# Supplementary Material

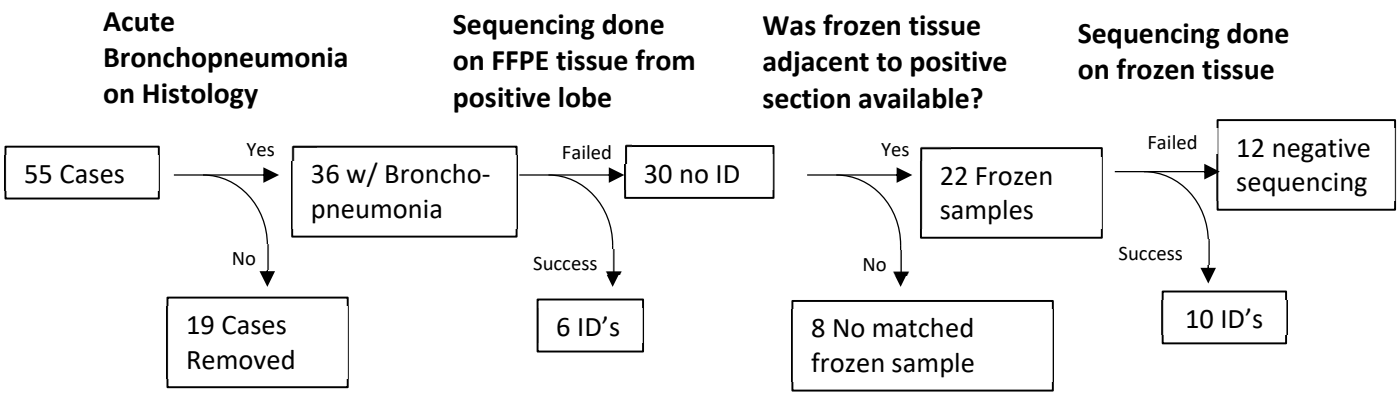
| Patient # | Patient Name | Hospital | Sex | DOB | Age at Death | Race | Ethnicity | BMI | Symptom Onset |
|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | |

| Hospital Admission | Did this patient undergo multiple hospitalizations? | Are outside hospital records available on this patient, either within the hospital system or CRIS via EMR? | Date of Death | Time of Death | Date of Autopsy | Time of Autopsy | Medical History |
|---|---|---|---|---|---|---|---|
| | | | | | | | |

| Surgical History | Enter all documented Home Medications | List any vasopressive medications received | List any antibiotics received | List any systemic steroids received | List any prophylactic anticoagulation | List any system anticoagulation received | List any paralytics received |
|---|---|---|---|---|---|---|---|
| | | | | | | | |

| List any inhaled vasodilators received | List COVID-specific treatments received | Was this patient placed on ECMO? | List any form of Renal Replacement Therapy patient received | Was this Patient Intubated? | Did this patient undergo tracheostomy? | If patient was not intubated, list all forms of respiratory care patient received | Did patient have chest tube(s) placed? |
|---|---|---|---|---|---|---|---|
| | | | | | | | |

**Supplementary Figure S1: Chart review form used to collect demographic and treatment data.**




**Supplementary Figure S2: Flow diagram illustrating sample selection criteria and results of 16S bacterial PCR and sequencing on lung tissue.**

**Supplementary Table S1: Characteristics of autopsy cases separated by site of autopsy.** P values are univariate analyses between sites.

| | NIH (n=44) | UW (n=11) | p value |
|---|---|---|---|
| Median age in years (IQR) | 62.5 (47.25,71) | 63 (53,72) | 0.8659 |
| Female, % | 31.80% | 36.40% | 0.9999 |
| Race, % | | | 0.0968 |
| White | 43.20% | 54.50% | |
| Black or African American | 40.90% | 9.10% | |
| Hispanic Ethnicity, % | 15.90% | 36.40% | 0.9999 |
| Median BMI (IQR) | 30.95 (25,36.9025) | 32.75 (27.3,38.15) | 0.6199 |
| Comorbidities | | | |
| COVID-related (IQR) | 2 (1,3) | 3 (2,4) | 0.0867 |
| Days from onset to death (IQR) | 21 (12,37.5) | 18 (16,22.5) | 0.4442 |
| Hospital duration, days (IQR) | 13 (6,30.25) | 13 (6.5,20.5) | 0.7281 |
| ICU duration, days (IQR) | 11 (4,26.5) | 13 (6,18) | 0.9865 |
| Intubated, % | 79.50% | 100.00% | 0.1787 |
| Intubation duration, days (IQR) | 11 (3,28) | 12 (2,18) | 0.5642 |
| Post-Mortem Interval, hours (IQR) | 22.2 (18.2,33.9) | 72 (36,84) | <0.0001 |
| Presence of Bronchopneumonia, (%) | 65.10% | 63.60% | 0.9999 |

**Supplementary Table S2: Premortem and postmortem pulmonary co-infection results.** Cases where premortem culture results align with postmortem sequencing are in red. To define semi-quantitative abundance in NGS sequencing three synthetic control DNA templates were spiked into reactions at defined copy numbers (2000, 200, and 20), with "major abundance" indicating organism-specific reads exceeding the most abundant synthetic template; "minor abundance" indicating those below the least abundant synthetic template, and "moderate abundance" falling between those levels.

| Patient ID | Premortem pulmonary co-infection diagnosed | Premortem respiratory pathogens cultured | Antibiotics received | Postmortem broncopeumnoia present | B&H stain positive | GMS stain positive | Sequencing Results |
|---|---|---|---|---|---|---|---|
| P1 | | | Linezolid, pipperacillin-tazobactam, ceftriaxone, vancomycin, clindamycin | Yes | Yes | Yes | [Major abundance]: Prevotella melaninogenica<br><br>[Moderate abundance]: Mycoplasma salivarium, Fusobacterium nucleatum<br><br>[Minor abundance]: Prevotella oulorum, Limosilactobacillus (Lactobacillus) fermentum |
| P2 | | | vancomycin, cefepime | | | | [Major abundance]: Pseudomonas aeruginosa<br><br>[Moderate abundance]: Enterococcus faecalis |
| P3 | | | vancomycin | | | | [Major abundance]: Pseudomonas aeruginosa<br><br>[Moderate abundance]: Bacteria of the Family Enterobacteriaceae |
| P4 | | | | | Yes | | [Moderate abundance]: Corynebacterium striatum, Staphylococcus aureus, Streptococcus salivarius group, Rothia mucilaginosa<br><br>[Minor abundance]: Streptococcus parasanguinis, Corynebacterium propinquum, Veillonella parvula, Veillonella species, Granulicatella adiacens, Streptococcus mitis group (not Streptococcus pneumoniae), Streptococcus gordonii, Staphylococcus epidermidis, Streptococcus mutans, Actinomyces gerencseriae, Corynebacterium accolens |
| P5 | Yes | K. pneumoniae, S. marcescens | Ceftriaxone, doxycycline, linezolid, piperacillin-tazobactam, vancomycin, amikacin, meropenem, polymyxin B, micafungin | Yes | | Yes | [Major abundance]: Staphylococcus aureus<br><br>[Minor abundance]: Pseudomonas putida group |
| P6 | | | Piperacillin-tazobactam, vancomycin | Yes | | | Minor abundance]: Mycoplasma salivarium, Fusobacterium nucleatum |
| P7 | | | amikacin, cefepime, meropenem, micafungin, vancomycin | Yes | Yes | | [Major abundance]: Lactobacillus gasseri<br><br>[Moderate abundance]: Prevotella oris, Prevotella scopos<br><br>[Minor abundance]: Prevotella jejuni, Streptococcus anginosus |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| P8 | Yes | MSSA, A. baumanii | amoxicillin, ampicillin-sulbactam, cefazolin, cefepime, ceftriazone, linezolid, meropenem, minocycline, polymixin B | Yes | Yes | | [Minor abundance]: Streptococcus pneumoniae, Neisseria subflava, Porphyromonas pasteri, Fusobacterium periodonticum or Fusobacterium pseudoperiodonticum, Porphyromonas species, Gemella sanguinis, Phocaeicola (Bacteroides) dorei, Haemophilus parainfluenzae, Streptococcus salivarius group |
| P9 | Yes | K. aerogenes, P. aeruginosa | Azithromycin, amikacin, ampicillin, cefazolin, cefapime, fluconazole | Yes | Yes | | [Moderate abundance]: Prevotella oris<br><br>[Minor abundance]: Prevotella jejuni, Veillonella species, Veillonella atypica, Prevotella melaninogenica, Staphylococcus aureus, Lactobacillus gasseri |
| P10 | | | none | Yes | Yes | | [Moderate abundance]: Bacteria of the Order Bacteroidales, Bacteria of the Phylum Bacteroidetes<br><br>[Minor abundance]: Prevotella melaninogenica, Prevotella nanceiensis |
| P11 | Yes | P. aeruginosa, E. coli | amikacin, cefepime, daptomycin, meropenem, metronidazole, vancomycin, linezolid, azithromycin, ceftriaxone | Yes | Yes | | |
| P12 | Yes | A. baumanii, MRSA, K. pneumoniae | ampicillin-sulbactam, ceftriaxone, meropenem, minocycline, polymyxin B, linezolid, piperacillin-tazobactam, cefepime, levofloxacin, micafungin, vancomycin | Yes | | | |
| P13 | Yes | K. pneumoniae, P. aeruginosa, K. oxytoca | ceftolozane-tazobactam, linezolid, cefepime, amikacin, ampicillin, meropenem, ceftriaxone | Yes | | | |
| P14 | | | piperacillin/tazobactam, vancomycin, doxycycline | Yes | Yes | | |
| P15 | | | levofloxacin, imipenem/cilastin, ceftriaxone | Yes | Yes | Yes | |
| P16 | | | vancomycin, piperacillin-tazobactam | | Yes | | |
| P17 | Yes | MRSA, MSSA | vancomycin, ceftaroline, daptomycin, piperacillin-tazobactam, micafungin | Yes | Yes | | |

| | | | | | |
|---|---|---|---|---|---|
| P18 | | | aztreonam, vancomycin | Yes | Yes |
| P19 | | | levofloxacin, vancomycin | Yes | |
| P20 | Yes | E. coli, P. aeruginosa | cefepime, ertapenem, meropenem, amikacin, ceftolozane-tazobactam, linezolid, fluconazole, vancomycin | Yes | |
| P21 | | | vancomycin, piperacillin-tazobactam | | |
| P22 | | | none | | |
| P23 | | | ceftriaxone, azithromycin | | Yes |
| P24 | | | ceftazidime, aztreonam, clindamycin, imipenem-cilastin, vancomycin | Yes | Yes |
| P25 | | | azithromycin, ceftriaxone, ampicillin-sulbactam, doxycycline | Yes | Yes |
| P26 | | | ceftriaxone, azithromycin, cefepime, piperacillin-tazobactam, vancomycin, linezolid | | |
| P27 | | | vancomycin, piperacillin-tazobactam | Yes | Yes |
| P28 | | | none | | |
| P29 | | | piperacillin-tazobactam, sulfamethoxazole-trimethoprim, amikacin, amphotericin B, ceftriaxone, fluconazole, micafungin, tobramycin, valgancyclovir, vancomycin, voriconazole | | |
| P30 | | | micafungin, piperacillin-tazobactam, vancomycin | Yes | Yes |
| P31 | | | azithromycin, ceftriaxone, linezolid, piperacillin-tazobactam | Yes | Yes |
| P32 | | | none | | |

| | | | | | | |
|---|---|---|---|---|---|---|
| P33 | | | none | Yes | Yes | |
| P34 | | | vancomycin, piperacillin-tazobactam, clindamycin | | | |
| P35 | Yes | P. aeruginosa | vancomycin, piperaciilin-tazobactam, ceftriaxone | | | |
| P36 | | | piperacillin-tazobactam | Yes | Yes | |
| P37 | | | piperacillin-tazobactam, vancomycin, meropenem, ampicillin-sulbactam | Yes | Yes | |
| P38 | | | azithromycin, cefazolin, meropenem | Yes | Yes | |
| P39 | | | amikacin, cefepime, linezolid, micafungin, piperacillin-tazobactam, voriconazole | Yes | Yes | Yes |
| P40 | Yes | K. pneumoniae | piperacillin-tazobactam, ampicillin-sulbactam, vancomycin, | Yes | | |
| P41 | Yes | MSSA | ampicillin sulbactam, ceftriaxone, doxycycline, | | | |
| P42 | Yes | P. aeruginosa | vancomycin, cefepime, metronidazole, fluconazole, trimethoprim-sulfamethoxazole, ceftriaxone, meropenem, piperacillin-tazobactam, daptomycin, micafungin, amikacin, ceftazidine-avybactam | | | |
| P43 | | | ceftriaxone, azithromycin | Yes | Yes | |
| P44 | Yes | MSSA | ceftriaxone, azithromycin, ampicillin, cefazolin, cefepime, fluconazole, gentamicin, linezolid, meropenem, micafungin, piperacillin-tazobactam, vancomycin | Yes | Yes | Yes |

| | | | | | |
|---|---|---|---|---|---|
| AU-10 | | | none | | |
| AU-13 | | | cefepime, piperacillin-tazobactam, ampicillin | Yes | Yes |
| AU-17 | | | azithromycin, vancomycin, cefepime, metronidazole | Yes | Yes |
| AU-18 | | | vancomycin, ceftazidime | | |
| AU-19 | | | ceftriaxone, doxycycline | Yes | |
| AU-20 | | | vancomycin | Yes | Yes |
| AU-21 | Yes | MRSA | vancomycin, meropenem, doxycycline | Yes | |
| AU-22 | | | vancomycin, cefepime | Yes | |
| AU-29 | | | vancomycin, ceftriaxone | Yes | Yes |
| AU-30 | Yes | E. cloacae, K. pneumoniae, MSSA | vancomycin, meropenem, doxycycline, voriconazole | | |
| AU-31 | | | ceftriaxone, vancomycin | | |

**Supplementary Table 3: Comparison of cases with pulmonary co-infection with and without fungal co-infection.** P values are univariate analyses between groups.

| | Fungal Pulmonary Co-infection (n=5) | Bacterial Only Co-infection (n=31) | p-value |
|---|---|---|---|
| **Median age in years (IQR)** | 41 (27,61) | 63 (58,70.5) | 0.0480 |
| **Female (%)** | 0/5 (0.0%) | 12/31 (38.7%) | 0.1464 |
| **Race (%)** | | | 0.5114 |
| **White (%)** | 3/5 (60.0%) | 14/31 (45.2%) | |
| **Black or African American (%)** | 1/5 (20.0%) | 11/31 (35.5%) | |
| **Hispanic Ethnicity (%)** | 2/5 (40.0%) | 6/31 (19.4%) | 0.3048 |
| **Median BMI (IQR)** | 39.2 (32.0,51.0) | 31.7 (26.3,38.5) | 0.2616 |
| **Major Comorbidities (IQR)** | 1 (1,1) | 2 (2,4) | 0.0273 |
| **Immunosuppression/Cancer (%)** | 0/5 (0.0%) | 5/31 (16.1%) | 0.9999 |
| **Pulmonary (%)** | 0/5 (0.0%) | 12/31 (38.7%) | 0.1464 |
| **Cardiac (%)** | 1/5 (20.0%) | 21/31 (67.7%) | 0.0637 |
| **Liver (%)** | 0/5 (0.0%) | 2/31 (6.5%) | 0.9999 |
| **Neuro (%)** | 0/5 (0.0%) | 3/31 (9.7%) | 0.9999 |
| **Diabetes Mellitus (%)** | 1/5 (20.0%) | 15/31 (48.4%) | 0.3549 |
| **CKD (%)** | 0/5 (0.0%) | 5/31 (16.1%) | 0.9999 |
| **Obesity (%)** | 4/5 (80.0%) | 18/31 (58.1%) | 0.6283 |
| **Days from onset to death (IQR)** | 31 (27,42) | 18 (10.5,26.5) | 0.0453 |
| **Hospital duration, days (IQR)** | 27 (22,33) | 10 (5.5,16.5) | 0.0016 |
| **ICU duration, days (IQR)** | 22 (22,29) | 7 (4,14.5) | 0.0207 |
| **Intubation duration, days (IQR)** | 22 (21.0,29.0) | 6 (1.0,13.5) | 0.1076 |
| **Post-Mortem Interval, hours (IQR)** | 19.8 (19.2,33.7) | 24.6 (19.5,36.5) | 0.5296 |
| **ICU Admission, (%)** | 5/5 (100.0%) | 29/31 (93.5%) | 0.9999 |
| **Intubated, (%)** | 5/5 (100.0%) | 27/31 (87.1%) | 0.9999 |
| **Pressor Use (%)** | 5/5 (100.0%) | 23/31 (74.2%) | 0.5655 |

| | | | |
|---|---|---|---|
| **RRT (%)** | 3/5 (60.0%) | 12/31 (38.7%) | 0.6296 |
| **ECMO (%)** | 2/5 (40.0%) | 5/31 (16.1%) | 0.2440 |
| **Abnormal Chest Imaging (%)** | 5/5 (100%) | 30/31 (96.8%) | 0.9999 |
| **Antibiotic Use, (%)** | 5/5 (100.0%) | 29/31 (93.5%) | 0.9999 |
| **HAP Coverage, (%)** | 4/5 (80.0%) | 20/31 (64.5%) | 0.6457 |
| **COVID-Specific Therapies** | | | |
| **Steroid Use, (%)** | 5/5 (100.0%) | 27/31 (87.1%) | 0.9999 |
| **Remdesivir, (%)** | 4/5 (80.0%) | 14/31 (45.2%) | 0.3377 |
| **Tocilizumab, (%)** | 2/5 (40.0%) | 2/31 (6.5%) | 0.1548 |
| **Convalescent Plasma, (%)** | 2/5 (40.0%) | 6/31 (19.4%) | 0.3048 |
| **Days from onset to death (IQR)** | 31 (27,42) | 18 (10.5,26.5) | 0.0453 |
| **Hospital duration, days (IQR)** | 27 (22,33) | 10 (5.5,16.5) | 0.0016 |
| **ICU duration, days (IQR)** | 22 (22,29) | 7 (4,14.5) | 0.0207 |
| **Intubation duration, days (IQR)** | 22 (21.0,29.0) | 6 (1.0,13.5) | 0.1076 |
| **Post-Mortem Interval, hours (IQR)** | 19.8 (19.2,33.7) | 24.6 (19.5,36.5) | 0.5296 |
| **ICU Admission, (%)** | 5/5 (100.0%) | 29/31 (93.5%) | 0.9999 |
| **Intubated, (%)** | 5/5 (100.0%) | 27/31 (87.1%) | 0.9999 |
| **Any Pre-mortem Infection** | 4/5 (80.0%) | 12/31 (38.7%) | 0.1493 |
| **Pulmonary** | 2/5 (40.0%) | 9/31 (29.0%) | 0.6309 |
| **Extra Pulmonary** | 4/5 (80.0%) | 11/31 (35.5%) | 0.138 |
| **Bacteremia** | 3/5 (60.0%) | 8/31 (25.8%) | 0.1544 |
| **UTI** | 0/5 (0.0%) | 2/31 (6.5%) | 0.9999 |
| **Fungemia** | 2/5 (40.0%) | 2/31 (6.5%) | 0.0843 |

| | | | |
|---|---|---|---|
| **Skin/Soft Tissue Infection** | 1/5 (20.0%) | 0/31 (0.0%) | 0.1389 |
| ***C. difficile* colitis** | 0/5 (0.0%) | 1/31 (3.2%) | 0.9999 |