Post-mortem COVID-19 Positive Testing: Institute of Forensic Sciences Experience

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Objective: The main objective was to present the experience of the Institute of Forensic Sciences of Puerto Rico in facing the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), also known as COVID-19. It has been found that some COVID-19 positive cases may continue to show post-mortem positive results for up to 49 days.

Methods: The *in vitro* technique of ID NOW COVID-19 was used in the analysis to evaluate the presence of SARS-Cov-2 in postmortem forensic cases. This isothermal method allows to amplify and identify the presence of the RNA-dependent RNA polymerase viral segment. Information on demographics, comorbidities, and the manner and cause of death was collected.

Results: A total of 612 subjects were sampled, of which 41 (6.7%) tested positive for COVID-19;14 (34.1%) of those subjects remained positive for more than 7 days Postmortem. Of the 41 positive cases, only 3 (7.3%) had been diagnosed with COVID-19 before their demise. The most common comorbidities were hypertension (36%), obesity (29%), and mental health conditions (50%).

Conclusion: Results from postmortem COVID-19 testing revealed that some cadavers remain COVID-19 positive for a longer period than expected. Despite this, based on the information collected from the cases that were tested more than once, there is no direct correlation between the cause of death and persistent COVID-19 positivity. We recommend that additional investigations be carried out, in which investigations viral load and the maximum time of the infectious phase are specifically evaluated. [P R Health Sci J 2022;41(4):197-201]

Key words: COVID-19, SARS-CoV-2, PCR, Post-mortem, Forensic pathology

n December 2019, an outbreak of a disease caused by severe acute respiratory syndrome (SARS)-coronavirus-2 (CoV-2) was reported in Wuhan, China (1). Due to its contagious capacity and rapid spread, by the end of January 2020 there were already confirmed cases in Europe, the United States, and Canada. As of the date of this publication, there have been more than 15 million infected patients in the United States and over 250,000 deaths. The arrival of coronavirus disease 2019 (COVID-19) has had major health, social, and economic impacts on all the nations of the world. The viral infection is mainly characterized by major symptoms such as fever, cough, muscle pain, shortness of breath, and diarrhea, among others, which can be classified as mild, moderate, and severe. Also, there is a group of patients that appear to be asymptomatic.

Puerto Rico has also become part of the pandemic, by October 31, 2020, we had 66,128 probable and confirmed COVID-19 cases and 822 deaths. The Institute of Forensic

Sciences (IFS) of Puerto Rico has received a fair number of suspected COVID-19 cases.

Usually, about 6,000 to 7,000 cadavers a year are referred for forensic evaluation and investigation. Not all the cadavers are autopsied. This pandemic year has changed how we manage and study these cases. Since COVID-19 is highly contagious, the pathologists at the institute suggested that the autopsies should be delayed until the cadavers that tested positive turned negative for COVID-19. It turned out that some of the corpses

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The authors have no conflict of interest to disclose.

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that were positive for COVID-19 when tested postmortem had been positive for more than 1 week (1,2,3).

This paper presents a series of cadavers with prolonged (7 days or more) positivity for COVID-19, which were tested using the isothermal nucleic amplification of COVID-19 at the IFS of Puerto Rico. Of the sampled subjects only 6.7% were positive for COVID-19 and of those that were positive 34.1% remained positive for an average of 44 days (Table 2).

Institute of Forensic Sciences of Puerto Rico

The IFS is a governmental agency that is composed of the following divisions: Medicolegal Investigation, Criminalistics Laboratory, Drug Testing Laboratory, Crime Scene Investigation, and Vehicle Analysis.

The Medicolegal Investigation Division recently added its own onsite COVID-19 laboratory for postmortem and live testing. The COVID-19 laboratory staff includes nurses, medical technologists, and an occupational health and safety officer. Prior to July 27, 2020, pathologists would sample the subjects suspected to have been infected with COVID-19 and send the swabs offsite to the Puerto Rico Department of Health laboratory for analysis. Subsequently the IFS became responsible for sampling and analyzing the specimens. The IFS is under the jurisdiction of the United States, government and must follow the guidelines established by the Center for Disease Control and Prevention (CDC) (4,5).

COVID-19 Laboratory

The presence of SARS-CoV-2 RNA was determined with the ID NOW COVID-19 assay, which is an in vitro diagnostic test. This test utilizes isothermal nucleic acid amplification technology for the qualitative detection of nucleic acid from the SARS-CoV-2 viral RNA. The test detects the virus by amplifying a unique RNA-dependent RNA polymerase segment.

The samples to be tested can be acquired by swabbing the nasal, nasopharyngeal or throat areas of suspected COVID-19 patients. Primarily, the IFS used nasopharyngeal swabs, which were collected by trained healthcare professionals.

The samples from both live and dead subjects were collected using a sterile swab without viral transport media and were stored at 2 tp 30°C in the COVID-19 laboratory of the IFS. Each sample was analyzed for SARS-CoV-2 RNA within 2hours after having been taken.

Medicolegal Investigation Division: Working During the COVID-19 Pandemic

Many procedures have changed since the COVID-19 pandemic arrived. The bodies received at the morgue are very carefully handled and stored. The bodies are kept in their pouches until a COVID-19 reverse transcriptase-polymerase chain reaction (RT-PCR) test is performed and the sample processed. Positive cases are sampled weekly until they become negative. This is done, as well, for mandatory autopsies, are performed for homicides, traffic accidents, and other cases that may require this kind of study. Some cases that remained positive were autopsied to minimize the loss of evidence.

The positive forensic cases that are to be autopsied are taken to an isolated, environmentally controlled negative pressure room. The space between the autopsy tables is 6 feet, and only the pathologist, the pathology assistant, and a floating assistant are allowed in that room. They all have the required equipment for working with contaminated cadavers, as mandated by the Occupational Safety, and Health Administration, National Institutes of Health, and the CDC. Before and after the autopsy procedure, the isolation room is treated with detergents, bleach, and alcohol-based sanitizers for decontamination purposes.

In accordance with the CDC guidelines, the protective personal equipment (PPE) utilized by the pathology staff

Table 1. Demographics, comorbidities, number of days of positiveness, cause, and manner of death.

	Age	Sex (female /male)	COVID-19 test, premortem (positive, negative, not done, unknown)	Length of positivity, postmortem (days)	Comorbidities					Place	Cause/
					Obesity (yes/no)	Diabetes mellitus (yes/no)	Hypertension (yes/no)	Chronic kidney disease (yes/no)	Mental health (yes/no)	of death	manner of death
1	28	М	Negative	49	Yes					Street	Gunshot/homicide
2	80	M	Not done	42					Yes	Home	Gunshot/suicide
3	23	M	Negative	25						Hospital	Gunshot/homicide
4	47	M	Unknown	44			Yes		Yes	Street	Gunshot/homicide
5	48	F	Unknown	43					Yes	Home	Overdose/accidental
6	58	M	Unknown	43					Yes	Street	Pedestrian/accidental
7	58	M	Unknown	27	Yes	Yes	Yes		Yes	Home	Asphyxia/suicide
8	50	M	Unknown	7						Street	Overdose/accidental
9	90	F	Not done	17			Yes		Yes	Asylum	COVID/natural
10	103	F	Positive	12			Yes		Yes	Hospital	COVID/ natural
11	84	F	Not done	20	Yes	Yes	Yes	Yes		Hospital	Cerebral Infarction/ natural
12	25	M	Positive	13	Yes					Hospital	Burns/accidental
13	26	M	Positive	15						Car	Gunshot/homicide
14	38	F	Negative	8						Street	Gunshot/homicide

Table 2. Percentages of sex, age, days positive for COVID-19, comorbidities, and place and manner of death.

Characteristics	n (%)
Age	54 years, average (36%)
Sex	
Female	5 females (36%)
Male	9 males (64%)
Number of days positive for COVID-19	44 days, average (36%)
Comorbidity	
Obesity	4 (29 %)
Diabetes mellitus	2 (13 %)
Hypertension	5 (36 %)
Chronic kidney disease	1 (7 %)
Mental health issues(s)	7 (50 %)
Place of death	
Street	5 (36%)
Home	3 (21%)
Hospital	4 (29%)
Car	1 (7%)
Asylum	1 (7%)
Manner of death	
Homicide	5 (36%)
Suicide	2 (13%)
Accidental	4 (29%)
Natural	3 (21%)

includes a surgical scrub suit, a surgical cap, an impervious gown with full sleeve coverage, eye, and face protection (face shield), shoe covers, double surgical gloves, and an N95 or N100 respirator (6,7,8).

The staff of the pathology division are tested once a month for COVID-19: those who worked with a positive COVID-19 case are tested 5 days after the date of the possible exposure. No employees in the Medicolegal Investigation Division of the IFS have tested positive for COVID-19.

Methods

This study was carried out by the Medicolegal Investigation Division with the collaboration of the COVID-19 laboratory, from March 26, 2020, through October 31, 2020. A variety of subjects were sampled, which samples were analyzed with RT-PCR and the ID NOW COVID-19 methodology.

In March 2020, the IFS began testing for COVID-19 before performing an autopsy, as a preventive measure to minimize exposure of the staff to the virus. There are no published references regarding how long a cadaver can be positive for COVID-19 or whether, if positive, it is or might be contagious (2). Other jurisdictions that have tested their cadavers for COVID-19 have not followed-up for results to become negative or the length of positivity. Approximately one-third of the tested (RT-PCR) cadavers in our jurisdiction that were positive for COVID-19, were positive for more than 7 days.

Since SARS-CoV-2 RNA is an intracellular virus, when the host dies, it would be expected that the infective virus would dissipate. Positive postmortem COVID-19 cases should become negative in a short period of time. We selected a 7-day period

as the limit that a cadaver should be positive for COVID-19 before our proceeding with a given cadaver's autopsy. We could not find any published references relating to the time interval of COVID-19 positive postmortem cases, or any related information, for that matter.

Subjects

Descriptions of the subjects:

• Subject 1:

This was a 28-year-old male who died of multiple gunshot wounds (homicide) and was negative for COVID-19 prior to death. The body was positive for COVID-19 for 49 days, postmortem. The comorbidity was obesity.

• Subject 2:

This 80-year-old male died of a gunshot wound to the head (suicide). A COVID-19 test was not done prior to his death. He was positive for COVID-19 for 42 days, postmortem. The comorbidity was mental health problems.

• Subject 3:

A 23-year-old male, died of multiple gunshot wounds (homicide). A premortem COVID-19 test was negative. He was positive for COVID-19 for 25 days postmortem. There were no comorbidities.

• Subject 4:

This was a 47-year-old male; he died of multiple gunshot wounds (homicide). Though his premortem COVID-19 status was unknown, his body was positive for COVID-19 for 44 days, postmortem. The comorbidities were hypertension and mental health problems.

• Subject 5:

This 48-year-old female died of drug overdose (accidental). She had unknown COVID-19 status, premortem. She

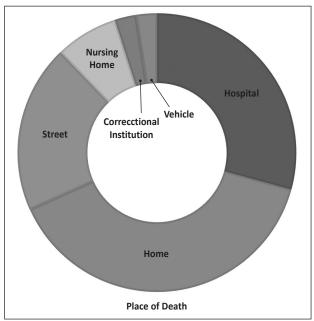


Figure 1. Places of death of subjects positive for COVID-19 for more than 7 days, postmortem.

was positive for COVID-19 for 43 days, postmortem. The comorbidity was mental health problems.

• Subject 6:

This 58-year-old male died in a traffic accident; it is unknown whether a premortem COVID-19 test was done. He was positive for COVID-19 for 43 days postmortem. The comorbidity was mental health problems.

• Subject 7:

The subject was a 58-year-old male who died of asphyxia (suicide). Whether a premortem COVID-19 test was done is unknown. He was positive for COVID-19 for 27 days, postmortem. The comorbidities were obesity, diabetes, hypertension, and mental health problems.

• Subject 8:

This is a 50-year-old male died of drug overdose (accidental). Whether a premortem COVID-19 test was done is unknown. He was positive for COVID-19 for 7 days, postmortem. There were no comorbidities.

• Subject 9:

A 90-year-old female, she died of COVID-19 infection (natural). A premortem COVID-19 test not done. She was positive for COVID-19 for 17 days postmortem. The comorbidities were hypertension and mental health problems.

• Subject 10:

This 103-year-old female she died of COVID-19 infection (natural). Her premortem COVID-19 test was positive. She was positive for 12 days postmortem, for COVID-19. The comorbidities were hypertension and mental health problems.

• Subject 11:

This was a 84 year old female who died of cerebral infarction (natural). A premortem COVID-19 test was not done. She was positive for COVID-19 for 20 days, postmortem. The comorbidities were obesity, diabetes, hypertension, and chronic kidney disease.

• Subject 12:

A 25-year-old male, this subject died of skin burns (accidental). While still hospitalized, his premortem COVID-19 test was positive. He was positive for COVID-19 for 13 days, postmortem. The comorbidity obesity.

• Subject 13:

This 26-year-old male died of multiple gunshot wounds (homicide). His premortem COVID-19 test was positive. He was positive for COVID-19 for 15 days, postmortem. There were no comorbidities.

• Subject 14:

The subject was a 38-year-old female who died of multiple gunshot wounds (homicide). Her premortem test for COVID-19 was negative. She was positive for COVID-19 for 8 days, postmortem. There were no comorbidities.

Results

A total of 612 cadavers were sampled, resulting in 41 positives and 571 negatives for COVID-19. The median age for all the

cases was 56 years and for the positive cases was 64 years. There were 18 (44%) females and 23 (56%) males in the positive group. Of the 41 positive subjects, only 3 (7.3%) were diagnosed with COVID-19 before death. The manners of death for the COVID-19 positive cases were classified as follows: There were 25 natural deaths (due to COVID-19), 7 traffic accidents, 6 homicides and 3 suicides. Most of the subjects died either at home, on the street, or at the hospital.

Of the 41 cadavers positive for COVID-19, 14 (34%) continued being COVID-19 positive for from 7 to 49 days after death (average, 44 days). The median age of this group was 48.5 years. One-third of the cases had a history of arterial hypertension and mental health disorders. We enumerated the subjects from 1 to 14 in chronological order by date of death. Also included were the gender, age, comorbidities, and number of days that they were positive for COVID-19 after death (Table 1). Of the 14 cases shown in Table 1, only 2 (14%) died due to COVID-19 infection; the rest died of other causes, including gunshot wounds and multiple traumas.

Medical comorbidities common to the subjects were arterial hypertension (5; 36%), obesity (4; 29%), diabetes mellitus (2; 13%), mental health related issues (7; 50%), and chronic kidney disease (1; 7%).

Discussion

The IFS is doing commendable work in terms of the COVID-19 testing of cadavers for the safety of all the employees involved in the procedure of receiving, processing, and dispatching the bodies.

Cadavers positive for COVID-19, and that remained positive in serial testing for 7 days or more, have not previously been studied; nor have data on this subject been published. Mainly the published articles focus on the pre and postmortem clinical findings, not inquiring about prolonged positiveness in cadavers. The IFS is the first entity to describe this phenomenon.

We recommend that further studies be done on viral RNA load in patients that were COVID-19 positive and died due to circumstances other than COVID-19. Our findings also suggest the importance of postmortem testing before autopsy and the implementation of appropriate safety procedures since COVID-19 may still be detectable and the viral infectivity cannot be ruled out (1,2,3,9).

It is important to point out that for the patients who had COVID-19 as an underlying cause of death, autopsies e were not mandatory; they were tested only once, postmortem, to minimize the exposure and possible contamination of the Medicolegal Investigation Division personnel.

The information collected from the subjects that were tested more than once for COVID19 shows the diversity of the characteristics of the cadavers tested. However, neither age, sex, comorbidities, nor cause or manner of death explain the persistence of postmortem COVID-19 positivity.

Resumen

Objetivos: El objetivo principal de este trabajo es presentar la experiencia del Instituto de Ciencias Forenses de Puerto Rico con el Síndrome Respiratorio Severo (SARS-CoV-2), también conocido como COVID-19. Se ha encontrado que alguno de los casos positivo a COVID-19 muestran resultados positivos hasta 49 días después de fallecer. Métodos: La técnica in vitro de ID NOW COVID-19 se utiliza en el análisis para evaluar la presencia de SARS-Cov-2 en casos post mortem medicolegales. Este método isotérmico permite amplificar e identificar la presencia del segmento viral RdRp. Se recopiló información sobre datos demográficos, comorbilidades y causa de muerte de estos cadáveres. Resultados: Un total de 612 cadáveres fueron muestreados para COVID-19, de los cuales 41 (6.7%) dieron positivo y 14 (34.1%) de estos sujetos permanecieron positivo más de 7 días. De los 41 casos positivos, sólo 3 (7.3%) habían sido diagnosticado con COVID-19 antes de su muerte. Las comorbilidades más comunes fueron hipertensión (36%), obesidad (29%) y enfermedad de salud mental (50%). Conclusión: Los resultados de las pruebas COVID-19 post mortem revelaron que algunos cadáveres siguen siendo positivos a COVID-19 durante un período de tiempo más largo de lo esperado. La información recopilada de los casos que se muestrearon más de una vez, no tiene correlación directa entre la causa de la muerte y la persistencia de la positividad a COVID-19. Recomendamos que se lleve a cabo investigaciones adicionales, donde se analice la carga viral de los cadáveres con positividad prolongada.

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