Acute Infectious Gastroenteritis

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Acute infectious gastroenteritis (AGE) is defined as a diarrhoeal disease of rapid onset presenting with the incidence of three or more soft or liquid stools, or three bouts of vomiting per 24 h, with addition of abdominal pain, or fever.

acute infectious gastroenteritis

aetiological agents biomarkers

1. Causative Agents of AGE

1.1. Rotavirus

Rotavirus is the most common agent causing AGE in children worldwide ^{[1][2][3][4][5][6]}. It has been suggested to be one of the most pathogenic causative agents in AGE which correlates with infectious diarrhoea, vomiting, and higher frequency of fever ^{[4][7][8][9]}. It was mostly reported among children aged 0–14 years old, with the highest incidence among children less than 5 years ^{[5][6][7][10][11][12]}. The prevalence was highly reported during the dry season ^{[6][12]}, bottle-fed children ^{[6][13][14]}, and children with group A blood type ^{[6][15][16]}. Moreover, laboratory investigations revealed a significantly high level of serum transaminase among children infected with rotavirus ^[17] ^{[18][19]}. Among many studies conducted worldwide, it was found that rotavirus infected children have a high chance of developing dehydration as a complication, and thus, must be closely monitored ^{[20][7][21][22]}.

1.2. Norovirus

Norovirus is an emerging cause of acute gastroenteritis, responsible for approximately 17–18% of all acute gastroenteritis cases worldwide, especially among developed countries ^{[23][24][25][26][27]}. The age group under 1 year old has the highest frequency of norovirus infection with additional risk factors which include male sex ^[26]. Norovirus is detected throughout the year, with the autumn and winter seasons observed to report a higher frequency of cases ^{[23][28][29]}; the detection rate of norovirus cases correlates positively with humidity ^[29]. Co-infection with rotavirus ^{[30][31]}, astrovirus ^[32], and *Salmonella* ^[31] may occur in certain cases. Norovirus infections are observed to cause more severe symptoms of gastroenteritis in children compared with rotavirus, especially after the introduction of the rotavirus vaccination period ^[33]. Individuals with AB blood group have less susceptibility towards GII.4 noroviruses, whereas those with the O blood group are more susceptible to GI.4, GII.4, GII.17, and GII.18-Nica viruses ^[34]. GII.4 is the most common norovirus genotype causing infection in children ^{[26][35][36]}. However, recent epidemiological data observed that GII.2[P16] is currently an emerging norovirus strain in East Asia and Europe ^[37]. Norovirus-infected patients are more likely to have a longer duration of infection and more

frequent vomiting in a day, but less likely to report fever in comparison with other causes of infective gastroenteritis [38].

1.3. Astrovirus

With an astrovirus-causing acute gastroenteritis' global average incidence of 11%, the highest prevalence of human astrovirus (HAstV) infections was in the group of children between 37 and 48 months old ^{[39][40]}. HAstV infection mainly occurs during the dry season in the African continent; meanwhile, the highest occurrence reported in the tropical areas is often in the rainy season and winter season in temperate climate countries ^{[39][41]}. Patients usually manifested with diarrhoea, fever, vomiting, and abdominal pain ^{[40][42]}.

1.4. Enteric Adenovirus Serotypes 40 and 41

Between 2.3 and 5% of the diarrhoea cases in children were caused by adenoviruses (AdV) serotypes 40 and 41 ^[43]. Enteric adenovirus serotypes 40 and 41 account for both sporadic or epidemic gastroenteritis in infants and young children, which are detected throughout the year with a summer peak between May and July ^{[43][44][45]}. Those infected with this pathogen will acquire mild fever and vomiting, abdominal pain, bloody diarrhoea, respiratory symptoms such as cough, and/or secondary lactose malabsorption ^{[45][46]}. Patients with adenovirus gastroenteritis have a significantly higher CRP (mean 3.4 mg/dL) and ESR value (mean 24 mm/h) compared with other gastroenteritis pathogens, and pulmonary-associated symptoms and vomiting frequency are increased in patients with this infective gastroenteritis ^[47]. Intravenous cytosine nucleotide analogue (CDV) that inhibits DNA polymerase has the highest in vitro activity against adenovirus, and is the preferred therapeutic agent. Elevation in lymphocyte counts (specifically CD4) were correlated with the clearance of AdV infection and improvement in survival ^{[48][49]}.

1.5. Salmonella

Salmonella has become a major foodborne pathogen across the globe, causing about 3.4 million cases and 681,316 annual deaths, with 63.7% of cases occurring in children under 5 years of age ^{[50][51][52]}. There were at least 150 non-typhoidal *Salmonella* serotypes that can cause gastroenteritis, with *Salmonella* Typhimurium and *Salmonella* Enteritidis being the most common serotypes ^{[51][53][54]}. A study in Greece recorded that the highest rate of infection was in August, with infants being the most vulnerable group ^[55]. The contributing factor of salmonellosis was mainly related to consumption of contaminated food and poor clean water supply ^{[56][57]}. Salmonellosis can cause the increase in C-reactive protein values (CRP), erythrocyte sedimentation rate (ESR) and body temperature ^[58]. In terms of management, the recommended empiric parenteral therapy includes cefotaxime or ceftriaxone, whereas oral therapy includes amoxicillin, trimethoprim-sulfamethoxazole, or azithromycin. *Salmonella* isolates have a high resistance towards at least one antimicrobial agent ^{[53][59]}, especially towards clindamycin, oxacillin, penicillin, and vancomycin, thus antibiotic susceptibilities of *Salmonella* must be determined for the targeted antibiotic therapy ^[54].

1.6. Escherichia coli

The WHO Global Burden of Foodborne Diseases report estimates that more than 300 million illnesses and nearly 200,000 deaths are caused by diarrheagenic *Escherichia coli* (DEC) globally each year. DEC is one of the major causal agents of diarrhoea in children under 5 years of age in developing countries ^{[60][61][62]}, whereas children under 2 years of age are at the highest risk of infection with Enteropathogenic *Escherichia coli* ^{[63][64]} and *Escherichia coli* O157:H7 ^{[63][65]}. However, the major cause of paediatric infections in certain areas such as Ahvaz, Iran, were non-O157:H7 *E. coli* ^[66]. The incidence of non-O157 Shiga Toxin-producing *Escherichia coli* has been increasing in recent years, including those caused by serotypes O26, O45, O103, O111, O121, and O145 ^[67]. Co-infections can occur especially between EPEC and *Campylobacter* spp. ^[68]. Interestingly, Enteropathogenic *Escherichia coli* cases are less frequently detected in Malaysia and its similar geographical/climatic areas, in comparison with a country such as Iran where it has been reported that acute gastroenteritis was greatly caused by this strain ^[69]. Most DEC are sensitive to ciprofloxacin and the empirical antibiotic of choice ^{[70][71]}.

1.7. Entamoeba histolytica

Other than viruses and bacteria, parasites such as *Entamoeba histolytica* also play a role in causing acute gastroenteritis in children ^{[72][73][74]}. Children infected with *Entamoeba histolytica* were mostly presented with tenesmus ^{[73][74][75]}, fever ^{[73][74]}, vomiting ^{[73][76]}, abdominal cramps ^{[73][75][76]}, and bloody diarrhoea ^{[77][74][75]}. Through laboratory investigations, it was found that infection with *Entamoeba histolytica* results in leukocytosis ^[72] ^{[73][76][78]}, elevation of CRP ^{[66][70][71][72][76][78]}, elevation of ESR ^[73], elevation of serum alkaline phosphatase, and serum transaminase levels ^{[73][79]}. Eating unwashed or raw vegetables ^{[79][80][81][82]} and poor water hygiene ^{[74][83]} ^[84] were identified to increase the risk of *Entamoeba histolytica* infection.

2. Biomarkers in the Detection of Common Aetiological Agents of AGE

2.1. Rotavirus

As the most common inflicted pathogen causing AGE in children, several biomarkers have been developed and available commercially for the detection of rotavirus. The widely available biomarker platform utilized is the enzyme immunoassay (EIA) to screen for the rotavirus antigen ^{[1][2][4][6][10][13][17][85][86][87]}. Amongst EIA kit used were Premier Rotaclone, Meridian Bioscience Inc., Cincinnati, OH, USA ^{[1][2][3][10][86][87]}, RIDASCREEN Rotavirus R Biopharm AG, Darmstadt, Germany ^{[6][86]}, and ProSpect Rotavirus Test, Oxoid Ltd., UK ^{[13][86]}. Rotavirus antigens can also be detected by using enzyme-linked immunosorbent assay (ELISA) ^{[3][7][9][11][19][22]} which includes ELISA kits such as Premier Rotaclone, Meridian Bioscience, Inc. ^[3], Fecal Rotavirus Antigen ELISA Kit (EDI, CA, USA) ^[7], ProSpecTM Rotavirus Microplate Assay, Oxoid ^[11] and Rota Antigen Test Device, Cambridge ^[19]. ELISA can also be used in the detection of rotavirus-specific IgM ^[21]. Other methods in detection of rotavirus antigen were immunochromatography ^{[16][18]} and latex agglutination ^{[14][85][88]}. In addition, polyacrylamide gel electrophoresis (PAGE) was carried out to determine the electropherotype of rotavirus strains ^{[1][10][85]}. Moreover, samples that were rotavirus positive for ELISA or EIA were sent for genotyping using reverse-transcription polymerase chain

reaction (RT-PCR) to determine whether they belong to particular G/P genotypes ^{[2][7][10][11][16][21][89][90]}. A multiplex real-time RT-PCR is an advanced approach for a high-throughput rotavirus genotype characterization for monitoring circulating rotavirus wild-type strains, which is more robust in identifying a novel strain ^[91].

2.2. Norovirus

Viral RNA and antigens are the main biomarkers for detection of norovirus infection. Real-time quantitative polymerase chain reaction, Multiplex Gastrointestinal Platforms, enzyme immunoassays (EIAs) and genotyping are the main methods used in laboratory diagnosis of norovirus ^[92]. Cepheid Xpert[®] Norovirus kit automates sample processing, nucleic acid extraction, and real-time reverse transcription polymerase chain reactions (RT-PCRs) for detection and differentiation of norovirus GI and GII, which account for the majority of norovirus infections worldwide ^{[93][94]}. Another real-time PCR platform, RIDA[®]GENE Norovirus, can also be used as an alternative in detecting the virus ^[95]. Although PCRs are highly sensitive and specific, they are expensive and require specialized techniques and equipment. Rapid diagnostic tests are usually carried out during outbreak screening and patient management, which include immunochromatographic test, enzyme immunoassay (EIA), enzyme-linked immunosorbent assay (ELISA) and fluorescence immunoassay (FIA). RIDA[®]QUICK Norovirus (R-Biopharm AG, Darmstadt, Germany) is one of the most used rapid immunochromatographic tests to detect norovirus ^{[96][97][98]}. RIDASCREEN[®] Norovirus 3rd Generation is an example of ELISA that is still currently in use ^{[101][102]}. For FIA, the Automated Fluorescent Immunoassay System NORO (AFIAS-Noro) assays (Boditech Med Inc, Gangwon-do, South Korea) are newly developed diagnostic tests for norovirus infections ^[88].

2.3. Astrovirus

Stool samples can be investigated using RT-PCR to detect the presence of human astrovirus (HAstV) ^{[39][40][47]}. One of the available kits is the RT-PCR Luminex Assay, with which a portion of the ORF2 capsid region is targeted by using a set of specific reverse primers labeled with biotinTEG at 5'-ends and specific probes sequences ^[103]. A one-step, accelerated, real-time RT-LAMP (rRTLAMP) assay can also be used by targeting the 5'-end of the capsid gene for rapid and quantitative detection of HAstV ^{[104][105]}.

2.4. Enteric Adenovirus Serotypes 40 and 41

Adenovirus antigen and hexon-coding gene in enteric adenovirus serotypes 40 and 41 can be recognised and used for screening and detection methods of this pathogenic agent infection. BioNexia RotaAdeno and RIDA Quick Rota-Adeno-Combi R-Biopharm, which are the immunochromatographic tests (ICT) and LIAISON Adenovirus chemiluminescence immunoassays (CLIA) can be utilised to detect enteric adenovirus antigen ^{[106][107]}. The samples from ICT and LIAISON CLIA can subsequently undergo RT-PCR for genotyping of hexon-coding genes by using a specific TaqMan Array Card, which is a 384-well singleplex real-time PCR format that has been recognised to detect multiple infection targets ^{[108][109]}.

2.5. Salmonella

As for *Salmonella* infection, the virulence genes include flagellin (*filcC*), *invA*, *invF*, *sitC*, *hilAgene*, *sipC*, *sipF* genes as well as heat stable enterotoxin gene, *parE* ^[110]. Multiplex real-time PCR such as RIDA GENE-gastrointestinal kits, EntericBio real-time Gastro Panel I, and Seeplex Diarrhea ACE detection has allowed a more rapid detection of multiple targets in a short period of time and some of the tests was followed by hybridisation to microarray/macroarray to achieve multiparametric detection of AGE aetiological agents ^[111]. In terms of sensitivity and specificity, RIDA GENE-gastrointestinal kits were reported to have 25% sensitivity and 99.7% specificity ^[112] ^{[113][114]}. As for EntericBio real-time Gastro Panel I, its sensitivity and specificity were much higher, which were 100% and 97.8%, respectively ^[113]. Seeplex Diarrhea ACE has a sensitivity of 40–100% and specificity of 96–100% ^{[112][115][116]}.

2.6. Escherichia coli

Most of the current omics approaches were focused on the detection of Shiga Toxin-producing *Escherichia coli* (STEC) infections. This is due to the fact that accurate diagnosis of STEC infection is very crucial because appropriate early treatment decreases the risk of serious complications and improves overall patient outcome, especially in children ^{[117][118]}. Although non-O157 serotypes account for the majority of STEC infections, they are significantly under-reported because frontline microbiology laboratories mainly focus on the detection of O157 STEC using specific agar-based methods ^[119]. CHROMagar STEC is a new chromogenic medium invented to improve detection of STEC, which detects O157 and non-O157 STEC through a chromogenic substrate ^{[119][120]}. However, PCR is a more sensitive test than culture ^{[119][120]}. RIDA[®]GENE real-time PCR kits EAEC, EHEC/EPEC, and ETEC/EIEC (R-Biopharm, Darmstadt, Germany) all can be used to detect the *aatA* and *aggR*, *eae*, and *elt* and *estA* genes of Enteroaggregative, Enteropathogenic, and Enterotoxigenic *Escherichia coli*, respectively ^[121]. These will ensure that different pathotypes of diarrheagenic *Escherichia coli* can be detected and differentiated successfully.

2.7. Entamoeba histolytica

Entamoeba histolytica infection can cause a significant decrease in serum leptin and has been suggested to be a potential biomarker for this pathogenic infection ^[122], which can be detected using the ELISA technique (kit supplied by RayBiotech.Inc, Guangzhou, China). Moreover, there was also a marked increase in HDL, obestatin, calprotectin and SIgA concentration level with a concurrent decrease in cholesterol, triglyceride, LDL and VLDL concentration levels. All of these serums can be analysed and measured using ELISA techniques ^[123]. ELISA (Techlab II *Entamoeba histolytica*) can also be used to detect *Entamoeba histolytica* antigen presented in stool with a specificity of 100% but with a low sensitivity of 19.2% ^[124]. Thus, multiplex PCR is a more favourable option as it has sensitivity of 100% with specificity of 95.8% ^[125]. During progression of amoebiasis, a small non-coding RNA known as microRNA (miRNA) is involved in promoting apoptosis in epithelial colon cells and comprehensive profiling of miRNA using Taqman Low-Density Arrays showed a significant interaction between miRNA and parasite presented ^[126]. Taqman Low-Density Arrays has a sensitivity of 92% and a specificity of 100% in diagnosis of *Entamoeba histolytica* infection ^[127].

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