Rapid microbial diagnosis of acute gastroenteritis: comparison of 3 multiplexed molecular tests on 194 pediatric stool samples. Are culture methods obsolete?

Alexandra BEUCHER1, Nicolas VERNET2, Guillaume MACCIO3, Paul O. VERHOEVEN3, Jean Michel ROQUE2, Amelie EPERCIEUX3, Yahia MEKKI1, François VANDENESCH1, Anne Marie FREYDIERE1, Olivier DAUWALDER1

1 Laboratoire de Bactériologie 2 Laboratoire de Virologie, Centre de Biologie et de Pathologie Est, Hospices Civils de Lyon, Bron, France, 3Laboratoire de Bactériologie-Virologie-Hygiène, Hôpital Nord, CHU de Saint Etienne, Saint Etienne, France.

Comparison of 3 multiplexed molecular tests [MMT] : CLART-Enterobact® (Genomica) [GEN], XTAG-GP® [Luminex] [LUM], Enteric Bacterial Panel® (BD) [EBP] and Enteric Virus Panel® (Diagnenode) [EVP] using a syndrome-based approach for the diagnosis of bacterial and viral acute gastroenteritis on a pediatric stool collection.

Objectives

Comparison of 3 multiplexed molecular tests [MMT] : CLART-Enterobact® (Genomica) [GEN], XTAG-GP® [Luminex] [LUM], Enteric Bacterial Panel® (BD) [EBP] and Enteric Virus Panel® (Diagnenode) [EVP] using a syndrome-based approach for the diagnosis of bacterial and viral acute gastroenteritis on a pediatric stool collection.

Material and Methods

Bacteriological methods
Conventional cultural methods and the European two steps algorithm combining immunological and molecular tests for the detection of toxigenic Clostridium difficile.

Viroligical routine methods
Immunological and molecular tests (ribopharm)

CLART-Enterobact® (Genomica)
XTAG-GP® (Luminex)
Bacterial Panel® (BD)
Virus Panel® (Diagnenode)

DNA/RNA Extraction with the Easy Mag® (bioMérieux) Automated DNA/RNA extraction

Micro-array
RT-PCR + hybridation
Full automated RT-PCR

15 pathogens
Bacterial pathogens
Campylobacter spp., C. difficile ToxA/B, E. coli O157, E. coli enteropathogenic LT/ST (ETEC), E. coli stx1/2, Salmonella spp, Shigella spp., Vibrio cholerae, Yersinia enterocolitica

15 pathogens
Bacterial pathogens
Campylobacter coli/C. jejuni, E. coli stx1/2, Salmonella spp., Shigella spp.

4 bacterial pathogens
Campylobacter coli/C. jejuni

2 viral pathogens
Norovirus I/II
Rotavirus

Time: 4-5 h
Time: < 5 h
Time: 2 h
Time: 2 h

Cost: 40-50€/test
Cost: 50€/test
Cost: 30€/test
Cost: 30€/test

Table 1. Results of the 3 MMT evaluated with 194 pediatric stool samples

<table>
<thead>
<tr>
<th>Test</th>
<th>Parameters</th>
<th>Toxicogenic C. difficile</th>
<th>Campylobacter (n=18)</th>
<th>Salmonella (n=13)</th>
<th>Shigella (n=6)</th>
<th>Norovirus (n=21)</th>
<th>Rotavirus (n=30)</th>
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<tbody>
<tr>
<td>Genomica</td>
<td>Assay</td>
<td>1st (n=3) 2nd</td>
<td>1st (n=3) 2nd</td>
<td>1st (n=3) 2nd</td>
<td>1st (n=3) 2nd</td>
<td>1st (n=3) 2nd</td>
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<tr>
<td>CLART</td>
<td>Se (%)</td>
<td>100 100</td>
<td>100 100</td>
<td>94.4</td>
<td>83.3 84.6</td>
<td>100 100</td>
<td>100 100</td>
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<tr>
<td>Enterobact®</td>
<td>Spe (%)</td>
<td>98.7 99.5</td>
<td>98.6 98.3</td>
<td>99.3 100</td>
<td>100 100</td>
<td>100 100</td>
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</tr>
<tr>
<td>[GEN]</td>
<td>PVP (%)</td>
<td>50.0 75.0</td>
<td>87.7 85.0</td>
<td>90.9 100</td>
<td>100 100</td>
<td>100 100</td>
<td>100 100</td>
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<tr>
<td></td>
<td>NPV (%)</td>
<td>100 100</td>
<td>100 100</td>
<td>99.4</td>
<td>98.6 98.9</td>
<td>100 100</td>
<td>100 100</td>
</tr>
<tr>
<td>Luminex</td>
<td>In (n)</td>
<td>35 0 35</td>
<td>35 0 35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
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<tr>
<td>XTAG</td>
<td>Se (%)</td>
<td>66.7 100</td>
<td>100 100</td>
<td>100 100</td>
<td>61.5 92.3</td>
<td>100 100</td>
<td>100 71.4</td>
</tr>
<tr>
<td>GPP® (LUM)</td>
<td>Spe (%)</td>
<td>100 100</td>
<td>96.6 100</td>
<td>73.9 98.3</td>
<td>93.6 100</td>
<td>100 100</td>
<td>100 88.3</td>
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<td></td>
<td>PPV (%)</td>
<td>100 100</td>
<td>75.0 100</td>
<td>14.5 80.0</td>
<td>33.3 100</td>
<td>100 100</td>
<td>52.5 95.2</td>
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<tr>
<td></td>
<td>NPV (%)</td>
<td>100 100</td>
<td>100 100</td>
<td>96.4 99.4</td>
<td>100 100</td>
<td>96.7 96.6</td>
<td>94.1 94.2</td>
</tr>
<tr>
<td></td>
<td>In(n)</td>
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<td>1 0 1</td>
<td>1</td>
<td>1</td>
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<td>1</td>
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<tr>
<td>BD Max</td>
<td>Se (%)</td>
<td>NI NI 88.9 88.9</td>
<td>84.6 84.6</td>
<td>100 100</td>
<td>81.0*</td>
<td>55.2*</td>
<td>55.2*</td>
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<tr>
<td>EBP® &amp; EVP®</td>
<td>Spe (%)</td>
<td>NI NI 99.4 99.4</td>
<td>100 100</td>
<td>100 100</td>
<td>97.7*</td>
<td>97*</td>
<td>97*</td>
</tr>
<tr>
<td>[BD]</td>
<td>PPV (%)</td>
<td>NI NI 94.1 94.1</td>
<td>100 100</td>
<td>100 100</td>
<td>81.0*</td>
<td>76.2*</td>
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<tr>
<td></td>
<td>NPV (%)</td>
<td>NI NI 98.9 98.9</td>
<td>98.9 98.9</td>
<td>100 100</td>
<td>97.7*</td>
<td>92.4*</td>
<td>92.4*</td>
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<tr>
<td></td>
<td>In(n)</td>
<td>/ / 3</td>
<td>3 0 3</td>
<td>0 **</td>
<td>0 **</td>
<td>0 **</td>
<td>0 **</td>
</tr>
</tbody>
</table>

Comparison with the bacterial gold standard [BGS] based on the positivity of bacteriological routine methods and/or the positivity of 2 on the 3 evaluated MMT

Comparison with virus gold standard [VGS] based on the positivity of one immunological and/or molecular tests used in routine practice

Discussion- Conclusion

This first frontline approach with MMT approves their utility compared to conventional methods allowing a significant reduction of the time to diagnosis of pathogen and, if positive, a targeted bacterial culture. The MMT use could guide, if required, antibiotic prescription which must be administrated early according to European pediatric guidelines. However, this study point out the importance of the extraction step (GEN) or the strength of the experimental protocol (LUM) and undergoes some sensitivity defects for virus and Salmonella suggesting that optimization is required before stopping bacteriological culture definitely.

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