"WHY WERE HARVEY ALTER, MICHAEL HOUGHTON, AND CHARLES RICE AWARDED THE NOBEL PRIZE FOR THEIR DISCOVERY OF THE HEPATITIS C VIRUS?"

Harshitha Ramu[1]

[1] Master's student Molecular Mechanisms of Disease, Radboud university medical center, Nijmegen, The Netherlands

Abstract

The 2020 Nobel Prize in Physiology or Medicine was jointly awarded to three scientists—Harvey J. Alter, Michael Houghton, and Charles M. Rice—for their discovery of the hepatitis C virus. Their work made a paramount impact on blood-borne hepatitis, a global health threat that can cause cirrhosis and different liver cancers in infected patients. The prior identification and characterisation of hepatitis A and B viruses were vital; however, a large number of hepatitis cases could not be explained. The discovery of the hepatitis C virus by these scientists aided in explaining the majority of the remaining chronic hepatitis cases and in the rapid development of diagnostic blood tests and antiviral drugs that have saved millions of lives and greatly reduced the burden on healthcare systems. The world is currently presented with an opportunity to eradicate hepatitis C virus infections altogether. However, exceptional international collaboration and financing are needed to implement widespread diagnostic testing and distribution of antiviral drugs to rid the world of the hepatitis C virus.

KEYWORDS: hepatitis C virus; Nobel Prize; antiviral drugs; infectious diseases

Hepatitis, commonly known as liver inflammation, is typically caused by viral infections, although other factors such as alcoholism, environmental toxins, and certain autoimmune conditions can contribute to its aetiology as well [1, 2]. The discovery of hepatitis viruses is one of the most important scientific breakthroughs of the past five decades. In the second half of the 20th century, two different hepatitis viruses, namely the hepatitis A and B virus, were identified [3, 4]. The hepatitis A virus is known to infect people through the ingestion of the virus in contaminated food and water, seldom causing severe long-term effects [5]. It is a highly contagious virus whose spread is now controlled in most countries by the introduction of a vaccine in 1995 [5]. The hepatitis B virus is transmitted through infected blood and other bodily fluids and is considered a more serious disease as it can stay dormant for long periods, after which it can induce liver cirrhosis and cancer [5]. Blood-borne hepatitis is a major concern for global health, annually causing over a million deaths worldwide, comparable to other serious conditions such as tuberculosis and HIV-AIDS [6]. Although the discovery of hepatitis A and B explained the cause of many hepatitis cases, a large number remained unexplained [7]. This article describes the work of three scientists that led to the discovery of the hepatitis C virus, which explained these infections, placing the world in a position to eventually eliminate viral hepatitis [8].

A mysterious viral agent

The detection and characterisation of infectious agents are pivotal for identifying disease outbreaks, for developing effective intervention strategies, and for epidemiological studies. Jaundice, a striking symptom common to hepatitis infections, was known since antiquity [3]. Epidemics of jaundice were a menace during the two world wars, and the cause was attributed to poor sanitary living conditions of soldiers [3]. Although the cause of these cases was traced back to the hepatitis A virus, it was only after the identification of the hepatitis B virus and the advent of novel molecular biology techniques that scientists identified the hepatitis A virus in stool samples [3]. Hepatitis A is now known to be an RNA virus belonging to the family *Picornaviridae*, transmissible through contaminated food and water [3]. The spread of the hepatitis A virus is greatly limited by the introduction of a vaccine in 1995 [5].

In 1967, Dr. Baruch Blumberg and his colleagues discovered the hepatitis B virus by finding significant amounts of the hepatitis B surface antigen, formerly termed the "Australia antigen", in the serum of an Aboriginal Australian [9]. The identification of this antigen resolved problems faced by scientists and healthcare providers for decades—the absence of a specific biomarker to distinguish between different types of viral hepatitides [10]. Two years later, the hepatitis B vaccine was invented, and Dr. Blumberg was awarded the Nobel Prize in Physiology or Medicine for his discovery [11]. Along with aiding physicians in the diagnosis of asymptomatic carriers of the virus, the safety of blood transfusions was greatly improved [12].

Around the same time, Harvey J. Alter was studying the incidence of hepatitis in patients who had received blood transfusions at the National Institute of Health. After performing diagnoses for the newly discovered hepatitis A and B viruses, Alter and his colleagues showed that a large proportion of cases remained unexplained [7]. In 1978, the same group demonstrated that the blood from these hepatitis A and B negative patients was able to infect chimpanzees and cause an increase in the levels of biomarkers associated with typical liver inflammation [13]. This unknown disease was appropriately named "non-A, non-B" hepatitis, a novel, severe form of hepatitis whose aetiology was yet to be determined. Assays involving chloroform inactivation, electron microscopy, and filtration studies all indicated the causative agent to be an enveloped virus of about 45-60 nm in diameter [14]. However, conventional cultivation experiments and serology approaches failed, partly due to the low amount of viral antigen in infected sera [14]. Scientists were frustrated for 15 years until the causative agent was finally identified and characterised as the hepatitis C virus due to the collective efforts of three scientists—Harvey Alter, Michael Houghton, and Charles Rice [8].

Discovery of the Hepatitis C Virus

After the establishment of the novel "non-A, non-B" viral hepatitis in post-transfusion patients by Alter and colleagues, the identification and characterisation of the unknown virus was of grave importance. As all conventional isolation strategies failed, the virus evaded detection for more than a decade until 1989, when Michael Houghton—a scientist at

"Why were Harvey Alter, Michael Houghton, and Charles Rice awarded the Nobel Prize for their discovery of the hepatitis C Virus?" - Ramu

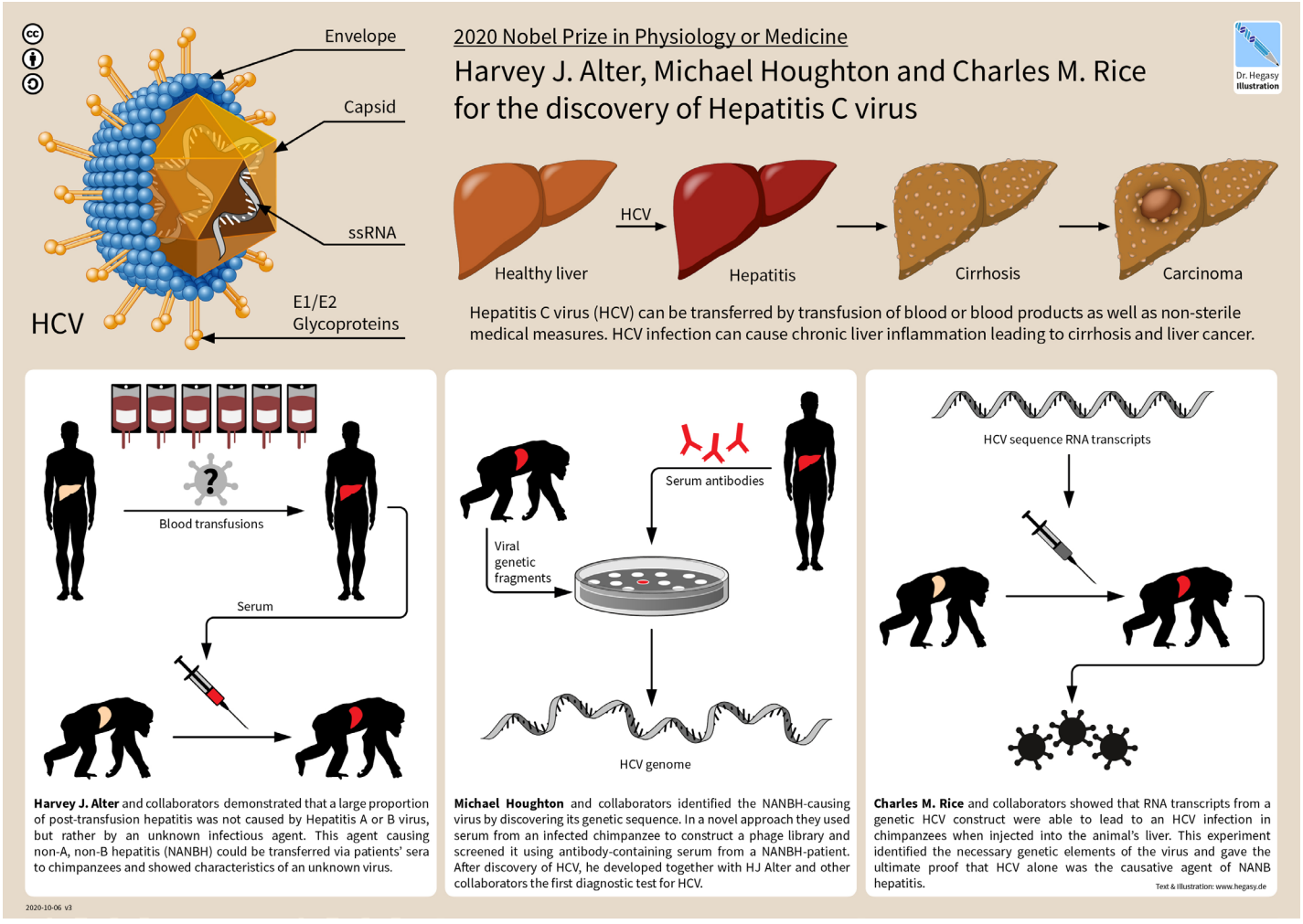

Figure 1: A summary of three key discoveries by Harvey J. Alter, Michael Houghton, and Charles M. Rice that led to the discovery of the hepatitis C virus. *Illustration: www.hegasy.de*

the pharmaceutical company Chiron—isolated the genetic sequence of the virus [15]. Houghton and his colleagues collected and curated a library of cDNA fragments, isolated from the blood of a chronically infected chimpanzee [15]. The rationale behind their experiments was to exclude fragments derived from the genome of the chimpanzee to isolate and identify the fragments that originated in the uncharacterised "non-A, non-B" hepatitis virus. The group achieved this by using antibodies isolated from the sera of chronically infected patients to identify the rare clones of DNA that encoded viral proteins [15]. After a series of rigorous experiments, one positive clone was identified to contain the partial sequence of the agent responsible for the mysterious infections [15]. Further research indicated that it was derived from an enveloped, positive-sense, single-stranded RNA molecule of approximately 10,000 nucleotides in length, belonging to the family of *Flaviviruses* [15]. The detection of antibodies in chronic "non-A, non-B" hepatitis patients specific to this newly discovered virus further validated the findings [16]. The novel virus was given the name "hepatitis C virus", and the molecular techniques used for its identification were recognised as potential approaches for the identification of other unknown infectious agents as well [17]. After this revolutionary discovery, the first diagnostic test for the hepatitis C virus was developed [18].

Although the discovery of the hepatitis C virus was conclusive, a crucial question remained: could the virus alone replicate and cause "non-A, non-B" hepatitis? This question was answered by scientist Charles M. Rice and colleagues at Washington University. Previously, the researchers observed an uncharacterised genomic region at the end of the viral genome that they speculated to be important for its replication [19]. Rice and his colleagues also detected specific genomic mutations in the virus isolated from patient samples that he hypothesised to prevent efficient viral replication [19]. Therefore, Rice and his colleagues genetically engineered a hepatitis C virus RNA transcript, excluding the debilitating genetic variants [19]. On injection of the construct directly into the livers of the chimpanzees, viral titres were detected in the blood, and pathological features similar to those observed in chronically infected human patients were observed [19]. These crucial experiments identified the genetic elements essential for viral replication and provided the missing piece of the puzzle that the hepatitis C virus alone was able to establish the "non-A, non-B" hepatitis observed in patients receiving blood transfusion [19].

A remarkable Nobel Prize-winning discovery

The discovery of hepatitis C viruses earned Harvey Alter, Michael Houghton, and Charles Rice the 2020 Nobel Prize in Physiology or Medicine [8]. Their findings are hailed as a scientific milestone in the ongoing battle against viral diseases (Figure 1) [8]. Owing to their work, highly specific and sensitive diagnostic blood tests have been developed, which have been instrumental in eliminating post-transfusion hepatitis in numerous countries worldwide [20].

An average of three to four million people gets infected with the hepatitis C virus annually, rendering them susceptible to cirrhosis, liver failure,

and hepatocellular carcinoma [21]. The novel discoveries provided a much-needed foundation with which efficient antiviral drugs have been developed [14]. These antiviral drugs are well-tolerated and are able to cure the disease; thus, these drugs provide the opportunity to save lives, reduce healthcare costs, and rid the world of the hepatitis C virus [14]. However, to reach this goal, collaborative international efforts are needed to advance widespread blood testing, to produce antiviral drugs at a large scale, and to distribute them across the globe.

Although the World Health Organization aims for the eradication of the hepatitis C virus by 2030, a progress report in 2020 indicated that merely one in five people with hepatitis C is diagnosed and even less receive treatment and are cured [22]. Lack of adequate financing remains the sole barrier to achieve widespread testing and treatment [22]. Therefore, the commitment of philanthropic funders and investments from public-private partnerships with industry are urgently needed to finish the work started by the Nobel Prize winners [22]. Increased financing can also aid in catalysing certain developing countries to re-assess and improve their health care programs to eliminate hepatitis C virus circulation [22]. Current large-scale investments and extensive international collaboration for COVID-19 suggests that it should be possible to mobilise resources to rid the world of the hepatitis C virus [23].

Acknowledgements

RAMS would like to thank Ronald P. van Rij, professor and head of the Laboratory of Experimental Virology at the Department of Medical Microbiology from the Radboudumc and Daphne Olischläger, BSc, for providing the author with feedback. Also, special thanks to Harshitha Ramu's classmate Arbaaz, BSc, for editing the manuscript before submission.

References

1. Floreani, A.*, et al.* Etiopathogenesis of autoimmune hepatitis. *J Autoimmun* **95**, 133-143 (2018).
2. Jamal, M.M.*, et al.* Alcohol and hepatitis C. *Dig Dis* **23**, 285-296 (2005).
3. Feinstone, S.M. History of the Discovery of Hepatitis A Virus. *Cold Spring Harb Perspect Med* **9**(2019).
4. Block, T.M.*, et al.* A historical perspective on the discovery and elucidation of the hepatitis B virus. *Antiviral Res* **131**, 109-123 (2016).
5. Thuener, J. Hepatitis A and B Infections. *Prim Care* **44**, 621-629 (2017).
6. Liu, Z.*, et al.* Disease burden of viral hepatitis A, B, C and E: A systematic analysis. *J Viral Hepat* **27**, 1284-1296 (2020).
7. Alter, H.J.*, et al.* Posttransfusion hepatitis after exclusion of commercial and hepatitis-B antigen-positive donors. *Ann Intern Med* **77**, 691-699 (1972).
8. Baumert, T.F. The Nobel Prize in Medicine 2020 for the Discovery of Hepatitis C Virus: Transforming Hepatology. *J Hepatol* **73**, 1303-1305 (2020).
9. Blumberg, B.S.*, et al.* A "NEW" ANTIGEN IN LEUKEMIA SERA. *Jama* **191**, 541-546 (1965).
10. Blumberg, B.S.*, et al.* Hepatitis and leukemia: their relation to Australia antigen. *Bull N Y Acad Med* **44**, 1566-1586 (1968).
11. Blumberg, B.S. & Millman, I. Vaccine against viral hepatitis and process. (Google Patents, 1972).
12. Alter, H.*, et al.* Hepatitis-associated antigen to test or not to test? *The Lancet* **296**, 142-143 (1970).
13. Bradley, D.W.*, et al.* Posttransfusion non-A, non-B hepatitis in chimpanzees. Physicochemical evidence that the tubule-forming agent is a small, enveloped virus. *Gastroenterology* **88**, 773-779 (1985).
14. Trepo, C. A brief history of hepatitis milestones. *Liver International* **34**, 29-37 (2014).
15. Choo, Q.L.*, et al.* Isolation of a cDNA clone derived from a blood-borne non-A, non-B viral hepatitis genome. *Science* **244**, 359-362 (1989).
16. Kuo, G.*, et al.* An assay for circulating antibodies to a major etiologic virus of human non-A, non-B hepatitis. *Science* **244**, 362-364 (1989).
17. Choo, Q.L.*, et al.* Hepatitis C virus: the major causative agent of viral non-A, non-B hepatitis. *Br Med Bull* **46**, 423-441 (1990).
18. Miyamura, T.*, et al.* Detection of antibody against antigen expressed by molecularly cloned hepatitis C virus cDNA: application to diagnosis and blood screening for posttransfusion hepatitis. *Proc Natl Acad Sci U S A* **87**, 983-987 (1990).
19. Kolykhalov, A.A.*, et al.* Transmission of Hepatitis C by Intrahepatic Inoculation with Transcribed RNA. *Science* **277**, 570-574 (1997).
20. Ward, J.W.*, et al.* Time for the Elimination of Hepatitis C Virus as a Global Health Threat. *The Liver: Biology and Pathobiology*, 935-952 (2020).
21. Jafri, S.-M. & Gordon, S.C. Epidemiology of Hepatitis C. *Clin Liver Dis (Hoboken)* **12**, 140-142 (2018).
22. Ward, J.W. The Nobel Prize for discovery of HCV is a call to end hepatitis. *The Lancet* **396**, 1733 (2020).
23. Kinsella, C.M.*, et al.* Preparedness needs research: How fundamental science and international collaboration accelerated the response to COVID-19. *PLoS Pathog* **16**, e1008902 (2020).