

Comparison of enzyme immunoassay (EIA) to immunodiffusion and complement fixation for coccidioidomycosis diagnosis

Rachel Lusk, MS IV, University of Arizona College of Medicine-
Phoenix

Laura Erhart, MPH, Arizona Department of Health Services
Arizona Infectious Disease Training and Exercise

August 2, 2012

Background

- Coccidioidomycosis
 - Prevalence in 2009: 155.1 cases per 100,000 population
 - Coccidioidomycosis infections were reported more frequently in Arizona than any other non-sexually transmitted reportable disease, excluding influenza.
- Coccidioidomycosis is reportable disease by laboratories and physicians in the state of Arizona.
 - Positive cases for public health surveillance utilize laboratory testing only, not clinical data

Diagnosis

- Laboratory diagnosis of coccidioidomycosis:
 - Culture or microscopy
 - can be difficult since the dry cough can prevent patients from providing a sputum sample, and most patients do not have lesions that require a biopsy
 - Serological testing
 - Complement Fixation
 - Immunodiffusion
 - Enzyme Immunoassay

Laboratory Diagnosis

- Enzyme immunoassay (EIA):
 - Considered most sensitive test
 - False positive rates, especially with the EIA IgM test, have been reported by many researchers
- Immunodiffusion (ID):
 - Qualitative
 - Considered to be as sensitive as EIA tests, but more specific
 - Require a longer incubation period than EIA exams
- Complement fixation (CF):
 - Quantitative, so can be used serially to determine the prognosis for the individual and efficacy of antifungal therapy
 - CF titers measure only an IgG response, which appears later in infection, and are not considered as sensitive as the EIA or ID tests
 - Some additional disadvantages: needs to be heat inactivated, and it needs overnight incubation.

Diagnosis

- Diagnosis of coccidioidomycosis can be very problematic for physicians.
 - Symptoms often nonspecific, and the lesions produced by infection can be comparable to lesions caused by other infections or neoplasms
 - This makes laboratory diagnosis important, but a lot of questions about how to utilize serology testing
 - EIA results can be obtained more quickly, but false positive rates and cross reactivity with other infections can make the results difficult to rely on.
 - Other tests may be more specific, but more difficult to obtain and require more time to obtain results.

Study Objective

To examine how EIA positive results compare with other tests from the same specimen, and the characteristics of cases that are only positive by EIA.

Methods

- Coccidioidomycosis laboratory results were provided by a commercial laboratory for a 6-month period (April-September 2009)
- Inclusion Criteria
 - Both EIA tests performed (IgM and IgG) and at least two of the other tests (ID IgM, ID IgG, or CF) run from the same specimen.
 - Only the earliest set of tests is included for individuals with more than one set.
 - EIAs are considered positive if one or both of the EIAs in the set are positive
 - EIAs are considered negative if both EIAs are negative.
 - Likewise, the comparison tests are considered positive if any of the included tests are positive and negative if all tests are negative.

Methods Continued

- Analysis included:
 - Calculation of sensitivity and positive predictive value for EIA compared to non-EIA tests
 - Descriptive statistics for EIA+/ID- patients
- Medical records requested for a 5% sample of patients that were positive for one or both of the EIA tests and negative for both immunodiffusion (ID) tests. (EIA+/ID-).
- Charts were reviewed to identify:
 - Subsequent positive non-EIA coccidioidal tests (ID, CF, tissue biopsy or culture)
 - Whether patient meets the ADHS clinical case definition for coccidioidomycosis
 - Illness characterized by one or more of the following: influenza-like signs and symptoms, including fever, chest pain, cough, myalgia, arthralgia, headache; pneumonia or other pulmonary lesion, diagnosed by chest x-ray; rashes, including erythema nodosum or erythema multiforme; involvement of bones, joints, or skin dissemination; meningitis; or involvement of viscera and lymph nodes.
 - Physician's diagnosis
 - Patient history of coccidioidomycosis

Results

10,243 sets of tests received

9,935 sets received where both EIAs and at least two ID tests run

8,564 sets with both EIAs and at least two other tests (CF, ID, or culture) for unique individuals

4,991 positive for EIA but negative for ID or CF

246 charts were reviewed (5%)

Results

	ID Positive	ID Negative	Total
EIA positive (IgM or IgG)	1639 (19 %)	4991 (58 %)*	6630 (77%)
EIA negative (both tests)	43 (0.5 %)	91 (22 %)	1934 (23%)
Total	1682 (20 %)	6882 (80 %)	8564

*Category from which charts were selected for review

Results: Subsequent Testing

(from medical record review)

Test	Number Tested	Number Positive	Percent Positive
Immunodiffusion (IgM or IgG)	37	4	11%
Complement Fixation	14	2	14%
Biopsy or Culture	28	6	2%
Any non-EIA test	79	12	15%

- Number of specimens with subsequent positive tests for the whole study population estimated based on medical record review results

Sensitivity and PPV

	Sensitivity	PPV
Original testing	97.4%	24.7%
With subsequent positive testing	97.8%	26.7%

Sensitivity: $(EIA+, ID+) / [(EIA+, ID+) + (EIA-, ID+)]$

PPV: $(EIA+, ID+) / [(EIA+, ID+) + (EIA+, ID-)]$

- Patients with subsequent positive tests considered “true positives” for purposes of sensitivity & PPV

Results: Chart Review

- For charts reviewed (n=246):
 - 206 (84%) met the ADHS Clinical Case Definition
 - Clinical definition: Influenza like signs and symptoms, pneumonia or other pulmonary lesion, erythema nodosum or erythema multiforme rash, meningitis, or involvement of bones, joints, etc., by dissemination
 - 61 (25%) patients were hospitalized
 - during testing or required hospitalization from illness that led to testing
 - 24 (10%) had a history of cocci
 - 28 (11%) patients were being screened for cocci
 - Patients that did not present with symptoms but were being screened for cocci for other reasons, such as being placed on immune modulating medications

Results: Clinical Diagnosis

- Most common diagnoses of charts reviewed (n=246):
 - Cocci: 56 (23%)
 - Pneumonia: 23 (9%)
 - Bronchitis: 19 (8%)
 - Upper Respiratory Illness: 13 (5%)
 - Other: 77 (31%)
 - Diagnosis not listed: 58 (24 %)
- Number that had subsequent positive test AND/OR met the clinical case definition: 206 (84%)
- Number that had subsequent positive test AND/OR met the clinical case definition and were diagnosed with cocci: 63 (26%)

Discussion

- Performance of EIA, compared to immunodiffusion, complement fixation, culture, or biopsy performed on the same specimen or in subsequent tests:
 - Sensitivity relatively high
 - Positive predictive value low
- Most (84%) patients with EIA+/ID- tests had clinical symptoms compatible with cocci
- Only 1/4 of EIA+/ID- patients were diagnosed with cocci by their clinician

Discussion

- Study has been conducted with data from a different AZ lab using the same methods

	This Lab	Other Lab
Sensitivity (with subsequent testing in parentheses)	97.4% (97.8%)	80.4% (83.8%)
Positive predictive value	24.72% (26.7%)	47.8% (61.5%)
Met ADHS clinical case definition	84%	98%
Physician diagnosed cocci	35%	86%
Number that had subsequent positive test AND/OR met the clinical case definition	84%	85%
Number that had subsequent positive test AND/OR met the clinical case definition and were diagnosed with cocci	26%	98%

Conclusions

- Test performance may vary between laboratories.
- Results from both laboratories demonstrate that subsequent laboratory confirmation with a different test may be useful for confirming EIA results.
- Most patients with EIA+/ID- tests have symptoms clinically compatible with cocci.
- Additional study is needed to determine how best to interpret EIA-positive results for clinicians and public health officials, in the absence of additional test results.

Acknowledgements

- ADHS
 - Laura Erhart, MPH
 - Clarisse Tsang, MPH
- Maricopa Department of Public Health & CDC
 - Rebecca Sunenshine, MD
- University of Arizona College of Medicine-Phoenix
 - Nathalie Petein, MD

Questions?

Rachel Lusk

rlusk@email.arizona.edu

(480) 272-3193