

## Supplemental Material

### Glossary of Terms

AF: Amniotic fluid

CSF: Cerebrospinal fluid

$C_T$ : Cycle threshold value derived from a real-time polymerase chain

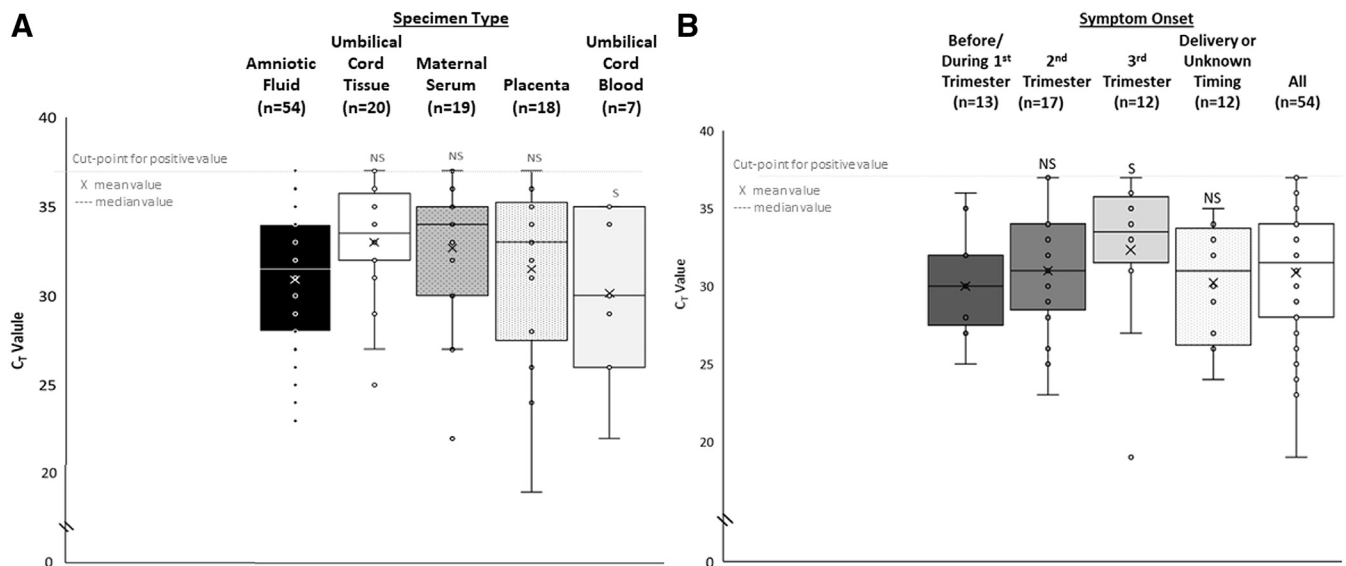
reaction; cycle threshold values are inversely proportional to viral load, with lower values representing a higher viral load

INS: Instituto Nacional de Salud, Colombia's National Institute of Health

rRT-PCR: Real-time reverse transcriptase-polymerase chain reaction  
 ZIKV: Zika virus (ZIKV+ denotes rRT-PCR positive Zika virus test result; ZIKV- denotes rRT-PCR negative Zika virus test result)

### SUPPLEMENTAL FIGURE 1

#### Box plots of cycle threshold values



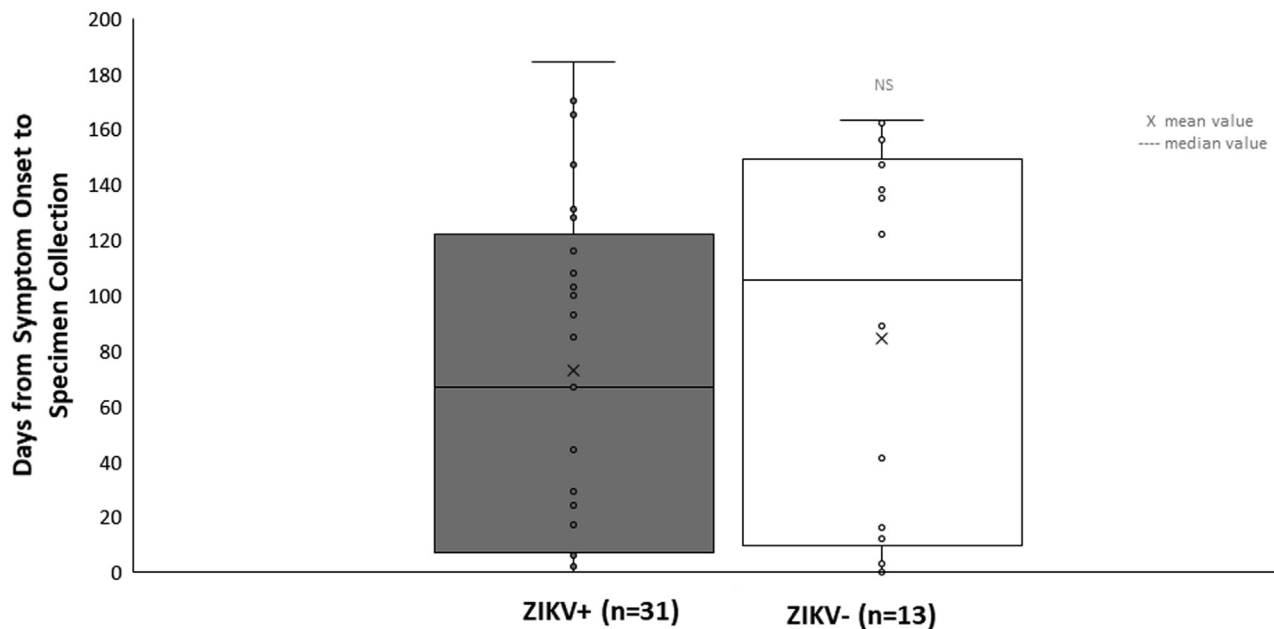
Box plots of **A**, cycle threshold values for Zika virus real-time reverse transcriptase-polymerase chain reaction-positive specimens by specimen type ( $n=118$  specimens<sup>1</sup>) and **B**, trimester of symptom onset, for amniotic fluid specimens only ( $n=54$ <sup>2</sup>). *Superscript 1* indicates missing cycle threshold values for the following ZIKV+ specimens: 6 amniotic fluid, 9 umbilical cord tissue, 3 maternal serum, 9 placenta, and 4 umbilical cord blood. *Superscript 2* indicates missing cycle threshold values for 6 ZIKV+ specimens.

$C_T$ , cycle threshold; NS, no significant difference compared with amniotic fluid specimens (**A**) or before/during first-trimester symptom onset (**B**) as Kruskal-Wallis probability value  $>.05$ ; PCR, polymerase chain reaction; S, significantly different compared with amniotic fluid specimens (**A**) or before/during first-trimester symptom onset (**B**) as Kruskal-Wallis probability value  $<.05$ ; ZIKV+, Zika virus real-time reverse transcriptase-polymerase chain reaction-positive specimens.

Mercedo et al. Zika virus in amniotic fluid. *Am J Obstet Gynecol* 2020.

## SUPPLEMENTAL FIGURE 2

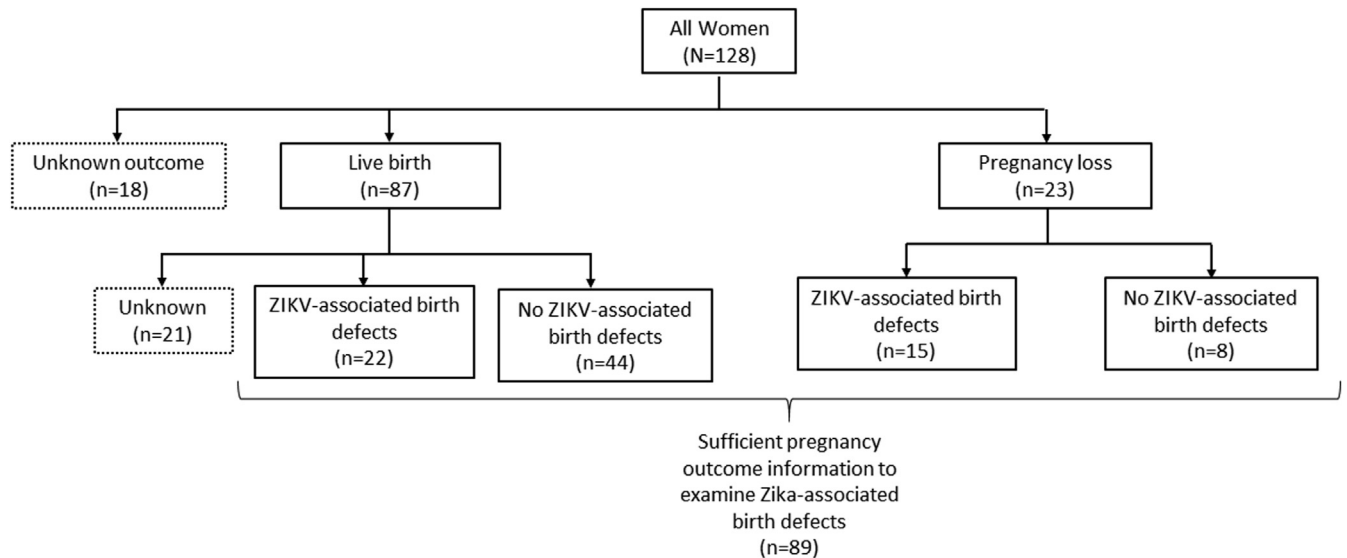
## Time from symptom onset to collection of prenatal amniotic fluid specimen



Time from symptom onset to collection of prenatal amniotic fluid specimen, among women with at least one positive Zika virus real-time reverse transcription virus real-time reverse transcriptase—polymerase chain reaction assay result (n=44). Among 75 women with a prenatal amniotic fluid specimen collected, 44 women had ZIKV symptom onset trimester information available and at least 1 specimen of any type that tested positive for ZIKV by real-time reverse transcriptase—polymerase chain reaction.

NS, Not significant when compared with ZIKV+ specimens (Kruskal-Wallis probability value >.05); PCR, polymerase chain reaction; ZIKV-, Zika virus real-time reverse transcription virus real-time reverse transcriptase—polymerase chain reaction negative specimens; ZIKV+, Zika virus real-time reverse transcription virus real-time reverse transcriptase—polymerase chain reaction positive specimens.

Mercado et al. Zika virus in amniotic fluid. Am J Obstet Gynecol 2020.

**SUPPLEMENTAL FIGURE 3**  
**Frequency of Zika virus-associated birth defects<sup>1</sup>**


Prevalence of Zika virus-associated birth defects<sup>1</sup> among women with amniotic fluid specimens (N=128). *Superscript 1* indicates Zika virus—associated birth defects that are defined as brain abnormalities (with or without microcephaly) or eye abnormalities identified prenatally (for pregnancy losses) or were confirmed postnatally (for live births).

ZIKV, Zika virus.

Mercado et al. Zika virus in amniotic fluid. *Am J Obstet Gynecol* 2020.