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## SARS-COV-2 VIRUS NOT FOUND IN ANIMAL SAMPLES FROM HUANAN SEAFOOD MARKET, SAYS STUDY

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File photo of the Wuhan Huanan Wholesale Seafood Market. | Photo Credit: AP

An analysis of environmental and animal samples for SARS-CoV-2 in the Huanan seafood market, Wuhan, China, in early 2020, detected the virus in environmental swabs but not in samples taken from animals, according to a study published in *Nature* on April 5. Further analysis of the genomic material in both virus-positive and virus-negative environmental samples revealed that a wide range of animals were present at the market. This included the raccoon dogs, which have been hypothesised to be potential intermediate hosts for the virus. However, the study did not find any animal species to be infected. The study has thus not been able to determine the origin of the virus.

George F. Gao and colleagues from China's Center for Disease Control and Prevention (CDCP), who carried out the study published in *Nature*, did find the virus in the environmental samples in the Huanan seafood market during the early stages of the pandemic. But no animal samples (raccoon dogs included) tested positive and they have not been able to provide evidence of potential spillover of the virus from animals to humans.

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"The environmental samples cannot prove that the animals were infected. Furthermore, even if the animals were infected, our study does not rule out that human-to-animal transmission occurred, considering the sampling time was after the human infection within the market as reported retrospectively. Thus, the possibility of potential introduction of the virus to the market through infected humans, or cold chain products, cannot be ruled out yet," they write.

The absence of the virus in samples collected from raccoon dogs and the inability of the study to establish the origin of the virus is in complete variance to the conclusions made by a team led by Michael Worobey, an evolutionary biologist at the University of Arizona, in a report posted in a public repository on March 20. Dr. Worobey's team had used the genome data posted in the Global Initiative on Sharing Avian Influenza Data (GISAID) by the researchers of the April 5 paper in *Nature*.

Since the clusters of early human cases in December 2019 was associated with the market, the researchers collected samples from the environment and animals (unsold goods found in

refrigerators and freezers, swabs from stray animals and their faeces) in and around the market from January 1 to March 30, 2020. In all, 1,380 samples were collected (923 environmental and 457 samples from 18 animal species). The virus was detected using RT-PCR in 73 environmental samples, but none of the animal samples, as per the study. Three live viruses were successfully isolated. The viruses from the market had 99.99-100% similarity with the human isolates.

The market is separated into two zones, the east and west zones, with seafood and animals mainly sold in the west zone and livestock meat in the east zone. Among the 828 samples collected from inside the market, 64 samples (7.7%) were positive for the virus. Of the 64 positive samples, 56 were collected from the market's west zone. Among the 14 samples from warehouses related to the market, five tested positive. "This may reflect the nature of SARS-CoV-2 presence in the market during the early phase of the outbreak," they write. Of the 51 samples collected from sewerage wells surrounding the market, three tested positive; all four samples from sewerage wells inside the market tested positive. The sampling was "biased" as shops in the west zone selling wildlife and those linked to early cases were prioritised for sampling.

As per the report, wastewater in the overground drainage led into the underground drainage inside the market and then flowed into the wells on the edge of the market. The virus in the overground drainage could be from infected humans and potential animal waste, they note. "These data suggest that either infected people and/or animals in the market contaminated the sewage or that the contaminated sewage may have further played a role in furthering the virus transmission within the case cluster in the market," they write.

The China CDCP researchers were able to isolate live viruses from three environmental samples (with CT values less than 30) collected from the ground and wall. Two of these samples were from the stalls with confirmed patients, as per the report. "Successful virus isolation from the original samples with low CT values revealed the existence of live SARS-CoV-2 with high titers in the environment of the HSM [Huanan seafood market]," they write. They obtained seven complete or near complete SARS-CoV-2 genome sequences (three sequences from three environmental samples and four sequences from cell supernatants).

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