UTILITY OF BIOMARKERS IN IBD

Matilda N. Hagan, MD
Melissa L Posner Institute for Digestive Health and Liver Disease, Mercy Medical Center
Clinical Assistant Professor of Medicine
University of Maryland School of Medicine

OBJECTIVES

• Role of various fecal and serological biomarkers in the diagnosis of IBD

• Recognize utility of biomarkers, drug metabolites or anti-drug antibodies in IBD treatment
IBD DIAGNOSIS

• Diagnosis made based on clinical symptoms, endoscopic and histological pattern and imaging.

• Endoscopic evaluation considered the gold standard
  – Invasive
  – Expensive
  – Inconvenience

NEED FOR ALTERNATIVE DIAGNOSTIC TOOLS

• Accurately distinguish IBD from non IBD patients
• Differentiate the subtypes of IBD, UC or CD
• Identify high risk individuals
• Predict clinical course and response to treatment
### SERUM BIOMARKERS IN IBD

<table>
<thead>
<tr>
<th>BIOMARKER</th>
<th>DESCRIPTION</th>
<th>CHARACTERISTICS</th>
</tr>
</thead>
<tbody>
<tr>
<td>CRP</td>
<td>Acute phase protein; Produced by hepatocytes driven by IL-6, TNF-α, IL-1β; mesenteric adipocytes in CD pts; Short half-live (~19 hours) responsive indicator of inflammation</td>
<td>Higher CRP in CD &gt; UC; 25% CD normal CRP even with active disease; Genetic variations in expression; Elevated in other inflammatory conditions: infection, autoimmune, malignancy</td>
</tr>
<tr>
<td>ESR</td>
<td>Rate at which RBC's settle in plasma in 1 hour</td>
<td>Influenced by age, gender, anemia, blood disorders, and pregnancy; Peaks less rapidly than CRP; Resolves more slowly</td>
</tr>
</tbody>
</table>

Bruce, S. Gastroenterology 2015;149:1275–1285

### FECAL BIOMARKERS IN IBD

<table>
<thead>
<tr>
<th>BIOMARKER</th>
<th>DESCRIPTION</th>
<th>CHARACTERISTICS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fecal Calprotectin</td>
<td>Damage-associated molecular pattern (DAMP) proteins (S100A8/S100A9); Calcium and zinc binding; 60% Cytosolic protein in granulocytes; Stable in feces for up to one week at room temp</td>
<td>Elevated in other conditions: neoplasia, polyps, non-steroidal anti-inflammatory enteropathy, increasing age, celiac disease, microscopic colitis, allergic colitis, and infections.</td>
</tr>
<tr>
<td>Fecal Lactoferrin</td>
<td>Neutrophil specific iron binding glycoprotein; Stable in feces</td>
<td>Sensitivity and specificity variable depending on cutoff.</td>
</tr>
</tbody>
</table>

Bruce, S. Gastroenterology 2015;149:1275–1285; Lewis, J.D. Gastroenterology 2011;140:1817–1826
CD- Serum And Fecal Biomarkers Correlate With Endoscopic But Not Clinical Disease Activity

<table>
<thead>
<tr>
<th></th>
<th>IL-6</th>
<th>Calprotectin</th>
<th>Lactoferrin</th>
<th>CDAI</th>
<th>SES-CD</th>
</tr>
</thead>
<tbody>
<tr>
<td>hSCRP</td>
<td>0.65</td>
<td>0.47</td>
<td>0.52</td>
<td>0.16</td>
<td>0.46</td>
</tr>
<tr>
<td>IL-6</td>
<td>0.45</td>
<td>0.55</td>
<td>0.23</td>
<td>0.15</td>
<td>0.43</td>
</tr>
<tr>
<td>Calprotectin</td>
<td>0.76</td>
<td>0.23</td>
<td>0.45</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lactoferrin</td>
<td>0.19</td>
<td>0.48</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CDAI</td>
<td>0.15</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Coefficients in red are significant at the 0.05 level.

CDAI=Crohn's disease activity index; hSCRP= high sensitivity C-reactive protein; IL=interleukin; SES-CD=Simple Endoscopic Score-Crohn's Disease

*164 adult patients with Crohn's disease undergoing clinically indicated colonoscopy.

FECAL CALPROTECTIN

- More sensitive than CDAI or CRP at detecting endoscopic disease activity
- Reliable surrogate marker of mucosal healing in CD
- Elevated levels associated with increased risk of clinical relapse
- Diagnose presence of endoscopic recurrence after resection in CD


Wright et al. Inflamm Bowel Dis 2016; 22: 1086-1094
DISTINGUISHING IBD VS. NON IBD

• No single fecal marker is adequately sensitive or specific to definitively include or exclude IBD

• Fecal calprotectin showed a high negative predictive value in differentiating IBD from IBS
  – Sensitivity and specificity of 89% and 81% (cutoff of 50 µg/g)
  – Sensitivity and specificity of 98% and 91% (cutoff of 100 µg/g)

• CRP ≤ 0.5 or Calprotectin < 40 µg/g had less than 1% chance or less of IBD

Lewis, J.D. Gastroenterology 2011;140:1817–1826

FC Predicts Clinical Relapse Of Disease Activity

25/43 (58%) CD relapsed; RR 10.6 (95% CI, 2.5–45.8; P = 0.002)
19/37 (51%) UC relapsed; RR, 18.2 (95% CI, 4.0–82.5; P=0.0002)

All IBD FC > 50, RR 13.4; (95% CI, 4.6–39; P = 0.0001)


*80 IBD pts (CD n= 43; UC, n=37) in remission 1-4 months
*Relapse defined as CDAI > 150 for CD or HBI > 4 for UC
USE OF BIOMARKERS IN CLINICAL ENDPOINTS

• Ideal cutoff for fecal biomarkers are evolving
  – Assessment of response to therapy
  – Mucosal healing
    • Calprotectin <50 µg/g
  – Post-op recurrence

Lewis, J.D. Gastroenterology 2011;140:1817–1826

Fecal Calprotectin Predicts Outcome After Induction Therapy With Anti-TNF In IBD

• 60 patients
  – (CD, n= 34, UC, n= 26)
  – Treated with infliximab, (n=42) or adalimumab, (n=18)
  – Documented FC level at baseline and after induction

• Post induction
  – 31 (52%) pts normalized FC (<100 µg/g)
  – 29 (48%) Elevated FC (≥100 µg/g)

Normal FC after induction predicts sustained clinical remission

Matilda N. Hagan, MD

FC Can Monitor For Disease Recurrence After Intestinal Resection

- POCER study pts n=174
- FC preoperatively, at 6, 12, 18 m
- Median FC (6+18m) recurrence 330 mg/g (IQR, 163–540) vs. 75 mg/g (IQR, 37–258) in remission

Wright, E et al. Gastroenterology 2015;148:938–947

SEROLOGICAL MARKERS

- Classical Markers
  - perinuclear anti-neutrophil cytoplasmic antibodies (pANCA)
  - anti-Saccharomyces cerevisiae antibodies (ASCA IgA, IgG)
  - Antibody to outer membrane porin (Anti-OmpC)
  - Pseudomonas fluorescens-associated sequence I-2 (anti-I2)
  - Flagellin (Anti-CBir1)


Copyright 2016 American College of Gastroenterology
ASCA TITERS

Found in ~60% CD, ~10% UC, <5% non IBD


pANCA TITERS

Present in ~60% of UC
~20% of CD patients

Deurr et al. Gastroenterology. 1991;100:1590 –1596
SEROLOGICAL MARKERS

- IBD vs. Non-IBD, meta-analysis
  - ASCA+/pANCA–
    - Sensitivity 55% and a specificity of 93% for CD
  - pANCA+
    - Sensitivity 55.3% and specificity 88.5% for UC

- Predict phenotype progression in indeterminate colitis
  - ASCA+/pANCA- predicted CD (80%)
  - ASCA-/pANCA+ predicted UC (63.6%)

Reese et al. Am J Gastroenterol 2006;101:2410–22

ROLE IN PROGNOSIS

- Seropositivity associated with more complicated disease in CD
- Elevated anti-CBir1 associated with development of CD of the pouch after IPAA
- Anti-Omp C in UC associated with colectomy

CD- PROPORTION OF COMPLICATED PATIENTS INCREASES WITH INCREASING QUARTILE

SEROLOGIC MARKERS

Lichtenstein, G et al. Inflamm Bowel Dis 2011;17:2488–2496

Presence Of pANCA Associated With Less Complicated Disease In CD Pts

Lichtenstein, G et al. Inflamm Bowel Dis 2011;17:2488–2496
SEROLOGICAL MARKERS

- **Novel Markers**
  - Antibodies to Flagellin A4-Fla2 and Fla-X
    - CD 59% had antibodies to A4-Fla2 and 57% to Fla-X.
    - IBS patients 29% A4-Fla2 and 26% Fla-X
    - 6% UC
  - Anti-laminaribioside carbohydrate IgG (ALCA)
  - Anti-chitobioside carbohydrate IgA (ACCA)
  - Anti-synthetic mannoside antibodies (AΣMA or AMCA)

Iskandar and Ciorba Translational Research 2012;159:313–325

- **ALCA and ACCA associated with CD**
  - Less prevalent than ASCA
  - positive in 34% to 44% of CD patients who were ASCA negative

- **Anti-synthetic mannoside antibodies (AΣMA or AMCA) associated with CD**
  - positive in 24% of patients with CD who were ASCA negative

Iskandar and Ciorba Translational Research 2012;159:313–325
TAKE HOME POINTS

• Unable to definitively diagnose IBD based solely on fecal and/or serological markers without endoscopy or imaging
• Role of serologies in IBD diagnosis is supportive
• Serology may be useful as a prognostic tool in subset of patients
• Combination of fecal and serological marker can perhaps exclude disease in a low probability clinical scenario

BIOMARKERS IN MONITORING IBD

• Change in IBD treatment paradigm from treating to improvement of clinical symptoms to now treating to reduce inflammation.

• Widely recognized that poor correlation between symptoms and presence of inflammation
  – Colonoscopy, serum or fecal biomarkers

Bruce, S. Gastroenterology 2015;149:1275–1285
BIOMARKERS IN MONITORING IBD

- Therapeutic Drug monitoring proposed as a way to optimize medical therapy to achieve endpoint of reduction in inflammation.

- Infliximab, adalimumab and azathioprine.
  - Able to detect drug levels and antibodies to drugs for infliximab and adalimumab
  - Measure thiopurine metabolites

Correlation of 6-TGN Level to Clinical Response*

Odds Ratio 5.0 for treatment response when 6-TGN >235

*92 pediatric IBD patients receiving 6-MP or AZA ≥4 months.

6-TGN Levels Correlate With Infliximab Trough Levels

*Cross Sectional study 72 pts on maintenance therapy with IFX and Aza


BIOLOGICS

- 30-60% of patients initially responsive to anti-TNF will lose response during first year of therapy (secondary loss of response)

- Anti-drug antibodies (ADA) implicated in secondary loss of response
  - Binding drug and neutralizing it
  - Increase clearance of the drug

Casteele, NV et al. The Journal of Clinical Pharmacology 2015 S5(S3) S39–S50
Factors Associated with Increase Anti-TNF Clearance

- Low albumin ≤3 g/dL
- Higher body weight
- Presence of antibodies to drug
- High baseline CRP
- High baseline TNF concentration
- Male gender

Dotan, I et al. Inflamm Bowel Dis 2014;20:2247–2259

Timing of Drug Monitoring?

- Proactive Strategy
  - Prior to symptom occurrence
  - Before loss of response
- Reactive Strategy
  - In response to symptom recurrence
  - After loss of response
THERAPEUTIC DRUG MONITORING

• Normalization of calprotectin or normalization or reduction of CRP week 10-14 of treatment
  – Predict clinical remission and mucosal healing

• Infliximab levels higher than 3 to 4 mcg/mL at week 8 to 14
  – predict long term response to maintenance therapy

• ADA increases probability of active disease even in the presence of therapeutic drug levels


Trough level Adapted InfliXImab Treatment (TAXIT) Trial

• RCT; Compared efficacy, cost-effectiveness and safety of concentration based dosing to clinical based dosing of infliximab in IBD cohort (n= 263) on maintenance therapy.
  – Dose optimization to infliximab 3–7 mg/mL in all patients
  – Randomization to TDM vs.. conventional approach
  – Primary end point of clinical and biochemical remission at 52 weeks

TAXIT

- Primary endpoint not achieved
- However, TDM group less flares compared with conventional group
  - CD dose optimization led to more pts in remission and decrease in CRP
  - Dose optimization associated with cost savings for patients with supra optimal drug levels
  - ? Proactive strategy superior to reactive strategy

Ben Horin S Gastroenterology 2015;148:1268–1281

Drug-Level Based Dosing Versus Symptom-Based Dose Adaptation (TAILORIX)

- Multicenter, double-blind, RCT
- Biologic naïve adults with active CD n=122
  - CDAI >220; CRP >5 and/or FCP >250
  - Endoscopic lesions
- Target for IFX dosing was a trough concentration > 3 ug/ml
- Primary endpoint sustained steroid-free clinical remission from week 22 to 54 AND absence of ulceration at 1 year based on centrally read endoscopies

D’Haens, G R et al DDW 2016
TAILORIX-RESULTS

- Results based on local endoscopy read
- Patients without ulceration at week 54: 36%; 43%; 48% (p=NS)
- Endoscopic remission: CDEIS <3 49%; 51%; 45% (p=NS)

![Graph showing primary endpoint results.]

D’Haens, G R et al DDW 2016

DRUG/ADA STRATEGIES

- Sub Therapeutic Drug Level AND Positive ADA → Persistent disease
- Therapeutic Drug Concentration Or Detectable Trough Level → Change To Another Anti-TNF
- Sub therapeutic Drug Level Or Undetectable Trough Level → Increase Dose Or Frequency
- Persistent disease → Change To Another Mechanism Of Action
- Addition of low dose immunosuppressant

ADA = anti-drug antibody

TAKE HOME POINTS

• Strong evidence that therapeutic drug monitoring should be used in the setting of secondary loss of response
• Optimal targets of drug levels for various clinical endpoints evolving
• Proactive TDM is evolving may improve goal of reduction of inflammation and cost reduction

THANK YOU
A 19 y o college student is referred to your office for evaluation for intermittent abdominal pain. Her cbc, cmp, esr, crp are normal. She denies a family history of IBD. Fecal calprotectin is >100 µg/g. Her serology panel shows ASCA -/pANCA -. A colonoscopy with TI intubation is normal. CT enterography was also negative. What is the next step in the management of this patient?

a) Watchful waiting  
b) Check other serologies associated with CD  
c) Start treatment for CD  
d) Test patient for celiac disease

A. Watchful waiting is inappropriate in this scenario.  

B. While other serologies can be positive in ASCA negative patients, they are unlikely to add benefit in this case since CD is unlikely to be the diagnosis.  

C. Elevated fecal marker alone is not diagnostic of IBD.  

D. This is the correct answer. Recognize that calprotectin can be elevated in other conditions besides inflammatory bowel disease such as celiac disease.

Reference:  
EVALUATING DISEASE ACTIVITY

• Positive correlation between fecal biomarkers and endoscopic disease activity
• Fecal biomarkers useful in patients who do not show elevated Crp
• Higher correlation with endoscopic activity
  – colonic disease > ileal disease

DISTINGUISHING IBD VS. NON IBD

• Fecal lactoferrin and fecal calprotectin superior to CRP in differentiating active IBD with inactive IBD and IBS.
• CRP≤ 0.5 or Calprotectin <40 µg/g had less than 1% chance or less of IBD