LABUPDATE no. 57

November 2024

# NEW ONE-STEP ALGORITHM FOR HEPATITIS C DIAGNOSIS

Hepatitis C virus (HCV) will establish persistence in up to 80% of persons who become infected, with a risk of subsequent long-term complications, including decompensated cirrhosis and hepatocellular carcinoma if left untreated. While previous treatment regimens were associated with significant side effects and a suboptimal success rate in treating chronic HCV infection, new direct-acting antiviral (DAA) treatment is paving the way for HCV cure and elimination. Successful treatment, defined as a sustained virological response (i.e. undetectable HCV RNA at 12 or 24 weeks after a course of treatment) is achievable in up to 99% of those with chronic HCV infection. Pan-genotypic DAA regimens also negate the absolute need to establish the HCV genotype prior to treatment initiation, typically for a 12-week course.

Even though curative treatment is now available, a mere 20% of persons with chronic HCV infection are estimated to be aware that they are infected, and only 15% have received treatment. To address this, the World Health Organization (WHO) has set the aim that 80% of eligible HCV-infected persons should receive treatment, with the goal to reduce HCV incidence by 90%, and HCV-related mortality by 65% to effectively eliminate HCV globally by 2030. The identification of chronic HCV infections by means of widespread diagnostic screening is key to achieving this goal, especially among those at high risk such as people who inject drugs (PWID) and men who have sex with men (MSM). Although blood and blood products are now routinely screened for HCV, any persons who received such products prior to the introduction of screening practices (prior to 1992 in South Africa) should also be tested for HCV. HCV infection has also been linked to prior inadequate infection control measures with parenteral administration of therapy in South African cohorts.

## LABORATORY DIAGNOSIS

The various laboratory assays available for laboratory diagnosis and monitoring are outlined in Table 1.

**Table 1: Hepatitis C laboratory assays**

| **Hepatitis C antibody test** | |
|---|---|
| **Test mnemonic:** | **HEPC** |

- Anti-HCV antibody detection by means of enzyme immunoassays (EIAs) remains the recommended screening method to detect HCV infection
- Current third generation HCV antibody EIA assays are more sensitive and specific than older generation assays, with a window period of approximately 8 weeks
- A negative HCV antibody result effectively rules out HCV infection except in very recent exposures
- A positive HCV antibody result always requires further testing with a direct virus detection method to distinguish between a current HCV infection and past resolved HCV infection (since HCV antibodies will remain detectable)
- HCV antibody tests should not be used to screen for reinfection in successfully treated or recovered patients

**Hepatitis C Immunoblot (Western Blot)**

| Test mnemonic: | HEPCWB |
|---|---|

- In low prevalence settings, the positive predictive value of HCV antibody EIA assays will be lower, which may increase the number of false positive HCV antibody results
- While further testing to determine HCV viraemia remains advisable in case of any HCV antibody positive result, Ampath has established from internal data that the likelihood of a detectable viraemia is significantly lower within what are considered low sample cut-off ranges
- In this group, the supplemental HCV Immunoblot (Western blot) is more useful to exclude false HCV antibody EIA reactivity, owing to its higher specificity

**Hepatitis C PCR / viral load**

| Test mnemonic: | HEPCPCR / HCVVL |
|---|---|

- Traditionally, molecular techniques to detect HCV ribonucleic acid (RNA), either with a qualitative polymerase chain reaction (PCR) or a quantitative PCR (viral load) is recommended for confirmation where the screening HCV antibody EIA is positive
- While this method remains the gold standard to confirm HCV infection, these assays are currently more expensive than serology assays and usually performed only in centralised molecular laboratories
- The hepatitis C viral load is also recommended to confirm a sustained virological response (SVR) 12 or 24 weeks after the completion of a course of DAA treatment to determine whether the patient has been successfully treated

**Hepatitis C core antigen test (NEW)**

| Test Mnemonic | HEPCAG |
|---|---|

- In view of the availability of pan-genotypic HCV treatment and the goal to eliminate viral hepatitis, there is a global drive towards simplified and more cost-effective HCV diagnostic algorithms
- Recent updates to international guidelines now recommend that an HCV core antigen assay can be used instead of molecular techniques to confirm the presence of HCV viraemia, which is not only a more cost-effective test to perform, but also an easy test to add to the specimen used for the HCV antibody test, using the same serology platform
- The automated chemiluminescent HCV core antigen immunoassay evaluated by Ampath is deemed a surrogate marker for viral replication, correlating with a viral load of approximately 3000 IU/mL or higher, with exceptional specificity
- The vast majority of HCV RNA positive specimens usually have HCV viral loads over 10 000 IU/ml
- Although only a small percentage of HCV-infected persons will have lower viral loads that might fall below the detection limit of the HCV core antigen assay, additional molecular testing should be considered in HCV antibody positive patients if the HCV core antigen tests negative

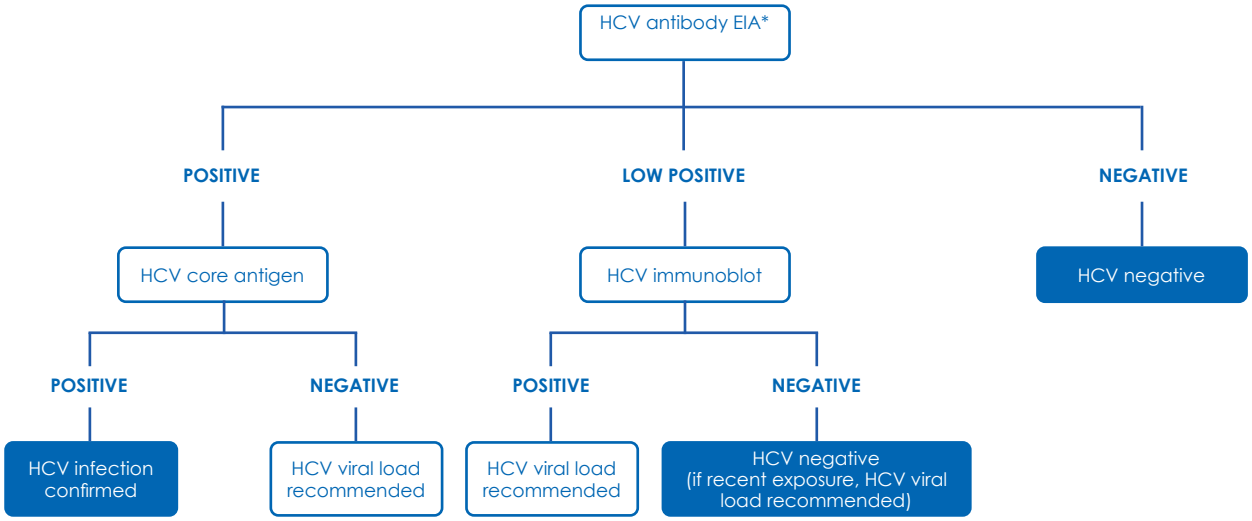
**Hepatitis C genotyping**

| Test Mnemonic: | HCVGEN |
|---|---|

- The need to perform HCV genotypic testing prior to treatment initiation is no longer considered an absolute requirement in international guidelines where pan-genotypic DAA treatment is available as a first-line regimen, and particularly in low-resource settings.

## NEW AMPATH HCV DIAGNOSTIC ALGORITHM

Ampath laboratories will introduce a new algorithm for HCV diagnosis to facilitate a shorter and more cost-effective pathway to treatment. As outlined in Figure 1, HCV antibody EIA-positive specimens will automatically be tested for the presence of an HCV core antigen on a serology platform. A positive HCV core antigen result serves as confirmation of the presence of HCV viraemia, with further assessment for DAA treatment recommended. For negative HCV core antigen test results, the positive HCV antibody result is




*EIA: Enzyme immunoassay

**FIGURE 1: HEPATITIS C VIRUS (HCV) DIAGNOSTIC ALGORITHM**

most likely in keeping with either a prior (resolved) HCV infection or a possible false positive result due to cross-reactive antibody responses. In a small percentage of patients, this may also be in keeping with a low viraemia below the detection limit of the HCV core antigen assay, as such further testing by means of HCV PCR or viral load is recommended.

Low positive-HCV antibody EIA results, on the other hand, will be submitted for a supplemental HCV antibody test, namely the HCV Immunoblot (Western blot) assay. If the HCV Immunoblot confirms the presence of HCV antibodies, confirmation of HCV viraemia by means of HCV PCR/viral load or HCV core antigen is recommended.

**NOTE:** The supplemental tests will not be added automatically for cash- paying patients.

## REFERENCES

1. Spearman CW, Dusheikob GM, Hellardd M, Sonderup M. Hepatitis C. 2019. Lancet 394:1451–466.
2. Manns MP, Maasoumy B. 2022. Breakthroughs in hepatitis C research: From discovery to cure. Nature reviews Gastroenterology and Hepatology 19(8):533–550.
3. Sonderup MW, Horak J, Smuts H, Saayman J, Boretti L, Black J. 2021. Expanding the epidemiological understanding of hepatitis C in South Africa: Perspectives from a patient cohort in a rural town. South African Medical Journal 1: 111(8):783–788.
4. Jiang X, Chang L, Yan Y, Ji H, Sun H, Guo F, Wang L. 2021. A study based on four immunoassays: Hepatitis C virus antibody against different antigens may have unequal contributions to detection. Virology Journal 3:18(1):137.
5. Galli C, Julicher P, Plebani M. 2018. HCV core antigen comes of age: a new opportunity for the diagnosis of hepatitis C virus infection. Clinical Chemistry and Laboratory Medicine (CCLM) 24:56(6):880–888.
6. World Health Organization. 2022. Updated recommendations on treatment of adolescents and children with chronic HCV infection, and HCV simplified service delivery and HCV diagnostics. World Health Organization; 18 October 2022.
7. Pawlotsky JM, Negro F, Aghemo A, Berenguer M, Dalgard O, Dusheiko G, Marra F, Puoti M, Wedemeyer H. 2020. European Association for the Study of the Liver. EASL recommendations on treatment of hepatitis C: Final update of the series. Journal of Hepatology 1:73(5):1170–1218.
8. Tillmann HL. 2014. Hepatitis C virus core antigen testing: role in diagnosis, disease monitoring and treatment. World Journal of Gastroenterology (WJG) 6:20(22):6701.