td>
<td>168.3 (57)</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

IL, interleukin, IFN-γ interferon-gamma, TNF-α tumour necrosis factor alpha, MMP2 Matrix metalloproteinase-2, ccCK caspase-cleaved cytokeratin, HA Hyaluronic acid, PIIINP Procollagen III amino terminal peptide, TIMP-1 Tissue inhibitor of metalloproteinase-1
Table 5  HCV treatment data and outcomes (n=29)

<table>
<thead>
<tr>
<th>No</th>
<th>LSM kPa</th>
<th>HCV Genotype</th>
<th>DAA regimen/duration (weeks)</th>
<th>Completed treatment</th>
<th>SVR</th>
<th>IDU at initial assessment (current IDU)</th>
<th>Alcohol use/AUDIT scores at initial assessment (current alcohol use)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>26.3</td>
<td>1a</td>
<td>SOF/LDV + RBV 12 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>Yes (22)</td>
</tr>
<tr>
<td>2</td>
<td>12.1</td>
<td>1a</td>
<td>OBV/PTVr/DSV + RBV 24 weeks</td>
<td>No only 20/24 weeks</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>Yes (23)</td>
</tr>
<tr>
<td>3</td>
<td>48.5</td>
<td>1a</td>
<td>OBV/PTVr/DSV+RBV + RBV 24 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>Yes (21)</td>
</tr>
<tr>
<td>4</td>
<td>4.9</td>
<td>3a</td>
<td>SOF/VEL 12 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>Yes (0)</td>
</tr>
<tr>
<td>5</td>
<td>3.8</td>
<td>1a</td>
<td>OBV/PTVr/DSV+RBV 12 weeks then SOF/LDV 12 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>Yes (2)</td>
</tr>
<tr>
<td>6</td>
<td>11.8</td>
<td>1a</td>
<td>EBR/GZR+ RBV 16 weeks</td>
<td>No only 8/16 weeks</td>
<td>No</td>
<td>Yes</td>
<td>Yes (14)</td>
</tr>
<tr>
<td>7</td>
<td>34.3</td>
<td>3a</td>
<td>SOF/VEL 12 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes (35)</td>
</tr>
<tr>
<td>8</td>
<td>7.9</td>
<td>3a</td>
<td>SOF/VEL 12 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>No (ex IDU)</td>
<td>No (0)</td>
</tr>
<tr>
<td>9</td>
<td>38.5</td>
<td>3a</td>
<td>GLE/PIB 12 weeks</td>
<td>No (ex IDU)</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes (40)</td>
</tr>
<tr>
<td>10</td>
<td>5.4</td>
<td>1a</td>
<td>OBV/PTVr/DSV 12 weeks then SOF/LDV 12 weeks</td>
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<td>Yes</td>
<td>No (ex IDU)</td>
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<td>11</td>
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<td>12</td>
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<td>EBR/GZR + RBV 12 weeks</td>
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<td>Yes</td>
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<td>No</td>
<td>Yes</td>
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<td>14</td>
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<td>3a</td>
<td>GLE/PIB 8 weeks</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
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<td>15</td>
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<td>EBR/GZR+RBV 16 weeks</td>
<td>Yes</td>
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<td>No (ex IDU)</td>
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<td>Yes</td>
<td>Yes</td>
<td>Yes (24)</td>
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<td>1a</td>
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LSM liver stiffness measurement, IDU injecting drug use, SVR sustained virological response, SOF/LDV sofosbuvir/ledispavir, OBV/PTVr/DSV Ombitasvir/Paritaprevir/Dasabuvir, SOF/VEL sofosbuvir/velpatasvir, EBR/GZR elbasvir/grazoprevir, GLE/PIB Glecaprevir/pibrentasvir, SOF/VEL/VOX sofosbuvir/velpatasvir/voxilaprevir * Treated as part of the STOP HCV1 study
Fig 1a Flow chart showing how the study cohort were selected

Site 1 n=1401

Site 2 n=474

Total eligible n=1875

Attend VALID clinic n=131

Total number recruited=127

Number excluded n = 4
  n=3 declined BBV testing
  n=1 was not registered at primary care practice.
Fig 1b Area Under the Curve for alcohol units/week in predicting clinically significant hepatic fibrosis (LSM ≥8kPa)

<table>
<thead>
<tr>
<th>Test Result Variable(s)</th>
<th>Area</th>
<th>Std. Error</th>
<th>Asymptotic Sig.</th>
<th>Lower Bound</th>
<th>Upper Bound</th>
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<tr>
<td></td>
<td>.699</td>
<td>.059</td>
<td>.001</td>
<td>.583</td>
<td>.814</td>
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Fig 1c Scatter plot showing correlation between Log values of LSM in kPa and ELF score (Spearman correlation 0.553, p value <0.001) (n=101)
Fig 1d Scatter plot showing correlation between Log values of LSM in kPa and APRI score (Spearman correlation 0.588, p value <0.001) (n=106)