# MOLECULAR EPIDEMIOLOGY OF FULL-LENGTH, EARLY INFECTION, HEPATITIS C VIRUS GENOMES AMONG INJECTING DRUG USERS WITH RECENT INFECTION: THE InC3 STUDY

Rodrigo C[1], Eltahla A[1], Bull RA[1], Luciani F[1], Grebely J[2], Dore GJ[2], Applegate T[2], Page K[3], Bruneau J[4], Morris MD[5], Cox AL[6], Osburn W[6], Kim AY[7], Shoukry NH[4], Lauer GM[7], Maher L[2], Hellard M[9,10,11], Schinkel J[8,12] , Prins M[8,12]  and Lloyd AR[1] on behalf of the InC[3] Study Group

[1]School of Medical Sciences, Faculty of Medicine, UNSW Australia, Sydney, NSW, Australia, [2]The Kirby Institute, UNSW Australia , Sydney, NSW, Australia, [3]Department of Internal Medicine, University of New Mexico, Albuquerque, NM, USA, [4]CRCHUM, Université de Montréal, Montreal, QC, Canada, [5]Department of Epidemiology and Biostatistics, University of California, San Francisco, San Francisco, CA, USA, [6]Department of Medicine, Johns Hopkins Medical Institutions, Baltimore, MD, USA, [7]Harvard Medical School, Boston, MA, USA, [8]Department of Internal Medicine, Division of Infectious Diseases, Tropical Medicine and AIDS, Center for Infection and Immunity Amsterdam, Academic Medical Center, Meibergdreef, Amsterdam, The Netherlands, [9]Burnet Institute, Melbourne, VIC, Australia, [10]Alfred Hospital, Department of Infectious Diseases, Melbourne, Australia, [11]Department of Epidemiology and Preventive Medicine, Monash University, Australia, [12]GGD Public Health Service of Amsterdam, Amsterdam, The Netherlands

**Background:** The molecular epidemiology of hepatitis C virus (HCV) infection has typically been studied in individual countries worldwide, however little is known about cross-continental patterns and about the characteristics of the viruses associated with incident infection.

**Methods:** The International collaborative of Incident HIV and Hepatitis C in Injecting Cohorts (InC3) is a collaboration of nine well characterized prospective cohorts on injecting drug users. The sample repository of InC3 collaboration was searched to find all samples within 180 days of infection.An optimized laboratory pipeline was used to generate full-length HCV sequences by next generation sequencing. The output of was analysed with a bioinformatics pipeline to generate consensus sequences and these were used in phylogenetic analysis to understand the cross-continental molecular epidemiology of early HCV infection and the demographic / behavioural associations with clustering.

**Results:** The cohorts were from three continents: North America (n=4), Australia (n=4) and Europe (n=1). One hundred and ninety full-length HCV genomes were amplified. Genomes from the most common subtypes (1a: n=94, 2b: n=15 and 3a: n=68) were used in maximum likelihood phylogenetic analysis to identify clusters of molecularly related sequences. Using full genome trees, 78 sequences (44%) were found to lie within 29 phylogenetic clusters/pairs (mean patristic distance cut-off; 0.01%, 95% bootstrap support). Of these, 13 each had exclusively Australian and North American sequences.  Older age, non-Caucasian ethnicity, Australian origin, and current imprisonment were significantly associated with clustering in bivariate analysis, and the first two associations remained statistically significant after binary logistic regression analysis.

**Conclusions:** We have established the largest, international, acute HCV genomic data repository available to-date worldwide. In the phylogenetic analysis of these sequences clustering was evident almost exclusively within-continents. This suggests that HCV evolves in micro-epidemics within geographically isolated communities and this can have implicatons for a universal vaccine.

**Disclosure statement:** The authors have no conflicts of interest to declare