

# [S1419] A COMPARISON AMONG FOUR NEUTROPHIL-DERIVED PROTEINS IN FECES AS INDICATOR OF DISEASE ACTIVITY IN ULCERATIVE COLITIS

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## BACKGROUND

Neutrophil-derived proteins in feces as an indicator of intestinal inflammation have been suggested as possible biomarkers for assessing disease activity in inflammatory bowel disease (IBD). Lactoferrin (Lf), Calprotectin (Cal), PMN-Elastase (PMN-e) and Lysozyme (Lys) are proteins found in activated neutrophils and have been shown to increase in feces during active IBD.

## AIM

To evaluate the diagnostic utility of measuring fecal concentrations of Lf, Cal, PMN-e and Lys as indicators of disease activity in patients with active and inactive ulcerative colitis (UC).

## METHODS

A total of 65 fecal specimens were collected over a 5-month period from 30 UC patients during times of active and inactive status of disease. The patient population had an age range of 25 to 62 yr with a male to female ratio of 1:2.

Disease activity was determined using the Colitis Activity Index (CAI - Rachmilewitz-Index) that includes a combination of lab parameters and symptoms with a score of >5 indicating active disease.

There were 22 active and 35 inactive time-points for the analysis. There were a total of 8 discrepant specimens that had elevated levels for all 4 proteins and a CAI range of 1 to 5, one specimen had normal levels for all 4 proteins and a CAI of 7.

Fecal Lf, Cal, PMN-e and Lys were measured by ELISA and reported as  $\mu\text{g/g}$  feces. Levels of  $\geq 7.25$ ,  $\geq 6.00$ ,  $\geq 0.062$  and  $\geq 0.60$  for Lf, Cal, PMN-e and Lys, respectively were considered elevated.

## RESULTS

**Table. 1:**

**Patient characteristics**

|  |                  |
|--|------------------|
| <b>Male : Female ratio</b>   | <b>10:20</b>     |
| <b>Age (in years; mean, SEM)</b>   | <b>42.7+/-10</b> |
| <b>Age of first manifestation (in years; mean)</b>                           | <b>29.40</b>     |
| <b>Age of diagnosis (in years; mean)</b>                                     | <b>32.4</b>      |
| <b>Disease spread: pancolitis (% , N)</b>                                    | <b>23.3 (7)</b>  |
| <b>Extraintestinal manifestations during exacerbation (lifetime) (% , N)</b> |                  |
| - Arthritis  | 20.0 (6)         |
| - Osteoporosis   | 10.0 (3)         |
| - PSC  | 3.3 (1)          |
| <b>Actual Medications (% , N)</b>  |                  |
| - Corticosteroids  | 0.0 (0)          |
| - 5-ASA tablets  | 50.0 (15)        |
| - Probiotics   | 13.3 (4)         |
| - Iron sulfate   | 3.3 (1)          |

**Table. 2:**

| <b>Rachmilewitz-Index</b>             | <b>Clinical activity index</b>              | <b>Score</b>   |
|---------------------------------------|---|----------------|
| <b>Bowel frequency</b>                | <b>&lt; 18; 18 to 35; 36 to 60; &gt; 60</b> | <b>0-1-2-3</b> |
| <b>Blood in stool</b>                 | <b>no - little - severe</b>                 | <b>0-2-4</b>   |
| <b>well being</b>                     |   | <b>0-1-2-3</b> |
| <b>Abdominell pain</b>                | <b>no - little - moderate - severe</b>      | <b>0-1-2-3</b> |
| <b>Fever</b>                          | <b>&lt; 38°C - &gt;38°C</b>                 | <b>0-3</b>     |
| <b>extraintestinal signs</b>          | <b>iritis - erythema nodosa - athritis</b>  | <b>0-3-3-3</b> |
| <b>Erythrocyte sedimentation rate</b> | <b>&gt; 50mm/h - &gt; 100mm/h</b>           | <b>0-1-2</b>   |
| <b>Haemoglobin</b>                    | <b>hb &lt; 100g/l</b>                       | <b>4</b>       |

CAI range of 1 to 5 indicates no disease activity, range > 5 indicates active disease.

| CAI                | 22 active    | 21 active *<br>1 discrepant | 43 inactive  | 35 inactive **<br>8 discrepant | all pts     | pts without<br>discrepant |
|--------------------|--------------|-----------------------------|--------------|--------------------------------|-------------|---------------------------|
|                    | sensitivity  |                             | specificity  |                                | correlation |                           |
| <b>Lactoferrin</b> | <b>86.4%</b> | <b>90.5%</b>                | <b>69.8%</b> | <b>85.7%</b>                   | <b>0.75</b> | <b>0.86</b>               |
| patients           | 19/22        | 19/21                       | 30/43        | 30/35                          |             |                           |
| >= 7.25 µg/ml      |              |                             |              |                                |             |                           |
| mean               | 253.00       | 264.99                      | 4.72         | 22.66                          |             |                           |
| standard error     | 83.38        | 86.39                       | 8.11         | 2.08                           |             |                           |

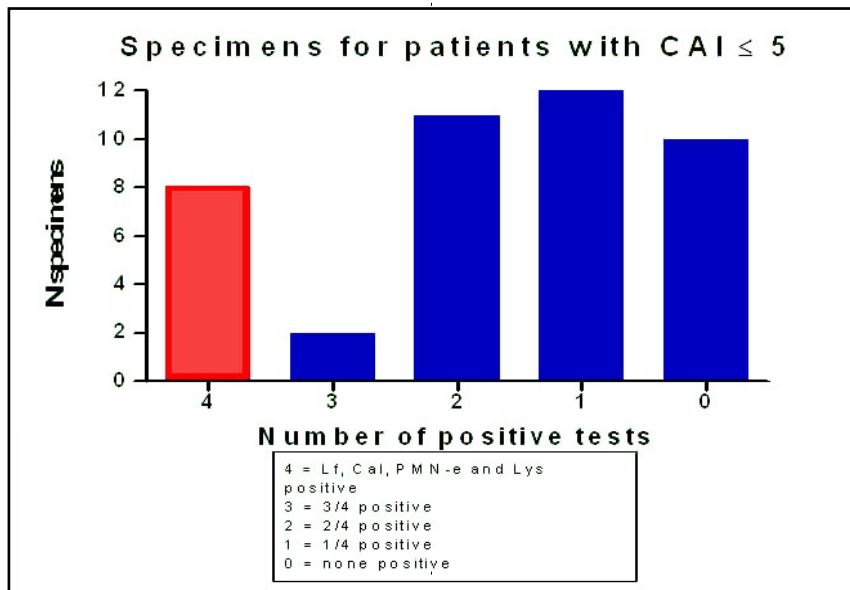
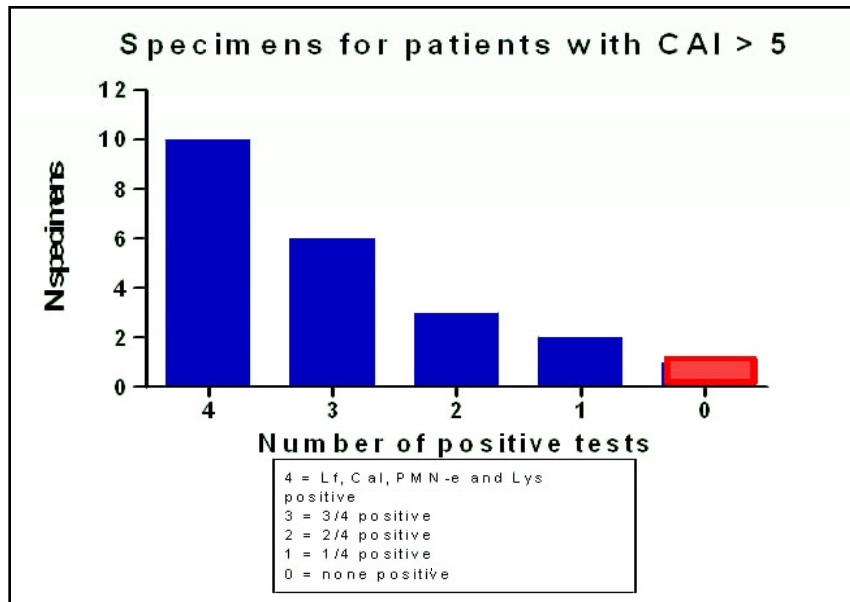
| CAI                 | 22 active    | 21 active *<br>1 discrepant | 43 inactive  | 35 inactive **<br>8 discrepant | all pts     | pts without<br>discrepant |
|---------------------|--------------|-----------------------------|--------------|--------------------------------|-------------|---------------------------|
|                     | sensitivity  |                             | specificity  |                                | correlation |                           |
| <b>PNM-Elastase</b> | <b>81.8%</b> | <b>85.7%</b>                | <b>48.8%</b> | <b>60.0%</b>                   | <b>0.60</b> | <b>0.68</b>               |
| patients            | 18/22        | 18/21                       | 21/43        | 21/35                          |             |                           |
| >= 0.062 µg/ml      |              |                             |              |                                |             |                           |
| mean                | 0.92         | 0.96                        | 0.13         | 0.25                           |             |                           |
| standard error      | 0.19         | 0.20                        | 0.07         | 0.03                           |             |                           |

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|---------------------|--------------|-----------------------------|--------------|--------------------------------|-------------|---------------------------|
|                     | sensitivity  |                             | specificity  |                                | correlation |                           |
| <b>Calprotectin</b> | <b>72.7%</b> | <b>76.2%</b>                | <b>60.5%</b> | <b>74.3%</b>                   | <b>0.65</b> | <b>0.74</b>               |
| patients            | 16/22        | 16/21                       | 26/43        | 26/35                          |             |                           |
| >= 6.0 µg/ml        |              |                             |              |                                |             |                           |
| mean                | 58.43        | 61.20                       | 8.53         | 18.52                          |             |                           |
| standard error      | 16.90        | 17.45                       | 18.18        | 3.08                           |             |                           |

| CAI             | 22 active    | 21 active *<br>1 discrepant | 43 inactive  | 35 inactive **<br>8 discrepant | all pts     | pts without<br>discrepant |
|-----------------|--------------|-----------------------------|--------------|--------------------------------|-------------|---------------------------|
|                 | sensitivity  |                             | specificity  |                                | correlation |                           |
| <b>Lysozyme</b> | <b>54.5%</b> | <b>57.1%</b>                | <b>48.8%</b> | <b>60.0%</b>                   | <b>0.55</b> | <b>0.63</b>               |
| patients        | 13/22        | 13/22                       | 21/43        | 21/35                          |             |                           |
| >= 0.6 µg/ml    |              |                             |              |                                |             |                           |
| mean            | 2.79         | 2.92                        | 0.88         | 1.69                           |             |                           |
| standard error  | 0.98         | 1.02                        | 0.43         | 0.18                           |             |                           |

\* 1 discrepant specimen that had normal levels for all 4 proteins and a CAI >5

\*\* 8 discrepant specimens that had elevated levels for all 4 proteins and a CAI range of 1 to 5



## CONCLUSIONS

Even though fecal ELISA tests do not replace the traditional methods for assessing IBD patients, these noninvasive and rapid assays can serve as a useful tool in combination as determined by the Gastroenterologist.

Among the neutrophil derived proteins, Lactoferrin had the highest sensitivity, specificity and correlation with disease activity for UC patients. Fecal lactoferrin is useful for assessing disease activity in inflammatory bowel disease.

## LITERATURE

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