# The field performance of hepatitis E diagnostics during an outbreak in Bentiu, South Sudan

A. Koyuncu[1,2], R. Nesbitt[2], C. Alvarez[3], K. Vincent Asilaza[4], J. Wamala[5], M. Haile[6], E. Gignoux[2,6], M. Albela[6], E. Gurley[1], F. Beden Loro[5], D. Biem[7], M. Rull[6], J. Rumunu[7], I. Ciglenecki[6], I. Eckerle[3], A. Azman [1,3,6]

[1]Johns Hopkins University, Baltimore, MD, USA; [2]Epicentre, Paris, France; [3]Geneva Centre for Emerging Viral Diseases, Geneva University Hospitals, Geneva, Switzerland; [4]Médecins Sans Frontières (MSF), Bentiu, South Sudan; [5]World Health Organization, Juba, South Sudan; [6]MSF, Geneva, Switzerland; [7]Ministry of Health, Juba, South Sudan

## Introduction

- Our understanding of hepatitis E epidemiology and burden has been hindered by the lack of routine identification and lab testing of suspected cases.
- PCR and ELISA tests for IgM antibodies are considered the gold standards used to confirm hepatitis E, but they require advanced lab capacity not available in communities most affected by hepatitis E.
- Rapid diagnostic tests (RDT) could be a useful surveillance tool, but their field performance has not been well characterized

## Objective

To evaluate the performance of PCR, IgM ELISA, and Assure IgM RDT during a hepatitis E virus (HEV) genotype 1 outbreak and assess the duration of viremia and antibody response

## Methods

- We used data from enhanced clinical hepatitis E surveillance (March - December 2022) in a health facility in Bentiu internally displaced persons camp
- All individuals with acute jaundice syndrome were considered suspected cases
- All suspected cases were asked a series of questions and tested with PCR (venous plasma) , IgM ELISA (venous plasma) and an IgM rapid test (capillary blood) (Assure, Genelabs Diagnostics).
- PCR and ELISA testing was conducted in a reference laboratory in Geneva.
- We used latent class models to estimate the sensitivity and specificity of each diagnostic test for confirming HEV.
- We explored how the sensitivity of each test varied as a function of time since jaundice onset, age and sex
- We used accelerated failure time models to estimate the median time to a PCR and ELISA negative test among those positive at baseline.

## Results

- Among 893 suspected hepatitis E cases (Table 1), 180 had viral RNA detected by PCR (20%), 245 had IgM antibodies detected by RDT (27%), and 236 had IgM antibodies detected by ELISA (26%). 363 had follow-up serum samples collected.
- The median time from first PCR positive result to a negative PCR result was 19 days (95% CI: 17, 21) with no differences by age or sex. (Figure 2)
- Individuals under 5 years old had a faster IgM antibody decay (median: 62 days; 95% CI: 39, 90) compared to over 5 (median: 126; 95% CI: 93, 185)

*Among suspected cases with careseeking delays up to 30 days:*

- The average positive predictive value for PCR was 85.7% (95% CI: 37.0, 98.1), IgM RDT was 75.2% (95% CI: 20.2, 95.1), and IgM ELISA was 76.2% (95% CI: 21.3, 95.4).
- The average negative predictive value for PCR was 91.6% (95% CI: 67.0, 99.7), for IgM RDT was 95.0% (95% CI: 83.5, 99.8), and for IgM ELISA was 98.2% (95% CI: 93.5, 99.9).
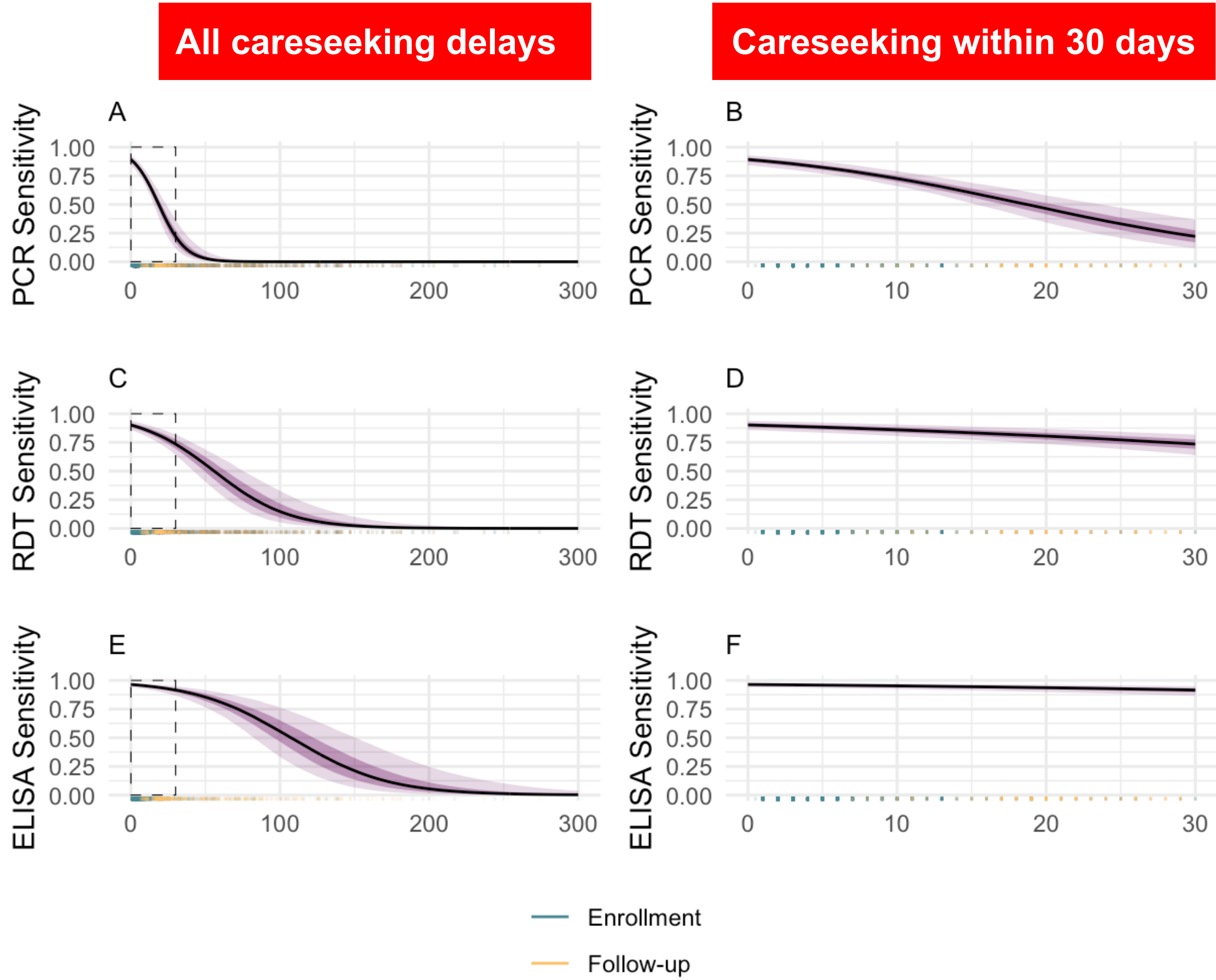


Figure 1. Sensitivity and specificity of diagnostic tests for detecting hepatitis E infections. Panel A, C, and E show sensitivity across all careseeking delays with a box around the first 30 days between jaundice onset and careseeking. Panels B, D, and F show sensitivity in the first 30 days between jaundice onset and careseeking.

## Conclusions

- Diagnostic tests for hepatitis E had high specificity in field conditions, though sensitivity decreased with delayed careseeking.
- Careseeking delays can greatly influence diagnostic test interpretation
- The Assure RDT had higher sensitivity than PCR and similar specificity to ELISA, underscoring its potential utility for surveillance and outbreak detection

Table 1. Characteristics of suspected hepatitis E cases who presented to care at a health facility between March and December 2022, Bentiu. Pan positive = PCR, RDT, and ELISA+, Pan negative: PCR, RDT, and ELISA-

| Characteristic N (col %) | Overall N=893 | Case type | | |
| --- | --- | --- | --- | --- |
| | | Pan-Positive N=169 | Mixed N=107 | Pan-Negative N=617 |
| Female | 406 (46) | 77 (46) | 48 (45) | 281 (46) |
| **Age** | | | | |
| 0-5 | 192 (22) | 44 (26) | 26 (24) | 122 (20) |
| 6-15 | 241 (27) | 69 (41) | 34 (32) | 138 (22) |
| 16-39 | 379 (42) | 50 (30) | 38 (36) | 291 (47) |
| 40+ | 81 (9) | 6 (4) | 9 (8) | 66 (11) |
| **Days since jaundice onset** | | | | |
| ≤1 week | 602 (67) | 132 (78) | 64 (60) | 406 (66) |
| >1 week to 2 weeks | 129 (14) | 30 (18) | 15 (14) | 84 (14) |
| >2 weeks to 1 month | 74 (8) | 6 (4) | 10 (9) | 58 (9) |
| >1 month to 2 months | 49 (6) | 1 (1) | 9 (8) | 39 (6) |
| >2 months | 38 (4) | 0 (0) | 9 (8) | 29 (5) |
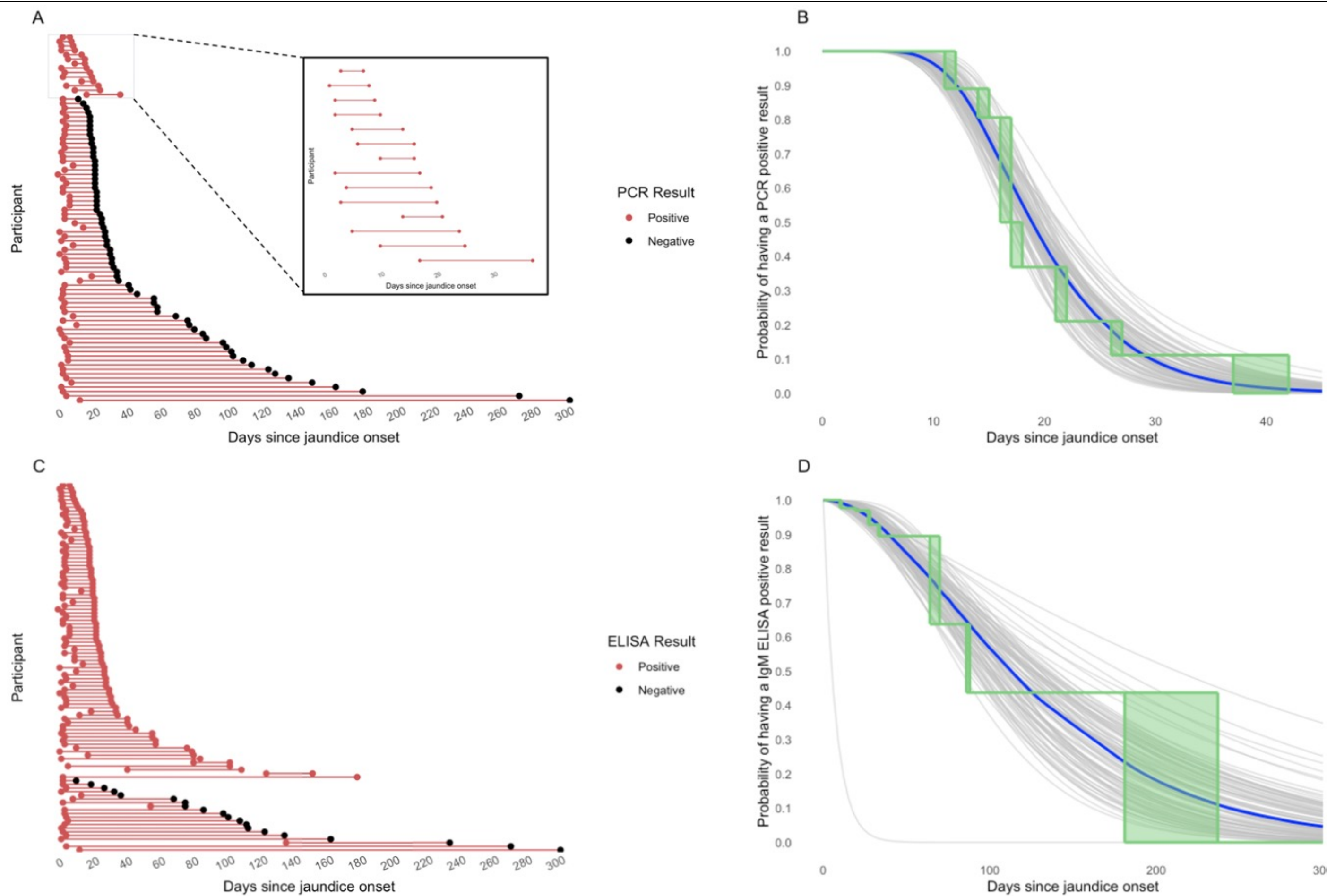


Figure 2. Longitudinal PCR (A & B) and IgM ELISA (C & D) results among suspected hepatitis E cases with a positive PCR and/or IgM ELISA result at enrollment. A & C: Cohort plot of participants who were PCR positive (A) or ELISA positive (C) at enrollment with a follow-up test result. B & D: Parametric and non-parametric estimates of the probability of a positive PCR (B) or IgM ELISA (D) result at follow-up. Rectangular regions represent estimates with similar likelihood in the non-parametric survival curve.

Table 2. Sensitivity and specificity of diagnostic tests for detecting true hepatitis E infections based on latent class analysis

| Test | Performance metric (95% Credible interval) | |
| --- | --- | --- |
| | Sensitivity: Careseeking within 30 days of AJS onset | Specificity |
| PCR | 73.4% (27.2, 90.0) | 98.2% (97.6, 98.7) |
| IgM RDT | 86.2% (74.0, 92.5) | 94.7% (93.4, 95.9) |
| IgM ELISA | 95.2% (90.9, 97.7) | 94.6% (93.0, 95.8) |

MEDECINS SANS FRONTIERES

English Charity Reg. No. 1026588