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Clinical Presentation: The initial symptoms of COVID-19 infection are generally mild and characterized most commonly by fever (43-98%), cough (68-82%), and shortness of breath (19-55%). In a recent report of >4700 cases in healthcare workers 92% had at least one of the symptoms of fever, cough, or shortness of breath. Other symptoms may also be present early in the illness such as headache (10-65%), myalgias (11-66%), sore throat (5-38%), loss of sense of smell or taste (up to 15-60%) and GI upset (nausea, vomiting, diarrhea 1-20%). Rhinorrhea (4-10%) and nasal congestion alone are relatively uncommon as opposed to many other respiratory viruses and seasonal allergies. Most persons infected with COVID-19 who are young and healthy will recover without problems. Those who are older or immunocompromised are more likely to progress to pneumonia which can be severe.

Testing Recommendations: Testing is widely available and all patients with symptoms suspicious for COVID-19 should undergo testing. Testing should be performed in **ALL** patients who present with symptoms typical of COVID-19 (cough, fever, shortness of breath), those with severe symptoms, and those with a combination of the less typical symptoms listed above. Asymptomatic testing can be performed in those who have had high-risk exposures and as part of pre-procedural screening. Patients with no symptoms who have no known exposures to a COVID case should not be tested. We recommend testing in the following patients:

1. Those with symptoms that could be compatible with COVID-19 (inpatient and outpatients).
 - o Differentiating COVID-19 from influenza is difficult and clinicians should follow the guidance below regarding evaluating patients when influenza virus is circulating.
 - o Differentiating pneumonia due to COVID-19 from other forms of pneumonia can also be difficult. See information below on differentiating types of pneumonia. Other respiratory testing (respiratory panels, culture, etc.) could also be obtained if appropriate.
 - o Patients with respiratory symptoms who do not undergo testing are presumed to be infected with SARS CoV-2 and should be isolated per available guidelines.

2. Anyone working in a healthcare environment who has symptoms, which could be compatible with COVID-19.
 - Because of their extensive and close contact with vulnerable patients in healthcare settings, even mild signs and symptoms of COVID-19 should be evaluated among healthcare personnel.
 - Testing directed by Employee Health or Student Health should be performed as directed. Employee Health is open 7AM – 430PM, Monday to Friday and can be reached at 402-552-3563. After hours and weekends, call the OUCH pager (888-OUCH).
3. Persons with high risk exposures to SARS-CoV-2
 - High risk exposure typically defined as being within 6 feet of someone with documented COVID-19 for at least 15 minutes (see information below)

Influenza Testing: Influenza symptoms can mimic COVID-19 and vice versa. When influenza begins to circulate in the community, patients who present with respiratory symptoms should undergo testing for both viral entities. To accommodate this we are offering SARS CoV-2/influenzaAB combination testing on our major COVID-19 testing platforms. These tests will only be offered in combination and individual assays for SARS-CoV-2 and influenza will not be available. Rapid influenza tests will not be available this season. Please utilize the following guidance in testing and interpreting the combination test.

- Symptomatic patients → Test for both influenza and SARS-CoV-2 using the combined assay.
 - Co-infection with both viral pathogens is rare but has been reported to occur
 - Decisions regarding the need for COVID-19 isolation and 1 or 2 tests to rule out COVID-19 should follow current protocols
 - Detection of influenza does not rule out SARS-CoV-2 infection and vice versa
 - Positive for Influenza
 - Place inpatients in contact/droplet isolation
 - Immunocompetent patients can exit isolation at 7 days as long as symptoms are improving (i.e. resolution of fever, improved cough, etc.)
 - Immunocompromised patients require a test be negative for influenza before exiting isolation. Testing should not occur until the following criteria are met:
 - 7 days after positive test
 - Resolution of fever and significant improvement in symptoms (cough, muscle pains, etc.)
 - Outpatients should remain at home until their fever has resolved for at least 24 hours without fever reducing medication and their symptoms are improving
 - Treatment
 - We recommend treatment of influenza in all symptomatic hospitalized patients.
 - Treatment of outpatients depends on the risk of severe disease. Treatment can be considered in all patients but is recommended in those at high risk for influenza complications (>65 years, <2 years, those with chronic medical conditions or immunocompromising conditions) and those who live in congregant settings (LTCF, homeless shelter, etc.). CDC guidance on treatment groups: <https://www.cdc.gov/flu/professionals/antivirals/summary-clinicians.htm>
 - Positive for SARS-CoV-2
 - Isolation and treatment of COVID-19 should follow current protocols
 - Positive for both influenza and SARS-CoV-2
 - Isolation should follow COVID-19 protocols

- Treatment of influenza is recommended
- Asymptomatic patients (procedural and admission based-screening) → Test for both influenza and SARS-CoV-2 using the combined assay.
 - Positive for SARS-CoV-2
 - Inpatients should be cared for on the COVID unit and monitored for symptoms development
 - Outpatients isolate at home and follow current guidance on timing of procedures
 - Positive for influenza
 - Inpatients should be placed in contact/droplet isolation and monitored for symptoms. Isolation can be discontinued 7 days after the positive test if no symptoms develop in all patients. Those who develop symptoms should follow the guidance above for symptomatic influenza.
 - Outpatients should isolate at home for 48 hours and monitor for symptom development. If no symptoms develop, they can exit home isolation after 48 hours.
 - Treatment is not recommended for asymptomatic influenza cases.

Screening for COVID-19 in Asymptomatic Patients: Due to the significance of asymptomatic and pre-symptomatic transmission screening for infection in asymptomatic patients, testing is recommended for close contacts of persons with documented SARS-CoV-2. In addition, testing of asymptomatic persons is recommended when undergoing high-risk procedures or entering high risk environments. Testing of asymptomatic persons should be restricted to the following situations:

- Before immunologically high-risk procedures such as bone marrow or organ transplant
- Pre-procedural testing meeting risk criteria
 - Not all procedures require testing and current institutional guidance should be followed
 - Testing should be arranged in the outpatient setting 48-72 before the procedure
- Admission screening of all inpatients, even if asymptomatic, is required
- Outbreak investigation as directed by infection control, employee health, or public health
- As required for admission for congregant settings (LTCF, homeless shelter, etc.)
- Those with documented close exposure* to a person infected with SARS-CoV-2
 - Close exposure includes the following:
 - Healthcare workers spending >15 minutes within 6 feet of a person with known COVID-19 while not wearing a procedure or N-95 mask (or higher level of protection)
 - Participation in an aerosol generating procedure (any duration) while not wearing appropriate PPE (N-95 or PPAR)
 - Non-healthcare workers spending >15 minutes within 6 feet of a person with known COVID-19 (quality of mask and fit can be considered in evaluating exposure)
 - Anyone identified by public health as having a high-risk exposure
 - The ideal testing interval for detection of infection is unknown, but nearly all patients who will develop symptoms do so by day 12-14. Knowing the average incubation period is around 5 days, a reasonable strategy would be to perform a first test at day 5-7. If this test is negative, this does not mean that a person can exit home isolation as viral shedding and symptoms may develop after this time. A second test could be performed around day 12-14 from exposure and if negative would mean it is safe to exit home isolation, potentially slightly early. Alternatively,

those without symptoms can exit home isolation after 14 days. Those who test positive should follow current guidance on duration of isolation.

- Testing is **NOT** recommended in the following situations:
 - Those with recent travel who do not have known exposure to persons with COVID-19
 - Some locations require testing before or after travel and some employers require testing before returning to work. While we do not generally recommend this practice if it is required for patients to undergo testing for these purposes, it is reasonable to obtain testing.

Ruling Out COVID-19:

- The currently utilized assays are very sensitive, particularly early in illness. Patients who have recently (within last 7 days) developed symptoms (either outpatient or inpatient) can usually be adequately ruled out with a single appropriately obtained NP swab. As viral levels in the nasopharynx begin to decline around a week into the illness a second test could be considered in patients who are 7 or more days into illness, especially if they have evidence of pneumonia. In these situations, a specimen from the lower respiratory tract would be preferred. Those with inconclusive results should have a second test obtained. Patients who need 2 tests to rule out COVID-19 should generally have a minimum of 18 hours between specimen acquisition.
- Patients who were initially screened while asymptomatic (surgical screening, etc.) and subsequently develop symptoms concerning for COVID-19 should undergo repeat testing
- Patients who have tested positive for COVID-19 should follow published isolation duration guidance available on our website based on their setting of care (inpatient vs. outpatient).

Respiratory Pathogen Panel Use: The new RPP includes SARS-CoV-2 RPP and has utility in diagnosing a variety of viral and bacterial pathogens, but it is expensive and may not impact clinical management. It should be reserved for use in populations where knowledge of specific viral pathogens beyond SARS-CoV-2 and influenza have clinical importance (i.e. RSV or Parainfluenza in stem cell transplant patient) or there is significant diagnostic uncertainty (i.e. atypical pneumonia not responding to antibiotics).

Current recommendations on use are outlined below.

- Patients with symptoms concerning for COVID-19 or influenza should be tested for both using the SARS CoV-2/influenzaAB combination test and the RPP should generally be avoided.
- For patients where there is strong suspicion for other respiratory pathogens or where COVID/influenza testing is negative the RPP may be considered
- The RPP is adequately sensitive for SARS-CoV-2 and can be used in the process of ruling out COVID, although testing using the current SARS CoV-2 test is preferred. The determination of the need for 1 or 2 tests to rule out COVID-19 should follow current protocols.
- Patients who have been tested with the RPP should not be retested for at least 7 days.
- Asymptomatic patients should **NOT** be tested using the RPP including pre-procedural and admission-based screening. Screening for COVID-19 should be done using the SARS CoV-2/influenzaAB test.

Directing COVID-19 Testing in Patients with Pneumonia: Differentiation of patients with COVID-19 infection from those with bacterial pneumonia can be difficult. The guidance below is provided to assist in deciding a patient’s management. Patients with syndromes highly consistent with bacterial infection should still have COVID-19 ruled out with at least 1 test, while those presenting with an “atypical pneumonia” with features concerning for COVID-19 should always be placed in appropriate isolation and tested.

	COVID-19	Bacterial Pneumonia
Symptoms	Fever, Cough, Shortness of Breath, Viral prodrome (headache, myalgias, nausea/vomiting/diarrhea)	Fever, Cough, Shortness of Breath, Purulent sputum
Lab Abnormalities	Normal/low WBC, Lymphopenia common Mild hepatitis	Leukocytosis, bandemia
Procalcitonin	Usually low	Elevated
Exposure	Close contact with confirmed or suspected case of COVID-19 Work or live in high risk environment	None
Imaging	Bilateral infiltrates, “atypical pneumonia” Multifocal ground-glass opacities	Lobar Infiltrate
Treatment	Supportive Care, consider remdesivir Airborne, contact, droplet Isolation	Antibiotic Therapy Precautions per etiology

Ordering COVID-19 Testing to Detect SARS-CoV-2 and/or Influenza:

- COVID-19 and influenza testing is available in One Chart using the COVID testing order panel. Clinicians should answer any necessary questions when ordering the assay to accurately reflect their reason for testing.
- Testing may be performed on a variety of molecular panels but only a single order is used to order all COVID testing. Tests currently in use include:
 - Roche Cobas 6800 (Turn around time (TAT) <24 hours)
 - NM Developed COVID Test (TAT <24 hours)
 - Hologic Aptima TMA SARS CoV2 (TAT <24 hours)
 - Cepheid SARS-CoV-2 (TAT <3 hours), limited supply
- In most cases, testing will be performed on the Roche Cobas 6800 platform. However, in certain circumstances, testing will be performed using our laboratory developed test, the Hologic Panther. Rapid testing will be performed using the Cepheid platform. The agreement between these assays is excellent.
 - Rapid testing is reserved for pre-solid organ transplant, laboring mothers, and very urgent surgical cases. Use outside of these parameters should be discussed with microbiology
- When ordering the test clinicians must designate if the patient has symptoms concerning for COVID-19/Influenza or if they are asymptomatic.
 - If symptomatic a patient flag for “COVID Rule Out” will be automatically added to the patient’s chart. The appropriate isolation should also be ordered (airborne, contact/droplet).
 - Patients who are asymptomatic and being screened upon admission or pre-procedurally will not be flagged in any way.
- If the test is positive for SARS-CoV-2 an infection flag for “COVID-19” will be automatically added to the patient’s chart.
- If the test is negative the “COVID Rule Out” flag will be automatically removed. Some patients will need additional testing to rule out COVID-19 (see below).

Interpreting COVID Testing Results: Three assays are currently being used to test for COVID-19 which may provide slightly different results.

Roche Cobas 6800 (COV19) interpretation

- This assay is designed to detect RNA from two genes within SARS CoV 2, the E- gene and the ORF1a-gene.
- If both the E- and ORF1a-genes RNA are not detected, the following will be resulted: **Not Detected - SARS CoV2 RNA NOT DETECTED by RT-PCR.**
- If both the E- and ORF1a-genes RNA are detected, or ORF1a- alone, the following will be resulted: **Detected - SARS CoV2 RNA DETECTED by RT-PCR.**
- If only the E-gene is detected, per the FDA EUA document, the following will be resulted: **Presumptive Positive – Sarbecovirus RNA DETECTED by RT-PCR.** Note that the E-gene is conserved with SARS-CoV and other coronaviruses found in bats. However, SARS CoV-1 has not been isolated since 2003 and infection from bats in highly unlikely. Thus, detection of the E-gene alone should be interpreted as positive for SARS-CoV-2.
- In rare cases, the sample will fail internal controls and the result will be **Invalid.** In this situation, we suggest recollection of the sample if clinically indicated.

Hologic Aptima TMA SARS CoV2 interpretation

- This assay is designed to detect RNA from two regions within the SARS-CoV-2 ORF1ab gene.
- Will be resulted as the following:
 - **Not Detected - SARS CoV2 RNA NOT DETECTED by RT-PCR.**
 - **Detected - SARS CoV2 RNA DETECTED by RT-PCR.**
- In rare cases, the sample will fail internal controls and the result will be **Invalid.** In this situation, we suggest recollection of the sample if clinically indicated.

Laboratory developed test (NE_{CoV19}) interpretation

- This assay is designed to detect RNA from two genes within SARS CoV 2, the E gene and the N-gene
- If the E-gene RNA is not detected, the following will be resulted: **Not Detected - SARS CoV2 E gene RNA NOT DETECTED by RT-PCR.**
- If the E-gene RNA is detected, the same sample will be re-tested on the next run for both the E-gene RNA plus a separate N-gene target to confirm if it is positive. If either or both the E- or N- target RNA is detected upon retesting, the following will be resulted: **Detected - SARS CoV2 E gene RNA DETECTED by RT-PCR.**
- However, if neither the E- or N-gene RNA is detected upon retesting, a result of inconclusive is reported as follows: **Inconclusive result - Please contact Microbiology director for further consultation at 402-888-5626**

Rapid Test Results (Cepheid)

- Results are reported as either detected or not detected

How to Manage Inconclusive Results: Inconclusive results most probably reflect low levels of virus in the sample but may also represent a false positive result. We recommend the following:

- For **inpatients** who have an initial inconclusive result on a NP swab, we recommended a new respiratory tract sample be obtained 24 hours after the first sample. If possible, a lower respiratory tract specimen (sputum, tracheal aspirate, etc.) should be obtained, particularly in those with evidence of pneumonia.
 - If the new test is positive or inconclusive, the patient should be considered positive for COVID-19.
 - If the new test is negative, the need for additional testing depends on the suspicion of disease.
 - If suspicion is low, consider negative for COVID-19.
 - If suspicion is high (atypical pneumonia, high risk exposure, etc.), consider third test, again preferentially of lower tract secretions if possible.
 - Discussions with infection control are appropriate to decide if additional testing is needed in cases where results are difficult to interpret. Please contact 888-4646 to discuss.

- For **outpatients** who have an inconclusive result, a second swab should be collected 24 hours after the first swab was obtained and re-tested.
 - If the second test is negative, the patient will be considered as negative for COVID.
 - If the second test is positive, the patient will be considered positive for COVID and a public health investigation will be opened.
 - If the second test remains inconclusive, interpretation is difficult. This may indicate presence of low-level viral RNA or be falsely positive. Out of an abundance of caution, the patient should be advised to continue home isolation similar to a patient with a positive result (10 days from symptom onset and at least 5 days of fever free without antipyretics and improving respiratory symptoms). If symptoms were to progress or worsen, obtaining additional specimens for repeat testing may be considered.

Role of Serology in COVID-19 Diagnosis:

- Infection with COVID-19 produces a rapid serologic response with 95% of persons with symptomatic infection demonstrating detectable IgG antibodies by day 14.
- Numerous serologic assays are available in the US. Many of these have not been well validated and may produce both false positive and negative results. Outside serologic testing is difficult to interpret.
- A serologic assay is available at Nebraska Medicine (DiaSorin SARS CoV2 IgG) which detects COVID-19 IgG antibodies. This is a qualitative assay which yields a positive or negative result only. It has been validated and is highly sensitive. Furthermore, this assay will not detect antibodies directed against seasonal coronaviruses.
 - IgM testing is not currently available nor are quantitative IgG levels. These may become available in the future.
- The current role of serologic testing is unknown. Possible use cases are noted below.
 - Patients who previously had a syndrome compatible with COVID-19 and were not tested
 - Evaluating timing of illness in asymptomatic patients with positive screening tests
 - High risk exposure patients who did not develop symptoms
 - Conflicting test results in an asymptomatic patient
 - Serology is NOT recommended to diagnose COVID-19 infection in patients with symptoms. Current molecular testing should be used in these situations.
- The duration of immunity in patients with positive serology is unknown and a positive serology does not mean that PPE recommendations can be disregarded.

Testing for Recurrent COVID-19 Infection:

Although there are very limited data to indicate that persons can become re-infected in the short term, there is concern that this may occur. The CDC states the following, “Patients previously infected with SARS-CoV-2 who have clinically recovered but later develop symptoms c/w COVID19, should be isolated and retested.” This means patients who had COVID-19 and recovered, and then develop symptoms suggestive of COVID-19 at a later date (typically months later) should be considered to have recurrent infection until proven otherwise. Unfortunately, if patients test positive it is unknown if reinfection is actually occurring or if this represents prolonged viral shedding. RT-PCR assays can remain positive for weeks or months and do not correlate with recovery of viable virus or transmissibility of disease.

We recommend patients with a history of COVID who have recovered and subsequently present with symptoms consistent with COVID be placed in COVID isolation and tested as per above guidance. Those who test positive will may remain in isolation until they meet criteria for exiting isolation as outlined. Consultation with the COVID ID MD is essential in sorting out these complex issues.

Frequently asked questions/answers regarding COVID testing at Nebraska Medicine

Which COVID test should I order?

- COVID-19 & Influenza A/B by PCR (COV19) is the appropriate test for almost all patients with a few exceptions noted below.

When will I have a result?

- You can expect a result within 24 hours of receipt in the microbiology lab.

Why does the turnaround time seem to vary?

- The turnaround clock starts once the specimen is received in the lab, not obtained. Specimens collected at outlying clinics or via drive through testing must be transferred via courier which takes additional time. You can check the order status in the “Micro” tab in One Chart.
 - “Pending” indicates the specimen is currently in the laboratory specimen receiving area or microbiology.
 - “Active” indicates the specimen has not yet arrived in the laboratory.
- Samples are grouped and run periodically throughout the day (“batched”). For example, one instrument runs 94 samples at a time. If we have 93 specimens and yours is number 94, you will have a result in about four hours. If your specimen is the first to arrive after we run a batch of 94, it will not go on our instrument until 93 more specimens are received and processed in the microbiology lab. This may take from a few hours (some weekdays) to most of a day (weekends).

When should I order a rapid COVID test?

- We have a **very** limited supply of rapid tests available due to allocation from the company. Guidance on the use of rapid tests is available above. Rapid tests should only be used when rapid knowledge of COVID status is essential to patient care. Example of these include the following:
 - Pre-solid organ transplant
 - Laboring mothers without previous COVID testing history
 - Pediatric Oncology patients being admitted to the hospital
 - Emergent surgery or procedure within next 4-6 hours
- Please note that general preoperative screening is not an appropriate use for the rapid test. Pre-operative screening specimens should be obtained and arrive in the microbiology laboratory by the afternoon of the day before the procedure. Specimens submitted within this time frame will be result by the next morning. Therefore, depending on the location where the specimen is obtained, it may need to be collected greater than 24 hours in advance of the procedure.
- If you feel your patient needs a rapid COVID test and does not fit the above criteria, please page Dr. Paul Fey, Director of Microbiology, at 402-888-5626.

This is an emergency. How long does it take to result a rapid test?

- Rapid tests are managed as any other stat specimens and will take approximately 75 minutes once received in the microbiology lab. This is the fastest, reliable testing modality currently available.

My patient has a procedure tomorrow and has not had a specimen collected yet. What do I do?

- To ensure pre-operative specimens will be result before a scheduled procedure, they should be collected by 12 pm the day before the procedure.
- If unforeseen events prevent this from happening, an infrastructure has been developed (a list generated via One Chart) allowing for patients with a procedure the following day to be identified and tested on the last “run” of the night. This is result between 11pm and 3am.

- If an inpatient specimen is collected after 7pm, or a specimen is collected at an outside clinic at a time that may prevent it from arriving in the microbiology laboratory before 7pm, please page the microbiology fellow on-call at 402-888-1529.

I need a result in >6 but <24 hours. What do I do?

- Please call the microbiology laboratory (402-552-2090) before the specimen is collected. We may be able to arrange testing so that you will have a result in time. Alternatively, we may advise you to place a rapid order.

Why can't I order a COVID test?

- If a patient has an active COVID+ flag, then One Chart will not allow users to enter a rapid test. This is because the patient has known COVID infection and the only reason to test would be to determine if the patient can exit isolation. Thus, only follow-up testing is indicated and available. If it has been less than 70 days since the COVID+ flag was resolved, then One Chart will not allow additional COVID testing. This is because of the likelihood of detected prolonged viral shedding which will not impact clinical care. If additional testing is required (e.g. the patient was moved off the COVID unit after 21 days, but is being discharged to a facility that requires two negative tests), please page Dr. Paul Fey, Director of Microbiology, at 402-888-5626 and we can place this order for you.

My patient previously tested positive for COVID-19. Should they be tested upon admission/before a procedure?

- If the patient's first positive test by PCR was between 21 and 90 days ago, treat the patient as COVID negative. Do NOT order a test as results during this time period will not be clinically meaningful (i.e. a positive test may indicate shedding or very low-level viral replication)

What if I need to change my order to a rapid COVID?

- One Chart will not allow you to cancel a lab order once the specimen has been received by the lab. Please page the microbiology fellow on call at 402-888-1529.

Does my patient have to go to the COVID floor?

- Decisions regarding location of patient care are dictated based on the presence of symptoms of COVID and should be managed using our current guidance. The COVID Infectious Disease Unit Physician is available via PerfectServe:
 - Search "Infectious Disease Nebraska Medicine"
 - Select "COVID-19" when asked what service you are trying to reach.
 - Select "COVID-19 Testing Questions Infectious Disease."

What does a "presumptive positive" result mean?

- Generally, this should be interpreted as a positive result, although it means only one of the two target genes was detected indicating that there may be a very low titer of virus in the sample. There are two target genes that our instrument detects, E and ORF1A. ORF1A is unique to SARS-CoV-2 and, therefore, when it is the only gene detected, we report this result as "Detected." However, the E-gene is shared with the original SARS virus and other bat coronaviruses. The FDA therefore requires us to report a "presumptive positive" when only the E-gene is detected. However, since SARS CoV-2 is the only SARS virus currently circulating in humans, detection of this target is indicative of current infection with SARS-CoV-2. More detailed information is available above.

What other tests are available that assess COVID status?

- The respiratory pathogen panel, aka BioFire RP 2.1 [Respiratory pathogen panel by PCR (viral/bacterial) in One Chart] detects SARS CoV-2 along with other viral entities. Due to a limited quantity of these tests, we ask that providers not order this test specifically to determine COVID status. Please note that the pneumonia panel (PNP) does not detect SARS CoV-2.
 - Order the respiratory pathogen panel ONLY if you need a multiplex PCR panel which detects additional viral targets. Note, however, that you do not need to order a separate COVID test as the RP2.1 panel detects SARS CoV-2. See guidance above for further information on appropriate use for the RP2.1.

What is the role for the antibody test (SARS CoV2 IgG Antibody)?

- See information above regarding serology use.
- High sensitivity for COVID-19 infection starting the second week after PCR positivity but exact utility still be evaluated
- Turnaround time on serologic testing is generally around 24 hours.

Specific laboratory question can be directed to: Kathie Rogers, PhD (402-552-3313 or katrogers@nebraskamed.com) or Paul D. Fey, PhD (phone (402) 559-2122, pager (402) 888-5626, email: pfey@unmc.edu)