

Hepatitis C virus serological and molecular status and circulating Th1-, Th2- and regulatory cytokines among young adults in Etsako, Edo State, Nigeria: a community-based cross-sectional study

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Abstract

Background

Hepatitis C virus (HCV) infection frequently persists without overt clinical manifestations and can progress silently to chronic liver injury. Anti-HCV immunoglobulin G (IgG) identifies serological exposure but cannot on its own distinguish active from resolved infection, whereas HCV RNA indicates detectable viral genetic material at the time of testing. This study determined anti-HCV IgG and HCV RNA positivity, and examined circulating interferon-gamma (IFN- γ), interleukin-5 (IL-5) and interleukin-10 (IL-10), among young adults in Etsako, Edo State, Nigeria.

Methods

This community-based cross-sectional study enrolled 200 adults aged 18–35 years (100 women, 100 men) recruited through a multistage household approach between January and June 2023. Venous blood was assayed for anti-HCV IgG, HCV RNA and the three cytokines. Categorical variables were summarised as frequencies and percentages with 95% confidence intervals; cytokine concentrations were summarised as mean $\pm$ standard deviation and compared with Welch's unequal-variance t-test, with two-sided $P < 0.05$ taken as significant.

Results

Anti-HCV IgG was detected in 17 of 200 participants (8.5%; 95% CI 5.4–13.2), compared with HCV RNA in 7 participants (3.5%; 95% CI 1.7–7.0). Paired testing classified 7 participants as antibody-positive/RNA-positive, 10 as antibody-positive/RNA-negative, none as antibody-negative/RNA-positive, and 183 as negative for both markers. Mean IFN- γ , IL-5 and IL-10 concentrations were numerically higher in RNA-positive than in anti-HCV IgG-positive participants, with the highest values in the antibody-positive/RNA-positive subgroup, but none of the between-group differences reached statistical significance. The three cytokines correlated positively with one another, and both HCV markers showed weak positive associations with each cytokine. No sociodemographic subgroup comparison reached statistical significance.

Conclusions

Antibody reactivity was more common than detectable viraemia in this population, underscoring the value of molecular confirmation after a reactive serological screen. The cytokine pattern is consistent with coordinated systemic immune activity accompanying HCV exposure, but the small RNA-positive subgroup limits the precision of these comparisons. Larger, longitudinal studies incorporating quantitative viral load and cellular immune measures are needed to clarify these relationships.

Background

Hepatitis C virus is a blood-borne pathogen capable of causing acute infection with few or no symptoms, which can progress over years to chronic hepatitis, hepatic fibrosis, cirrhosis and hepatocellular carcinoma. Its public-health importance has grown alongside the availability of highly effective direct-acting antivirals, which cure most treated individuals; timely diagnosis, confirmation of current infection and linkage to treatment are therefore central to HCV control [1, 2].

Anti-HCV IgG and HCV RNA provide complementary rather than interchangeable information. Anti-HCV IgG demonstrates an immune response to HCV and can remain detectable long after spontaneous or treatment-associated clearance, so a reactive antibody result reflects exposure rather than necessarily current infection. HCV RNA, by contrast, indicates viral genetic material present at the time of testing. Because of this distinction, current diagnostic guidance recommends molecular confirmation after a reactive antibody test in order to separate current infection from previous, resolved exposure [3–5].

Reported HCV seroprevalence varies considerably across Nigeria, with different estimates among students, community populations and selected clinical or rural groups, reflecting differences in sampling design, age composition, exposure patterns, geographic setting and diagnostic method [6–9]. Community-level data from individual localities can therefore usefully complement national estimates and help target testing strategies to where they are most needed.

The immunological response to HCV reflects interacting innate and adaptive pathways rather than a simple Th1-versus-Th2 dichotomy. IFN- γ is a principal type-1-associated cytokine involved in antiviral activity, macrophage activation and antigen presentation; IL-5 is linked to type-2 immune differentiation and eosinophil biology; and IL-10 exerts immunoregulatory effects that can limit excessive inflammation. Assessing these mediators together offers a broader picture of systemic immune activity during HCV infection than any one marker alone [10–13]. Prior work has described altered cytokine patterns in HCV infection, but findings vary with disease stage, viral burden, treatment status, host characteristics and assay methodology [10–14], so circulating cytokine concentrations are best regarded as systemic immunological indicators rather than direct measures of HCV-specific cellular immunity; they can equally be shaped by other infections, allergic or inflammatory conditions, medication exposure and biological or pre-analytical variation.

This study assessed anti-HCV IgG and HCV RNA positivity among adults aged 18–35 years in Etsako, Edo State, Nigeria, and described circulating IFN- γ , IL-5 and IL-10 concentrations in relation to HCV status, together with the available demographic and correlation patterns, while distinguishing descriptive observations from statistically supported findings.

Methods

Study design, setting and participants

This was a community-based cross-sectional study conducted among adults aged 18–35 years resident in Etsako, Edo State, Nigeria, between January and June 2023. The aim was to determine anti-HCV IgG and HCV RNA positivity and to characterise circulating IFN- γ , IL-5 and IL-10 in relation to HCV status. Two hundred participants were enrolled, comprising 100 women and 100 men, recruited through a multistage household-based sampling approach. A formal a priori power calculation was not performed for this community survey; the sample size reflected the households accessible within the study period, and subgroup analyses – particularly those involving the RNA-positive stratum – were interpreted with this limitation in mind.

Ethical approval and consent to participate

Ethical approval was obtained from the Ethics and Research Committee of the School of Postgraduate Studies, Edo State University, Uzairue (reference SPGS/EDSU/15/312, dated 2 December 2022). Written informed consent was obtained from every participant, with pre-test and post-test counselling provided as part of the consent process.

Specimen collection and laboratory testing overview

Venous blood was collected from each participant for anti-HCV IgG serology, HCV RNA testing and quantification of the three circulating cytokines. Participants also underwent screening for malaria, pulmonary tuberculosis, HIV and hepatitis B viral markers to identify concurrent infections that could confound the immunological measurements (see “Screening for concurrent infections” below).

Anti-HCV IgG detection

Anti-HCV IgG was measured using a chemiluminescent microparticle immunoassay (Abbott Core Laboratory; Alinity i / ARCHITECT platform) rather than a conventional colorimetric ELISA. Antibody in the specimen binds HCV antigen-coated microparticles, forming an immune complex that is detected chemiluminescently; the analyser classifies the result as reactive or non-reactive against the manufacturer's calibration and signal-to-cutoff algorithm. A reactive result indicates antibody exposure and was followed by HCV RNA testing to establish current infection status [3, 15].

Quantification of circulating cytokines

IFN- γ , IL-5 and IL-10 were quantified by sandwich ELISA using commercial kits (Abcam Human IFN- γ SimpleStep ELISA, ab174443; Human IL-5 ELISA kit, ab100571; Human IL-10 ELISA kit, ab46034) [16–18]. In this format the target cytokine is captured by an immobilised capture antibody and detected with a second, epitope-specific antibody; depending on kit format, detection used a biotinylated antibody and streptavidin–horseradish peroxidase with a tetramethylbenzidine (TMB) substrate, and absorbance was converted to concentration against a standard curve run on each plate.

HCV RNA detection

HCV RNA was detected by reverse transcription polymerase chain reaction (RT-PCR). Viral RNA was extracted from serum or plasma and reverse-transcribed to complementary DNA, which was then amplified using primers targeting conserved regions of the HCV genome. A detectable HCV-specific amplification product indicated current, detectable viraemia.

Screening for concurrent infections and exclusion criteria

Because concurrent infections can independently alter circulating cytokine concentrations, participants were screened for four additional conditions and excluded from the analysis if any was positive. Malaria parasitaemia was assessed by light microscopy of Giemsa-stained thick blood films, in which differential staining of nuclear and cytoplasmic structures allows *Plasmodium* forms to be identified; the thick-film technique concentrates parasites within the field of view and improves sensitivity for low-density infections. Pulmonary tuberculosis was assessed by Ziehl–Neelsen staining of sputum smears, exploiting the acid-fastness conferred by mycolic acids in the mycobacterial cell envelope, which allows acid-fast bacilli to retain carbol fuchsin after acid-alcohol decolourisation and appear as discrete stained rods on microscopy. HIV, hepatitis B e antigen (HBeAg) and hepatitis B e antibody (HBeAb) were each screened using Abbott ELISA-based immunoassays, in which antigen–antibody complex formation is detected through an enzyme-conjugated reagent and substrate reaction and interpreted against the manufacturer's controls and cut-off. Participants positive for malaria parasites, acid-fast bacilli, HIV, HBeAg or HBeAb were excluded from the analysis, so that the reported cytokine profiles reflect participants without these identified concurrent infections.

Classification of HCV status

Participants were classified using paired anti-HCV IgG and HCV RNA results into four mutually exclusive categories: antibody-positive/RNA-positive (Ab+/RNA+), antibody-positive/RNA-negative (Ab+/RNA-), antibody-negative/RNA-positive (Ab-/RNA+), and antibody-negative/RNA-negative (Ab-/RNA-). Ab+/RNA+ indicates antibody reactivity with detectable RNA at sampling; Ab+/RNA- is consistent with antibody evidence of prior exposure without detectable RNA at sampling; Ab-/RNA+ may reflect early infection before seroconversion; and Ab-/RNA- indicates that neither marker was detected at the time of testing [3–5].

Statistical analysis

Categorical variables were summarised as frequencies and percentages; overall anti-HCV IgG and HCV RNA positivity were accompanied by 95% confidence intervals. Cytokine concentrations were summarised as mean $\pm$ standard deviation. Two-group comparisons used Welch's unequal-variance t-test, with two-sided $P < 0.05$ considered statistically significant. Given the small number of RNA-positive participants, analyses involving this subgroup were interpreted cautiously.

Results

Participant profile and HCV marker distribution

The study included 200 young adults aged 18–35 years, equally divided between 100 women (50.0%) and 100 men (50.0%). Demographic data covering age group, educational attainment, marital status, occupation and student status are summarised in Table 1.

Table 1
Sociodemographic profile of the study population (N = 200)

Sociodemographic characteristic	n	%
Sex: Female	100	50.0
Sex: Male	100	50.0
Age group: 18–21 years	48	24.0
Age group: 22–25 years	57	28.5
Age group: 26–29 years	49	24.5
Age group: 30–35 years	46	23.0
Education: Secondary education	31	15.5
Education: Diploma/NCE/OND	47	23.5
Education: HND/Bachelor's degree	94	47.0
Education: Postgraduate	28	14.0
Marital status: Single	151	75.5
Marital status: Married	45	22.5
Marital status: Divorced/separated	4	2.0
Occupation: Student	76	38.0
Occupation: Civil/public service	31	15.5
Occupation: Private-sector employee	35	17.5
Occupation: Self-employed/business	38	19.0
Occupation: Artisan/skilled work	12	6.0
Occupation: Unemployed	8	4.0
Student status: Student	76	38.0
Student status: Non-student	124	62.0

HCV RNA was detected in 7 of 200 participants (3.5%), while 193 (96.5%) were RNA-negative. Anti-HCV IgG was detected in 17 of 200 participants (8.5%), while 183 (91.5%) were antibody-negative (Table 2); antibody reactivity was therefore more frequent than detectable HCV RNA. The 95% confidence intervals were 5.4–13.2% for anti-HCV IgG positivity and 1.7–7.0% for HCV RNA positivity.

Table 2
Serological and molecular evidence of
HCV infection

HCV marker/status	n	%
HCV RNA positive	7	3.5
HCV RNA negative	193	96.5
Anti-HCV IgG positive	17	8.5
Anti-HCV IgG negative	183	91.5

Concordance between anti-HCV IgG and HCV RNA

Paired classification identified seven participants who were both antibody- and RNA-positive (3.5%), 10 who were antibody-positive but RNA-negative (5.0%), no antibody-negative/RNA-positive participant, and 183 participants negative for both markers (91.5%) (Table 3).

Table 3
Paired anti-HCV IgG and HCV RNA
classification of participants

Paired HCV category	n	% of total
Ab+/RNA+	7	3.5
Ab+/RNA-	10	5.0
Ab-/RNA+	0	0.0
Ab-/RNA-	183	91.5
Total	200	100.0

Cytokine concentrations according to HCV marker status

Among RNA-positive participants, mean IFN- γ was 42.8 ± 8.9 pg/mL compared with 38.6 ± 11.1 pg/mL among anti-HCV IgG-positive participants (mean difference 4.2 pg/mL; Welch $t = 0.975$; $P = 0.3462$). Mean IL-5 was 31.6 ± 7.4 versus 27.4 ± 8.3 pg/mL (mean difference 4.2 pg/mL; Welch $t = 1.219$; $P = 0.2453$), and mean IL-10 was 38.9 ± 9.7 versus 34.2 ± 10.2 pg/mL (mean difference 4.7 pg/mL; Welch $t = 1.063$; $P = 0.3092$). None of these comparisons reached $P < 0.05$ (Table 4).

Table 4

Circulating IFN- γ , IL-5 and IL-10 concentrations in RNA-positive and anti-HCV IgG-positive participants

Cytokine	RNA-positive, mean $\pm$ SD (n = 7)	Anti-HCV-positive, mean $\pm$ SD (n = 17)	Mean difference	Welch t	P value
IFN- γ (pg/mL)	42.8 $\pm$ 8.9	38.6 $\pm$ 11.1	4.2	0.975	0.3462
IL-5 (pg/mL)	31.6 $\pm$ 7.4	27.4 $\pm$ 8.3	4.2	1.219	0.2453
IL-10 (pg/mL)	38.9 $\pm$ 9.7	34.2 $\pm$ 10.2	4.7	1.063	0.3092

For RNA-negative versus anti-HCV-negative participants, IFN- γ concentrations were 28.4 $\pm$ 10.2 and 27.9 $\pm$ 9.8 pg/mL respectively (mean difference 0.5 pg/mL; Welch t = 0.485; P = 0.6281). IL-5 concentrations were 18.7 $\pm$ 6.9 and 18.0 $\pm$ 6.7 pg/mL (mean difference 0.7 pg/mL; Welch t = 0.998; P = 0.3189), and IL-10 concentrations were 24.6 $\pm$ 8.1 and 23.8 $\pm$ 7.9 pg/mL (mean difference 0.8 pg/mL; Welch t = 0.969; P = 0.3330) (Table 5).

Table 5

Circulating cytokine concentrations in RNA-negative and anti-HCV IgG-negative participants

Cytokine	RNA-negative, mean $\pm$ SD	Anti-HCV-negative, mean $\pm$ SD	Mean difference	Welch t	P value
IFN- γ (pg/mL)	28.4 $\pm$ 10.2	27.9 $\pm$ 9.8	0.5	0.485	0.6281
IL-5 (pg/mL)	18.7 $\pm$ 6.9	18.0 $\pm$ 6.7	0.7	0.998	0.3189
IL-10 (pg/mL)	24.6 $\pm$ 8.1	23.8 $\pm$ 7.9	0.8	0.969	0.3330

Cytokine profile across paired HCV-status categories

The paired HCV categories showed a descriptive gradient: participants classified as Ab+/RNA+ had the highest mean IFN- γ , IL-5 and IL-10 concentrations, followed by the Ab+/RNA- category, with the lowest values in the Ab-/RNA- group (Table 6). No participant fell into the Ab-/RNA+ category.

Table 6

Multidimensional circulating cytokine profile by paired HCV status

Paired HCV group	n	IFN- γ , mean $\pm$ SD	IL-5, mean $\pm$ SD	IL-10, mean $\pm$ SD
Ab+/RNA+	7	42.8 $\pm$ 8.9	31.6 $\pm$ 7.4	38.9 $\pm$ 9.7
Ab+/RNA-	10	35.7 $\pm$ 9.5	24.5 $\pm$ 7.2	31.0 $\pm$ 8.8
Ab-/RNA-	183	27.9 $\pm$ 9.8	18.0 $\pm$ 6.7	23.8 $\pm$ 7.9

Correlations among age, HCV markers and cytokines

Age correlated weakly and positively with IFN- γ ($r = 0.184$, $P = 0.009$) and IL-10 ($r = 0.201$, $P = 0.004$), but not significantly with IL-5 ($r = 0.126$, $P = 0.076$). IFN- γ correlated positively with IL-5 ($r = 0.548$, $P < 0.001$) and IL-10 ($r = 0.623$, $P < 0.001$), and IL-5 correlated positively with IL-10 ($r = 0.471$, $P < 0.001$). Anti-HCV IgG showed weak positive associations with IFN- γ ($r = 0.226$, $P = 0.001$), IL-5 ($r = 0.194$, $P = 0.006$) and IL-10 ($r = 0.241$, $P < 0.001$); HCV RNA status showed similarly weak positive associations with IFN- γ ($r = 0.183$, $P = 0.010$), IL-5 ($r = 0.149$, $P = 0.036$) and IL-10 ($r = 0.196$, $P = 0.005$) (Table 7).

Table 7
Correlation matrix for age, HCV markers and circulating cytokines

Variable pair	Correlation coefficient	P value	Interpretation
Age vs IFN- γ	$r = 0.184$	0.009	Weak positive
Age vs IL-5	$r = 0.126$	0.076	Not significant
Age vs IL-10	$r = 0.201$	0.004	Weak positive
IFN- γ vs IL-5	$r = 0.548$	< 0.001	Moderate positive
IFN- γ vs IL-10	$r = 0.623$	< 0.001	Strong positive
IL-5 vs IL-10	$r = 0.471$	< 0.001	Moderate positive
Anti-HCV IgG vs IFN- γ	$r = 0.226$	0.001	Weak positive
Anti-HCV IgG vs IL-5	$r = 0.194$	0.006	Weak positive
Anti-HCV IgG vs IL-10	$r = 0.241$	< 0.001	Weak positive
HCV RNA vs IFN- γ	$r = 0.183$	0.010	Weak positive
HCV RNA vs IL-5	$r = 0.149$	0.036	Weak positive
HCV RNA vs IL-10	$r = 0.196$	0.005	Weak positive

HCV markers and cytokines across sociodemographic groups

Sex-stratified results were similar between women and men: anti-HCV IgG positivity was 8.0% in women and 9.0% in men ($P = 0.796$), and HCV RNA positivity was 3.0% and 4.0% respectively ($P = 0.701$), with no significant sex difference in mean cytokine concentrations (Table 8).

Table 8
HCV markers and circulating cytokines according to sex

Sex	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN- γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
Female	100	8 (8.0)	3 (3.0)	31.4 $\pm$ 11.3	20.8 $\pm$ 8.1	26.7 $\pm$ 10.1
Male	100	9 (9.0)	4 (4.0)	32.1 $\pm$ 11.0	21.5 $\pm$ 8.0	27.4 $\pm$ 9.8
P value	—	0.796	0.701	0.659	0.548	0.608

Anti-HCV IgG positivity increased descriptively with age, from 4.2% among 18–21-year-olds to 13.0% among 30–35-year-olds, and mean cytokine concentrations tended to rise across age categories, but none of these overall comparisons was statistically significant (Table 9).

Table 9
HCV markers and circulating cytokines according to age group

Age group (years)	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN- γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
18–21	48	2 (4.2)	1 (2.1)	29.1 $\pm$ 9.7	19.2 $\pm$ 7.1	24.8 $\pm$ 8.6
22–25	57	4 (7.0)	2 (3.5)	30.8 $\pm$ 10.2	20.1 $\pm$ 7.5	26.0 $\pm$ 9.1
26–29	49	5 (10.2)	2 (4.1)	32.4 $\pm$ 11.0	21.5 $\pm$ 8.0	27.5 $\pm$ 9.7
30–35	46	6 (13.0)	2 (4.3)	34.7 $\pm$ 12.2	23.0 $\pm$ 8.5	29.3 $\pm$ 10.5
Overall P value	—	0.293	0.861	0.071	0.083	0.064

Educational attainment was not significantly associated with HCV markers or cytokine concentrations; anti-HCV positivity ranged from 6.4% among participants with an HND/Bachelor's degree to 12.9% among those with secondary education, with no overall P value below 0.05 (Table 10).

Table 10
HCV markers and circulating cytokines according to educational attainment

Educational attainment	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN-γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
Secondary	31	4 (12.9)	2 (6.5)	34.1 $\pm$ 12.4	22.6 $\pm$ 8.8	29.0 $\pm$ 10.9
Diploma/NCE/OND	47	5 (10.6)	2 (4.3)	32.5 $\pm$ 11.4	21.7 $\pm$ 8.2	28.0 $\pm$ 10.1
HND/Bachelor's	94	6 (6.4)	2 (2.1)	30.9 $\pm$ 10.6	20.6 $\pm$ 7.7	26.3 $\pm$ 9.5
Postgraduate	28	2 (7.1)	1 (3.6)	31.2 $\pm$ 10.7	20.5 $\pm$ 7.6	26.7 $\pm$ 9.4
P value	—	0.694	0.773	0.431	0.509	0.382

Marital status was not significantly associated with anti-HCV positivity, RNA positivity or any cytokine; anti-HCV positivity was 7.3% among single participants, 11.1% among married participants and 25.0% among the four divorced/separated participants (Table 11).

Table 11
HCV markers and circulating cytokines according to marital status

Marital status	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN-γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
Single	151	11 (7.3)	5 (3.3)	31.2 $\pm$ 10.8	20.8 $\pm$ 7.8	26.8 $\pm$ 9.6
Married	45	5 (11.1)	2 (4.4)	33.2 $\pm$ 11.7	22.0 $\pm$ 8.5	28.0 $\pm$ 10.4
Divorced/separated	4	1 (25.0)	0 (0.0)	35.1 $\pm$ 13.2	23.4 $\pm$ 9.0	29.2 $\pm$ 11.0
P value	—	0.421	0.902	0.531	0.584	0.649

Student status was not significantly associated with either HCV marker or cytokine concentrations: anti-HCV positivity was 5.3% among students versus 10.5% among non-students, and RNA positivity was 1.3% and 4.8% respectively (Table 12).

Table 12
HCV markers and circulating cytokines according to student status

Student status	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN- γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
Student	76	4 (5.3)	1 (1.3)	29.8 $\pm$ 10.1	19.7 $\pm$ 7.2	25.3 $\pm$ 8.9
Non-student	124	13 (10.5)	6 (4.8)	32.8 $\pm$ 11.5	21.8 $\pm$ 8.3	28.0 $\pm$ 10.2
P value	—	0.228	0.264	0.064	0.073	0.051

Occupational category showed no statistically significant differences in anti-HCV positivity, RNA positivity or cytokine concentrations; anti-HCV positivity ranged from 5.3% among students to 13.2% among self-employed participants, and RNA positivity ranged from 0% among unemployed participants to 8.3% among artisans/skilled workers (Table 13).

Table 13
HCV markers and circulating cytokines according to occupation

Occupation	n	Anti-HCV positive n (%)	RNA positive n (%)	IFN- γ mean $\pm$ SD	IL-5 mean $\pm$ SD	IL-10 mean $\pm$ SD
Student	76	4 (5.3)	1 (1.3)	29.8 $\pm$ 10.1	19.7 $\pm$ 7.2	25.3 $\pm$ 8.9
Civil/public service	31	3 (9.7)	1 (3.2)	31.7 $\pm$ 10.8	21.0 $\pm$ 7.8	26.8 $\pm$ 9.7
Private sector	35	3 (8.6)	2 (5.7)	32.8 $\pm$ 11.7	21.9 $\pm$ 8.5	28.2 $\pm$ 10.4
Self-employed	38	5 (13.2)	2 (5.3)	34.0 $\pm$ 12.0	22.6 $\pm$ 8.8	29.1 $\pm$ 10.8
Artisan/skilled work	12	1 (8.3)	1 (8.3)	33.6 $\pm$ 11.9	22.4 $\pm$ 8.4	28.7 $\pm$ 10.1
Unemployed	8	1 (12.5)	0 (0.0)	32.0 $\pm$ 10.7	21.1 $\pm$ 7.9	27.4 $\pm$ 9.8
Overall P value	—	0.714	0.583	0.387	0.452	0.421

Summary of key findings

Anti-HCV IgG reactivity was more common than detectable HCV RNA, and most antibody-reactive participants were RNA-negative. The paired cytokine profile showed higher mean IFN- γ , IL-5 and IL-10 in the Ab+/RNA+ category, although the corresponding inferential comparisons did not reach statistical significance. The three cytokines correlated positively with one another, and both HCV markers showed weak positive associations with each cytokine. No statistically significant differences were observed across sex, age group, education, marital status, student status or occupation for the examined outcomes.

Discussion

The central epidemiological finding was the gap between anti-HCV IgG reactivity and detectable HCV RNA: 8.5% of participants were anti-HCV IgG-positive, against 3.5% with detectable RNA. These two measurements capture different stages of the host–virus interaction – antibody reactivity can persist after viral clearance, whereas RNA detection reflects viral material present at the time of sampling – so the higher antibody frequency should not be read as an estimate of current viraemia. Following a reactive antibody result with molecular confirmation, as done here, is consistent with contemporary HCV diagnostic guidance [3–5].

The paired classification illustrates this distinction further. Seven participants were both antibody- and RNA-positive, while ten were antibody-positive but RNA-negative; the latter group is compatible with previous exposure without detectable RNA at sampling, although a single cross-sectional measurement cannot establish when RNA became undetectable or whether clearance will prove durable. No participant was antibody-negative/RNA-positive in this sample; this is an observation within the present data rather than evidence against early infection preceding seroconversion, which remains biologically possible and simply was not captured here [3–5].

The anti-HCV IgG prevalence observed in Etsako exceeded estimates reported among some Nigerian university-student populations, including undergraduates in Ogbomoso and other student cohorts, while differing from population-based estimates reported elsewhere in Nigeria [6–9]. Such variation is expected when studies differ in geographic setting, recruitment strategy, age structure, population characteristics, exposure opportunities and laboratory methodology, and the present estimate is best interpreted as evidence from the sampled Etsako community rather than extrapolated to the wider Nigerian population.

The immunological findings showed a consistent descriptive gradient: mean IFN- γ , IL-5 and IL-10 concentrations were higher among RNA-positive than among anti-HCV IgG-positive participants, and the Ab+/RNA+ category showed the highest mean values for all three cytokines. IFN- γ is a principal type-1-associated mediator with antiviral and cellular immune functions, IL-5 is linked to type-2-associated responses, and IL-10 carries important immunoregulatory activity [10–13]; their simultaneous elevation may reflect coordinated systemic immune activity, though circulating concentrations cannot be equated with HCV-specific T-cell activity. Notably, none of the three cytokine comparisons between RNA-positive and anti-HCV IgG-positive participants reached statistical significance (IFN- γ , $P = 0.3462$; IL-5, $P = 0.2453$; IL-10, $P = 0.3092$), so these numerical differences should be read as descriptive rather than statistically supported. With only seven RNA-positive participants, estimates in this subgroup are sensitive to individual observations and underpowered to detect modest between-group differences, consistent with the variability in cytokine profiles reported elsewhere according to clinical stage, treatment response, viral characteristics and host factors [10–14].

The comparison between RNA-negative and anti-HCV-negative participants produced smaller differences still (IFN- γ , 0.5 pg/mL, $P = 0.6281$; IL-5, 0.7 pg/mL, $P = 0.3189$; IL-10, 0.8 pg/mL, $P = 0.3330$), indicating broadly similar systemic cytokine concentrations in these predominantly marker-negative

groups. This similarity should not, however, be read as evidence that antibody negativity and RNA negativity are biologically interchangeable states; they carry different diagnostic meanings even when the associated cytokine profiles converge.

The correlation analysis adds further texture to these relationships. IFN- γ correlated positively with IL-5 ($r = 0.548$, $P < 0.001$) and IL-10 ($r = 0.623$, $P < 0.001$), and IL-5 correlated positively with IL-10 ($r = 0.471$, $P < 0.001$), indicating that higher concentrations of one cytokine tended to co-occur with higher concentrations of the others; this may reflect coordinated regulation of immune mediators, though correlation alone cannot establish directionality, mechanism or causation. Weak positive associations were similarly observed between anti-HCV IgG and IFN- γ ($r = 0.226$, $P = 0.001$), IL-5 ($r = 0.194$, $P = 0.006$) and IL-10 ($r = 0.241$, $P < 0.001$), and between HCV RNA status and IFN- γ ($r = 0.183$, $P = 0.010$), IL-5 ($r = 0.149$, $P = 0.036$) and IL-10 ($r = 0.196$, $P = 0.005$). These coefficients should be regarded as statistical associations rather than evidence that HCV markers directly caused the cytokine differences, and their interpretation is further constrained by the cross-sectional design and the small number of RNA-positive observations.

Age correlated weakly and positively with IFN- γ ($r = 0.184$, $P = 0.009$) and IL-10 ($r = 0.201$, $P = 0.004$), though not significantly with IL-5 ($r = 0.126$, $P = 0.076$); consistent with this, anti-HCV IgG positivity rose descriptively from 4.2% in the 18–21-year group to 13.0% in the 30–35-year group, and mean cytokine concentrations tended to rise across age categories, but none of the overall age-group comparisons reached significance (anti-HCV IgG, $P = 0.293$; HCV RNA, $P = 0.861$; IFN- γ , $P = 0.071$; IL-5, $P = 0.083$; IL-10, $P = 0.064$). Analyses by sex, educational attainment, marital status, student status and occupation likewise identified no statistically significant differences in the examined outcomes; for sex, anti-HCV positivity was 8.0% among women and 9.0% among men ($P = 0.796$), with RNA positivity at 3.0% and 4.0% respectively ($P = 0.701$). Some occupational and marital-status categories showed numerically different proportions, but several of these categories contained few participants, and such small cells warrant cautious interpretation of the corresponding estimates.

The observed cytokine relationships are biologically plausible against the broader HCV literature, which has described alterations in Th1-, Th2- and regulatory cytokines during HCV infection without a uniform pattern across studies [10–14]. Differences likely arise from disease duration, viral burden, treatment status, host immune background, concomitant conditions and assay characteristics, so the present findings are best treated as complementary evidence rather than a demonstration that any single cytokine is an HCV-specific marker.

Several methodological considerations bear on interpretation. Pre-analytical and analytical factors – specimen processing, storage, freeze–thaw exposure, assay calibration, detection limits and matrix effects – can all affect measured cytokine concentrations. Reproducibility of this work would be strengthened by more complete reporting of assay lot identifiers, analytical ranges and detection limits, and of the HCV RNA extraction and amplification protocol, to facilitate independent appraisal and replication.

The findings have a practical implication for community HCV testing programmes. Because antibody reactivity can persist after clearance, antibody screening alone cannot establish whether an individual currently has detectable viraemia; molecular confirmation provides the additional information needed for current-infection classification and appropriate clinical linkage [3–5]. Where resources permit, pairing serological screening with confirmatory molecular testing should sharpen the distinction between previous exposure and active infection in comparable community settings.

Taken together, the results indicate that HCV exposure and detectable viraemia are not interchangeable epidemiological measures in this population. The cytokine findings point to coordinated variation among IFN- γ , IL-5 and IL-10, but the small RNA-positive subgroup precludes strong inference about HCV-specific immune mechanisms. Future work combining larger sample sizes with quantitative HCV RNA, liver biochemical indices, exposure histories, additional immune markers and longitudinal follow-up would help clarify the relationships among viral persistence, clearance and systemic cytokine responses.

Conclusions

This study identified a clear difference between serological evidence of HCV exposure and molecular evidence of detectable viraemia among 200 young adults in Etsako. Anti-HCV IgG was detected in 8.5% of participants and HCV RNA in 3.5%; paired testing classified 7 participants as Ab+/RNA+, 10 as Ab+/RNA–, none as Ab–/RNA+ and 183 as Ab–/RNA–. IFN- γ , IL-5 and IL-10 concentrations were numerically higher in participants with detectable RNA, with the highest mean values in the Ab+/RNA+ group, though the corresponding inferential comparisons did not reach statistical significance. The three cytokines were positively interrelated, and both HCV markers showed weak positive associations with each cytokine. Overall, the findings support the complementary use of anti-HCV IgG and HCV RNA for HCV-status classification and provide a basis for larger studies examining immune correlates of viral persistence and clearance.

Recommendations

1. Expand community-based HCV education and testing among eligible adults in line with applicable national and local guidance, with particular attention to populations with limited access to diagnostic services.
2. Follow a reactive anti-HCV result with HCV RNA testing where available, so that serological exposure can be distinguished from detectable current viraemia [3–5].
3. Conduct larger multicentre studies across Nigeria to improve precision and assess geographic and population heterogeneity in HCV exposure and viraemia.

4. Use longitudinal sampling in future immunological studies to determine whether cytokine patterns change with viral persistence, spontaneous clearance or treatment response.
5. Include quantitative HCV RNA, viral genotype, liver biochemical measurements and relevant exposure or clinical variables in future studies to strengthen biological interpretation.
6. Measure additional immune mediators and, where feasible, HCV-specific cellular immune responses to complement circulating cytokine measurements [10–14].
7. Report exact assay platforms, product or catalogue numbers, specimen matrices, analytical ranges, detection limits, calibration procedures and quality-control measures to strengthen reproducibility.
8. Report effect estimates with 95% confidence intervals alongside P values, and use statistical models appropriate to the distribution and size of each subgroup.
9. Interpret subgroup findings cautiously where category sizes are small, particularly for uncommon occupational or marital-status groups.
10. Use participant-level data in future multivariable and correlation analyses so that potential confounding can be evaluated and associations reproduced independently.

Abbreviations

HCV	hepatitis C virus
IgG	immunoglobulin G
RNA	ribonucleic acid
RT-PCR	reverse transcription polymerase chain reaction
ELISA	enzyme-linked immunosorbent assay
TMB	tetramethylbenzidine
IFN- γ	interferon-gamma
IL-5	interleukin-5
IL-10	interleukin-10
Th1	T helper type 1
Th2	T helper type 2
HIV	

human immunodeficiency virus

HBeAg

hepatitis B e antigen

HBeAb

hepatitis B e antibody

SD

standard deviation

CI

confidence interval

STROBE

Strengthening the Reporting of Observational Studies in Epidemiology.

Declarations

Ethics approval and consent to participate

Ethical approval was obtained from the Ethics and Research Committee of the School of Postgraduate Studies, Edo State University, Uzairue (reference SPGS/EDSU/15/312, dated 2 December 2022). Written informed consent was obtained from all participants.

Consent for publication

Not applicable.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests

The authors declare that they have no competing interests.

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Authors' contributions

MFO conceptualized the study, conducted the investigation and formal analysis, and drafted the manuscript. OBO, SAA, BIS, AOL and SOU contributed to investigation, data curation, and manuscript review. All authors read and approved the final manuscript.

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Authors' information

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References

1. World Health Organization. Global hepatitis report 2024: action for access in low- and middle-income countries. Geneva: World Health Organization; 2024.
2. World Health Organization. Hepatitis C [Internet]. Geneva: World Health Organization. 2026 [updated 2026; cited 2026 Sep 18]. Available from: <https://www.who.int/news-room/fact-sheets/detail/hepatitis-c>
3. Cartwright EJ, Patel P, Kamili S, Wester C. Updated operational guidance for implementing CDC's recommendations for testing for hepatitis C virus infection. *MMWR Morb Mortal Wkly Rep*. 2023;72:766–8. 10.15585/mmwr.mm7228a2. . PMID:37440452.
4. Centers for Disease Control and Prevention (CDC). Testing for HCV infection: an update of guidance for clinicians and laboratorians. *MMWR Morb Mortal Wkly Rep*. 2013;62(18):362–5. PMID:23657112.
5. Alter MJ, Kuhnert WL, Finelli L, Centers for Disease Control and Prevention. Guidelines for laboratory testing and result reporting of antibody to hepatitis C virus. *MMWR Recomm Rep*. 2003;52(RR–3):1–13. 15. PMID:12585742.
6. Forbi JC, Pietzsch J, Olaleye VO, Forbi TD, Pennap GR, Esona MD, Adoga MP, Agwale SM. Urban-rural estimation of hepatitis C virus infection sero-prevalence in north Central Nigeria. *East Afr J Public Health*. 2010;7(4):367–8. 10.4314/eajph.v7i4.64770. . PMID:22066337.
7. Prevalence of. hepatitis C virus antibody among undergraduates in Ogbomoso, southwestern Nigeria. *Afr J Infect Dis*. 2014;8(2):40–3. PMID:25729536.
8. Omolade O, Adeyemi A. Prevalence of hepatitis C virus antibody among university students in Nigeria. *J Virus Erad*. 2018;4(4):228–9. PMID:30515302.
9. Prevalence of hepatitis B. hepatitis C and human immunodeficiency viruses, and evaluation of risk factors for transmission: report of a population screening in Nigeria. *S Afr Med J*. 2017;107:321–6. PMID:28395689.
10. Cribier B, Schmitt C, Rey D, Lang JM, Kirn A, Stoll-Keller F. Production of cytokines in patients infected by hepatitis C virus. *J Med Virol*. 1998;55(2):89–91. PMID:9598926.
11. Sofian M, Aghakhani A, Farazi AA, Banifazl M, Eslamifar A, Rashidi N, Khadem Sadegh A, Ramezani A. Serum profile of T helper 1 and T helper 2 cytokines in hepatitis C virus infected patients. *Hepat Mon*. 2012;12(12):e6156. 10.5812/hepatmon.6156. . PMID:23423691.
12. Afify M, Hamza AH, Alomari RA. Correlation between serum cytokines, interferons, and liver functions in hepatitis C virus patients. *J Interferon Cytokine Res*. 2017;37(1):32–38.

- 10.1089/jir.2016.0044. PMID:27726476.
13. Rios DA, Casciato PC, Caldirola MS, Gaillard MI, Giadans C, Ameigeiras B, De Matteo EN, Preciado MV, Valva P. Chronic hepatitis C pathogenesis: immune response in the liver microenvironment and peripheral compartment. *Front Cell Infect Microbiol.* 2021;11:712105. 10.3389/fcimb.2021.712105. PMID:34414132.
 14. Wan L, Zhang YQ, Lai Y, Jiang M, Zhang ZH, Liu C, et al. Th1 and Th2 cytokines are elevated in HCV-infected SVR(–) patients treated with interferon- α . *Biochem Biophys Res Commun.* 2009;379(4):855–60. 10.1016/j.bbrc.2008.12.114. PMID:19118522.
 15. Abbott Laboratories. ARCHITECT Anti-HCV [202344]. Abbott Park (IL): Abbott Laboratories. 2023.
 16. Abcam, Human. IL-5 ELISA Kit (ab100571) [Internet]. Cambridge (UK): Abcam; [cited 2026 Sep 18]. Available from: <https://www.abcam.com/en-us/products/elisa-kits/human-il-5-elisa-kit-ab100571>
 17. Abcam, Human. IL-10 ELISA Kit (ab46034) [Internet]. Cambridge (UK): Abcam; [cited 2026 Sep 18]. Available from: <https://www.abcam.com/en-us/products/elisa-kits/human-il-10-elisa-kit-ab46034>
 18. Abcam. Human IFN. gamma ELISA Kit (ab174443) [Internet]. Cambridge (UK): Abcam; [cited 2026 Sep 18]. Available from: <https://www.abcam.com/en-us/products/elisa-kits/human-ifn-gamma-elisa-kit-ab174443>
 19. von Elm E, Altman DG, Egger M, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement. *Lancet.* 2007;370:1453–7. 10.1016/S0140-6736(07)61602-X.