ORIGINAL ARTICLE

NEONATAL GASTROINTESTINAL MUCORMYCOSIS (GIMM), A MIND-BOGGLING PRESENTATION IN LMIC

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ABSTRACT

Background: Gastrointestinal Mucormycosis (GIMM) is a rare infection acquired by ingesting contaminated products. It primarily affects the stomach, followed by the colon and ileum. Premature and low birth weight neonates are explicitly susceptible. Contaminated nasogastric tubes, endotracheal tubes, wooden tongue depressors, the warm, humid environment of the Intensive Care Units (ICU), contaminated enteral feeds, and parenteral infusions are considered culprits in a hospital. The most important differential diagnosis of MM is necrotizing enterocolitis. The gold standard methods to diagnose Mucormycosis are histopathology, culture & sensitivity, and molecular testing in high-risk patients.

Material and Methods: It is a retrospective descriptive study conducted from January 2015 to January 2016 at Children's Hospital Lahore. All neonates who presented with intolerance to feed, abdominal distension, failure to pass meconium, respiratory distress, and pneumoperitoneum needing abdominal surgery were included. Per-operatively specimens were obtained like gangrenous gut resections or serial six biopsies in case of suspected Hirschsprung disease. An end-to-end anastomosis or an ileostomy stoma was performed depending on the patient's clinical condition.

Results: Of the 121 neonates, enrolled preterm were (55.3%) and low birth-weight males were (67.7%). They presented with abdominal distension (100%), and had (51.2%) pneumoperitoneum. Most neonates were suspected of having necrotizing enterocolitis (NEC) and showed dilated bowel loops on the x-ray abdomen (93.3%).After surgeries, two complications, short gut syndrome (8.2%) and intolerance to feed (13.2%), were seen in these patients. Three pre-term neonates suspected clinically as NEC were found to have GIMM. The histopathology report showed numerous broad aseptate fungal hyphae with extensive necrotizing inflammation and infarction of the bowel wall.

Conclusion: GIMM clinically mimics NEC and should be suspected in any neonate, in the absence of pneumatosis intestinalis on abdominal X-ray and no response to antibacterial therapy. The timely diagnosis and rapid management can subsequently decrease the mortality.

Keywords: Mucormycosis, Necrotizing enterocolitis, Neonates

BACKGROUND

The prevalence of Mucormycosis (MM) in neonates is 0.14% in India; the global annual prevalence varies greatly. Usually, MM presents in diabetic people, leukemic, post tuberculous, renal failure, critically ill bedridden patients, or solid organ transplant, and hematopoietic stem cell transplant patients. Recently it is also documented in COVID-19 patients. Hot, humid environments of tropical and subtropical areas favor the growth of Mucorales. MM produce widely disseminated infection in rhino-orbital-cerebral, pulmonary, gastrointestinal, and cutaneous forms. Gastrointestinal mucormycosis (GIMM) is a rare infection; acquired by ingesting fermented milk, dried bread, fermented porridges, spore-contaminated herbals, and homeopathic remedies.[1-4]

GIMM primarily affects the stomach, followed by the colon and ileum; other sites like the liver, spleen, and pancreas have also been reported. Emphysematous gastritis, mural thickening, and air-containing collections in small and large bowel walls are commonly been seen on imaging. Among these, colonic MM has the highest mortality rate. MM invades the arterial wall causing thrombus formation and subsequent transmural ischemic necrosis of the gut segment, bowel infarction, perforation, and peritonitis. Histologically, there are broad aseptate ribbon-like, branching at right angles hyphae are also found Fig1 E, F. Angioinvasion is MM's characteristic feature leading to vascular thrombosis. MM can also lead to sepsis and multi-organ failure.[5-9]

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This article can be sited as: Ashraf M, Sahrish F, Akram A, Ahmad A, Arshad S, Rasul AA. Neonatal gastrointestinal mucormycosis (GIMM), a mind-boggling presentation in LMIC. Infect Dis J Pak 2022; 33(4): 129-135.

In the neonatal age group, premature and low birth weight neonates are explicitly susceptible to GIMM. The assumed pathophysiology is the ingestion or inhaling fungal spores from the air. In a hospital setting, contaminated nasogastric tubes, endotracheal tubes, wooden tongue depressors, the warm, humid environment of the Intensive Care Units (ICU), contaminated enteral feeds, and parenteral infusions are considered culprits. Early detection, reversal of predisposing factors, and the combination of surgical debridement of affected tissues and antifungal medications are the mainstay of MM. The most important differential diagnosis of MM is necrotizing enterocolitis (NEC). The following gold standard methods used to diagnose MM are Histopathology, Culture & sensitivity, and molecular testing in high-risk patients.[10-12]

MM, formerly known as Zygomycosis, is now placed in the Mucorales order and Mucoromycotina subphylum. MM is one of the 27 species of Mucorales and the 11th genera. The most commonly affecting genus is Rhizopus, followed by Lichthemia; and Mucor. The Mucorales have been widely found in dirt, dung, and putrefied food. The most frequently involved site is the Rhino-cerebral area. However, we report the least frequent type of MM involving the neonatal gastrointestinal tract from the pediatric tertiary care center.[12-14]

The exact incidence and prevalence of GIMM in neonates are unknown; however, increasing incidence and prevalence of MM are reported all around the globe. A global study indicated a higher majority of MM in Europe (34%) than in Asian countries (31%). An umbrella study by Chakrabarti *et al*. showed a 70 % higher prevalence of MM in India. However, data regarding the true prevalence and incidence of GIMM in neonates has yet to be reported to date around the globe. The reporting incidence is increasing due to improvements in diagnostic facilities, as Histopathology is an essential diagnostic tool unavailable in the peripheral center across Pakistan.[15-18] There is minimal epidemiological data on neonatal GIMM from Pakistan; and worldwide. Most data is from case series and reports, not randomized clinical trials. Therefore, this study presents retrospective cases of GIMM in neonates who were clinically suspected of necrotizing enterocolitis at a tertiary care center.

MATERIAL AND METHODS

This was a retrospective descriptive study conducted in pediatric emergency department of Children's Hospital Lahore, January 2015 to December 2016. The sample size was estimated using the sample size formula for estimating proportions. n=Z^2.p.(1-p)/E^2 where Z is 1.96 (95% confidence interval), p=0.05 or 5% estimated prevalence of muormycosis cases and E is the desired margin of error 5%. The sample size needed is 73.

Total 121 neonates with intolerance to feed, abdominal distension, failure to pass meconium, respiratory distress; and pneumoperitoneum were included. An exploratory laparotomy was performed in children with perforation only (51% or 62). Per-operatively different specimens were sent like gangrenous gut resections; serial six biopsies in case of suspected Hirschsprung disease. The autolyzed and mislabeled biopsy specimens with poor morphology were excluded. An end-to-end anastomosis or an ileostomy stoma was performed depending on the patient's clinical condition. The neonates were advised long-term follow-ups for stoma closures, and the development of any subsequent complications i.e., gut atresia, adhesion obstructions, and short gut syndromes. Statistical analysis was done on Statistical Packages of Social Sciences (SPSS) version 23. Qualitative variables like age, gender, clinico-histopathological features were summarized as frequencies and percentages. No statistical association was determined between any variables. This study was duly approved by Institutional Ethical Review Board vide letter # IRB/CHICH/2017/2/17. The detection of MM is a red alert and communicated with the relevant doctor immediately after examining the slides.

RESULTS

The main clinicopathological findings of all neonates are summarized in Table-I. Most of the neonates were males; presented with abdominal distension (100%) followed by pneumoperitoneum (51.2%). The majority of the neonates were preterm (55.3%) and had low birth weight (67.7%).

The majority of neonates were preterm and suspected of NEC, the majority having raised CRP levels. The majority of the patients showed dilated gut loops on the x-ray abdomen (93.3%). The mean age of patients was ten days. After one year, two complications i.e., short gut syndrome (8.2%) and intolerance to feed (13.2%) were seen in these patients.

The detailed clinical; and per-operative findings and outcomes of neonates presented with GIMM are summarized in Table-II. There were three pre-term neonates with a male: female ratio of 2:1. Which

showed GIMM suspected clinically as NEC. The histopathology report showed numerous broad aseptate fungal hyphae with extensive necrotizing inflammation and infarction of the bowel wall. In addition, extensive angioinvasion was found, leading to tissue necrosis and infarction.

Table-I: Main clinicopathological features of neonates presented in surgical emergency.

S #			NEC (72)	HD (49)	Total (121)
1.	Risk factors	Preterm	63(87.5%)	4(8.1%)	67(55.3%)
		Low birth weight	70(97.2%)	12(24.4%)	82(67.7%)
2.	Clinical presentation	Intolerance to feed	69(95.8%)	4(8.1%)	73(60.3%)
		Abdominal distension	72(100%)	41(83.6%)	113(93.3%)
		Constipation	43(59.7%)	49(100%)	92(76.0%)
		Respiratory distress	37(51.3%)	23(46.9%)	60(49.6%)
		Pneumoperitoneum	39(54.1%)	23(46.9%)	62(51.2%)
3.		Abdominal Examination			
	Physical Examination	Distended abdomen	72(100%)	49(100%)	121(100%)
		Shiny abdomen	35(48.6%)	23(46.9%)	58(47.9%)
		Visible veins on the abdomen	35(48.6%)	13(26.5%)	48(39.6%)
		Erythema on the abdomen	17(23.6%)	7(14.2%)	24(19.8%)
4.		Rectal examination			
		Gush of air	0(0%)	27(55.1%)	27(22.3%)
		Gush of stool	0(0%)	33(67.3%)	33(27.2%)
5.	Laboratory investigations	Neutropenia	69(95.8%)	7(16.6%)	76(62.8%)
		Thrombocytopenia	61(84.7%)	17(40.4%)	78(64.4%)
		Raised CRP levels	72(100%)	23(54.7%)	95(78.5%)
		PT APTT deranged	59(81.9%)	13(30.9%)	72(59.5%)
6.	X-Ray Abdomen	Multiple Dilated gut loops	72(100%)	41(97.6%)	113(93.3%)
		Multiple air-fluid levels	69(95.8%)	39(92.8%)	108(89.2%)
		Pneumatosis Intestinalis	37(51.3%)	31(73.8%)	68(56.1%)
		The paucity of air in the pelvis	3(4.1%)	37(88.0%)	40(33.0%)
7.	Management	Surgical exploration	72(100%)	49(100%)	121(100%)
		IV antibiotics	72(100%)	49(100%)	121(100%)
8.	Complications	Intolerance to feed	7(9.7%)	3(6.1%)	10(8.2%)
		Short gut syndrome	9(1.25%)	7(1.4%)	16(13.2%)

Table-II: Main clinicopathological features of neonates presented with GIMM.

S#	Age/ sex	Predisposing factors	Clinical Presentation / Examination	X-ray Examination	Operative-findings	Post-op-findings/ follow up
1.	7-days Pre-Term Low Birth weight	Preterm Low birth weight	Weak cry Severe Respiratory distress Massive Abdominal distension 5th day	Pneumo-peritoneum	A laparotomy was performed through right transverse supraumbilical incision. On exploration, jejunum with a perforation was noted. A jejunal resection and end-to-end jejunal anastomosis with gangrenous gut resection was performed (Figure-1. C)	Shifted to NICU Treated with antibiotics, abdominal distension Death on 2nd postop-day
2.	11 days Pre-Term Low Birth weight	Preterm Low birth weight	Intolerance to feed Moderate Respiratory distress Constipation Massive	Pneumo-peritoneum	A laparotomy was performed through right transverse supraumbilical incision. On exploration, an ileal perforation was noted and end-to-end ileal anastomosis with	Shifted to NICU Treated with antibiotics, abdominal distension Death on 4th postop-day

			Abdominal distension 9th day		gangrenous gut resection was performed (Figure-1D)	
3.	5- days Pre-term Low Birth weight	Preterm Low birth weight	Severe respiratory distress Massive Abdominal distension on 3rd day	Pneumo-peritoneum	A laparotomy was performed through right transverse supraumbilical incision. On exploration, multiple ileal and colonic perforations were noted and end-to-end ileal anastomosis and colostomy with perforated gut resections was performed	Shifted to NICU Treated with antibiotics, abdominal distension Death on 3rd postop-day

Table-III: Comparison of main demographic data of neonates presented with GIM

Sr#	Authors	Type	No. of Neonates	Premature Low birth weight	Duration	Site in GIT	Region	Outcome
1.	Sahrish *et al*.	Original research	Three males	Present	2015-2016	Colon only	Pakistan	Death (3)
2.	Mahajan N *et al*.[6th]	Original research	Eight Male: female 5:3	Preterm (3) Low birth Weight (6)	2022	Small bowel and Large bowel	India	Survived (6) Death (2)
3.	Kajal P *et al*.[20th]	Case report	One male	Present	2022	Sigmoid colon Ileum	India	Survived
4.	Ramakrishnan S *et al*.[21st]	Case report	One male	Present	2020	tongue	India	Survived
5.	Muthukanagarajan SJ, al.[22nd]	Case report	One female	Present	2016	jejunum	India	Death
6.	Jun H *et al*.[23rd]	Case report	One male	Present	2017	Stomach colon	Korea	Survived
7.	Sathe PA *et al*.[24th]	Case report	One male	present	2015	Terminal ileum	India	Survived
8.	Mathur NB *et al*.[25th]	Case report	One male	Present	2013	stomach	India	Survived
9.	Patra S *et al*.[26th]	Original research	Male: female 1:1	Present	2003-2011	Small bowel and Large bowel	India	Survived (3) Death (3)
10.	Öztürk MA *et al*.[27th]	Case report	One male	Present	2011	Ileocecal valve ascending colon	Turkey	Death
11.	Gupta R *et al*. [12th]	Case report	One male	Present	2010	Jejunum Tongue	India	Survived
12.	Sarin YK *et al*.[28th]	Case report	One male	Present	2010	Terminal ileum	India	Survived
13.	Jain D *et al*. [29th]	Case report	One male	Present	2009	Terminal ileum	India	Survived
14.	Dhingra KK *et al*.[30th]	Case report	One male	Present	2008	Terminal ileum	India	Survived

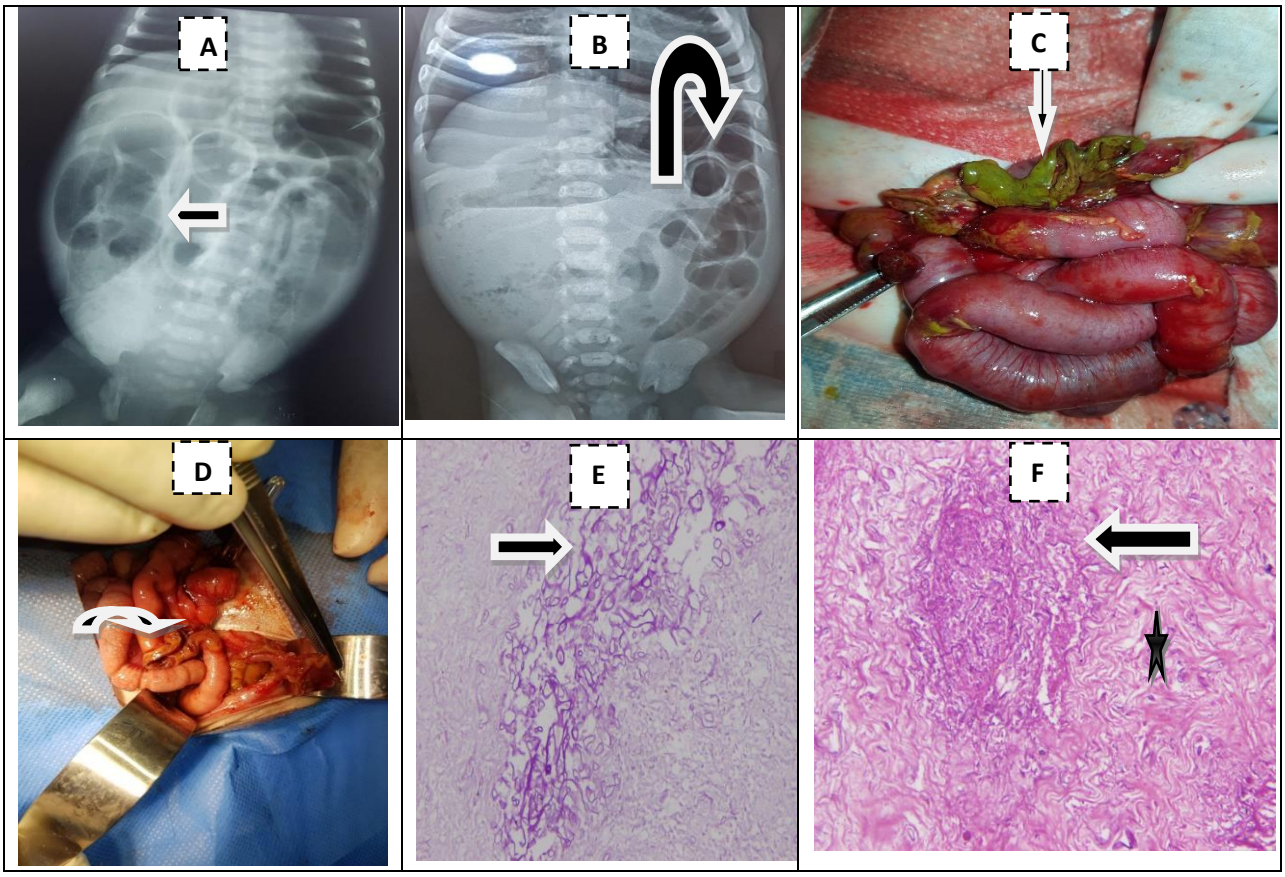

Figure1 A: X-ray abdomen AP view showed dilated stomach and multiple massive dilated gut loops **B:** X-ray abdomen AP view showed numerous air-fluid levels and the presence of gas within the small or large intestine wall **C-D:** Intraoperative examination showed necrotic green-colored gut segments with multiple perforations, and adjacent dilated gut loops. **E-F:** Histopathological examination showed numerous empty-looking broad aseptate hyphae of MM angioinvasion and destruction of the bowel wall. (H&E and PAS X 400) (All Arrows and Asterix highlight all the mentioned findings).

DISCUSSION

MM should be clinically suspected in all patients with localized bluish-black colored discoloration at any site, not responding to antibiotic treatment. MM involving GIT is rarely seen. GIMM affects various areas in GIT, including the tongue, palate, and stomach, small and large intestines. GIMM clinically mimics Necrotizing enterocolitis (NEC) in neonates, not responding to conventional antibiotic treatment, and cannot be distinguished radiologically. The presence of GIMM disguised as NEC is mind-boggling for clinicians.[19] A detailed review of neonatal GIMM from different regions of Asian countries is explained in Table-3. The primary demographic data regarding mean age, gender, prematurity, low birth weight, and a more common site of involvement were similar to other loco-regional reports.[6, 20-29]

In a meta-analysis by Jeong W *et al*., GIMM was seen in only 8% of cases with 54% mortality.[31] According to the literature, the colon, followed by the stomach, ileum, and appendix, are frequently reported sites. In the index study, the most common location was the terminal ileum and colon, similarly reported by other authors. [20, 22, 24-29] However, some authors reported rare lingual involvement in neonates as well.[21,23] An increasing trend of GIMM case reports is noted now. [6,25,31]

Grossly, the necrotic lesions of GIMM expand rapidly with ulcerating and necrotic edges forming a crater-like appearance as shown in per-operative images (Figure-I, C-D). Broad, aseptate, right-angle branching hyphae are found in hematoxylin and eosin (H&E) stained tissue sections on microscopic examination and can be highlighted further by PAS or Gomori methenamine-silver (GMS) stains. The most characteristic feature is blood vessel invasion, resulting in arterial thrombosis and subsequent ischemic necrosis. Dense ischemic infarction of surrounding tissues can also be noted. (Figure-I, F).

Emergency diagnosis, reporting, and initiation of antifungal therapy within six days are strongly associated with improved patient survival. The prognosis of GIMM is invariably grave; the reported mortality is 75% and 64%, as in our index study, all

three patients died despite surgical debridement and antifungal therapy. However, in contrast, Indian authors also reported the survived neonates.[23-25] Despite surgical management and antifungal treatment, the high mortality rate is attributed to ambiguous clinical features and zero clinical suspicion of GIMM in neonates. These children had rapidly progressive diseases and succumbed to death. Generally, GIMM should be suspected in any preterm infant with clinical features of NEC, with the absence of pneumatosis intestinalis on the abdominal radiograph and not responding to conservative management, including antibacterial therapy.

CONCLUSION

GIMM has a crooked presentation and clinically mimics NEC. GIM should be suspected in any neonate, in the absence of pneumatosis intestinalis on abdominal X-ray and no response to antibacterial therapy. The management includes a rapid institution of appropriate antifungal therapy, like Amphotericin B, and surgical resection of the involved intestine.

AUTHOR CONTRIBUTION

Madiha Ashraf: Conception of study
Fariha Sahrish: Drafting the work
Aysha Akram: Revising it critically
Alia Ahmad: Interpretation of data
Shehla Arshad: Conception of study
Saima Irum: Interpretation of data

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