الملخص العربى

إن زيادة استخدام المضادات الحيوية يتوافق مع زيادة مقابلة في حالات الاسهال الناتجة عن المضادات الحيوية والمشاكل المرتبطة به والمضاعفات ، وما يرتبط بها من تكاليف الرعاية الصحية، وجعل الوقاية أو العلاج من الاسهال الناتج عن المضادات الحيوية أولوية عالية.ان بكتيريا الكلوستريديم ديفيسيل هي السبب الرئيسي لحالات الاسهال المرتبطة بالمضادات الحيوية في المرضى في المستشفيات اللتى طرحت تحديات جديدة بسبب ظهور سلالات شديدة الضراوة من بكتيريا الكلوستريديم ديفيسيل التي قد تكون مقاومة للعلاج. ان التشخيص المختبري السريع الدقيق أمر بالغ الأهمية للحد من المراضة من بكتيريا الكلوستريديم ديفيسيل والسماح بتنفيذ تدابير محددة لمكافحة العدوى.

الغرض من هذه الدراسه هو قياس نسبه الأصابه ببكتيريا الكلوستريديم ديفيسيل فى حالات الأسهال الناتجه عن المضادات الحيويه فى الأطفال وايضا لمعرفة عوامل الخطورة الأخرى المسببه لبكتيريا الكلوستريديم ديفيسيل.

وقد أجريت هذه الدراسة على خمسين مريض من اقسام مستشفى كلية الطب جامعة عين شمس يعانون من الأسهال الناتج عن عدوى المستشفيات وكانت الدراسة على مدار ٨ أشهر (أبريل - نوفمبر٢٠١٤)، و يتكون هولاء المرضى من ثلاثين ذكر(٦٠ %) وعشرين أنثى (٤٠%) ، وكان متوسط السن 11.5 (4.8-21.3) months

كل الأطفال الذين تشملهم الدراسه خضعوا للآتى:

١- أخذ تاريخ مرضى مفصل مع الأهتمام بالأسهال(المدة ،عدد مرات التبرز الأعراض المصاحبه،تاريخ أخذ اى علاج) و ما يصاحبه من أعراض الجهاز الهضمى (ألم بالبطن، قئ، أنتفاخ، دم شرجى) وتاريخ تناول المضادات الحيويه (النوع، تايخ البدء،المدة) وايضا عوامل الخطوره (تناول مثبطات المناعة و أستخدام الأنبوبه المعدية وحدوث جراحة سابقه فى الجهاز الهضمى)

٢-الفحص اكلينيكى الشامل والذى يشمل قياس الوزن والطول، قياس الحراره، فحص علامات الجفاف، العلامات الحيوية (سرعة التنفس، سرعة نبض القلب، مقياس حرارة المريض) ،الفحص العام، وفحص البطن(من حيث وجود انتفاخ- الم-تضخم بالأعضاء الداخلية).

٣-الأختبارات المعملية المبدئية:

I- صورة دم كاملة بالأضافة الى العد التفريقى قبل وبعد حدوث الأسهال.
II- تحليل المتفاعل البروتينى ج قبل وبعد حدوث الأسهال.
III-تحليل براز.
IV-مزرعة براز، و قد تم الحصول على عينات البراز وزرعها مباشرة في المستنبتات البكتيرية ذات الكواشف الملونة كما تم زرعها فى آغار الدم.

وقد وجد أنه ليس هناك اختلاف ذو دلالة أحصائيه بالنسبه لسبب دخول المستشفى بين المجموعة الموجبة للكولسترتيم دفيسيل و المجموعة السالبة. كما وجد أن نسبة حدوث الأسهال الناتج عن عدوى المستشفيات 1045/100,000 ، وتمثل الكلوسترتيم ديفيسيل ١٨% من الأسهال الناتج عن عدوى المستشفيات،و نسبة حدوث الكلوسترتيم ديفيسيل 188/100,000 .

أما بالنسبة لعوامل السن والجنس ، فلا يوجد أختلاف ملحوظ بين المجموعتين.

أما بالنسبة للأسهال يوجد اختلاف ذو دلالة أحصائيه،حيث يزداد مدة الأسهال وعدد مرات التبرز فى المرضى المصابيين بالكلوسترتيم ديفيسيل.

أما ما يتعلق بالأعراض الأكلينيكية والعلامات المرضية المصاحبة للأسهال يوجد اختلاف ذو دلالة أحصائيه، ففى المرضى المصابيين بالكلوسترتيم ديفيسيل يرتفع معدل الأصابة بأعراض الحراره ،و ألم البطن.

أما بالنسبه للفحص الأكلينيكى يوجد اختلاف ذو دلالة أحصائيه ،حيث يرتفع معدل الأصابة بأنتفاخ البطن فى المرضى المصابيين بالكلوسترتيم ديفيسيل.

وبالنسبة لأختبارات الدم يوجد اختلاف ذو دلالة أحصائيه، فقد وجد أن مرضى الكلوسترتيم ديفيسيل يرتفع لديهم متوسط عدد خلايا الدم البيضاء والبروتين المتفاعل ج.

وبالنسبة لمزرعة البراز،المرضى الموصابين بالكلوسترتيم ديفيسيل يمثلون ١٨% من المرضى، أما المصابين بالبكتريا الأخرى يمثلون ١٠% من المرضى و المرضى الذين لم يصابوا بالبكتريا ٧٢% من المرضى. ولذالك فان سبب الأسهال هو أسهال فيروسى ٦٠% من المرضى ، أسهال بكتيرى ٢٨% من المرضى، أو أسهال طفيلى ١٢% من المرضى.

Abstract

Background: Once a nosocomial diarrhea, Clostridium difficile infection (CDI) now appears frequently in exposure to antibiotics, The incidence of Clostridium difficile-associated diarrhea (CDAD) is increasing worldwide, So this clinical study was carried to evaluate children who developed diarrhea during hospitalization in Children's hospital , Ain shams university.

Aim : This study measure the frequency of clostridium difficile as a cause of nosocomial diarrhea and also determine the risk factor associated with the development of Clostridium difficile-associated diarrhea in children.

Patients and Methods: Fifty patients suffering from diarrhea, admitted in Ain Shams Pediatric Hospital over a period of 8 months (April - November, 2013) were enrolled in the study. All enrolled patients were subjected to detailed history documentation, clinical examination and laboratory investigations in the form of complete blood count (CBC), C-reactive protein (CRP) at admission and after onset of diarrhea, Stool samples of patients with diarrhea that developed 3 or more days after hospital admission were submitted for stool analysis and culture, detection of clostridium difficile by using Toxigenic Clostridium difficile selective medium(cycloserine cefoxitin fructose agar (CCFA)

Results: The frequency of nosocomial diarrhea was 1045/100,000, Clostridium difficile represented 18% of cases with ND, the frequency of clostridium difficile associated diarrhea was 188/100,000 of hospitalized children, The cause of nosocomial diarrhea was found to be viral diarrhea 60% of patients, bacterial diarrhea 28% of patient and the parasitic cause 12% of patients, the antibiotic combinations was found to be the independent risk factor for the clostridium infection.

Conclusions: CDI is considered to be the main cause of bacterial infectious diarrhea in nosocomial settings and Receiving combinations of antibiotics represents the main risk to develop diarrhea associated to *C.difficile*.

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Aim of work

The aim of this study was to measure the frequency of clostridium difficile as a cause of nosocomial diarrhea and also to determine the risk factor associated with the development of Clostridium difficile-associated diarrhea in children.

Appendix

Appendix : Causes of admission in the studied sample

Measure	N	Diagnosis
Chest diseases	18	• 8 cases with bronchopneumonia • 5 cases with bronchial asthma • 3cases with acute bronchiolitis • 2cases with chronic stridor (laryngomalacia)
Renal diseases	8	• 4 cases with nephrotic syndrome • 3 cases with glomerulonephritis • A case of acute pyelonephritis
Blood diseases	8	• A case of autoimmune hemolytic anemia • A case of chronic hemolytic anemia • 3 cases with ITP • A case of henoch-schoenlein purpura • A case of drug-induced purpura • A case of hemophilia A
Cardiac diseases	7	• 4 Cases with congenital heart disease • A case of cardiomyopathy • 2 cases with congestive heart failure
Neurogenic diseases	5	• 3 Cases with cerebral palsy • A case of convulsion(epilepsy) • A case of Guillain-Barre syndrome
Nutritional disorder	2	• A case of rickets(Renal osteodystrophy) • A case of hypocalcemic teteny
Gastrointestinal diseases	1	• A case of ulcerative stomatitis with dehydration

Discussion

Diarrhea is defined as at least 1 day with ≥3 unformed stools or a significant increase in stool frequency above baseline **(Cohen et al., 2010)** , Nosocomial diarrhea is an acute episode of diarrhea in a hospitalized patient that was not present on admission and arises after ≥3 days of hospitalization **(Polage et al., 2012**)

Nosocomial diarrhea is a common complication in hospitalized patients but its causes and significance are underappreciated. Diarrhea predisposes patients to infections, contributes to morbidity and mortality, and increases hospital length of stay and costs . Physicians frequently focus on Clostridium difficile infection (CDI) as the primary cause of diarrhea (**Polage et al., 2012**)

Clostridium difficile is the main cause of nosocomial diarrhea. Diarrhea associated with *C. difficile* has increased incidence, morbidity, and mortality in the last few years. due to the emergence of hypervirulent strains, increased use and abuse of antibiotics, and the increase of susceptible at-risk populations(**Vecchioa and Zacurb et al., 2012)** .

This study was designed in an attempt to determine the frequency of clostridium difficile as a cause of nosocomial diarrhea and also to determine the risk factors associated with the development of Clostridium difficile-associated diarrhea in children. It included 50 patients admitted in Ain Shams Pediatric Hospital.

The result of the present study showed that there was no significant difference between patients with CDI and other causes of ND regards the cause of admission, this agree with those of (**Lee et al., 2012**) who found that there was no significant difference between between CDI and other causes of ND regarding the cause of admission.

Regarding the cause of nosocomial diarrhea, In the present study , viral diarrhea represented 60% of patients , bacterial diarrhea represented 28% of patients and the parasitic cause represented 12% of patients . These results were in agreement with

those of **(Gutierrez-Gimeno et al., 2009)** who found that Nosocomial viral diarrhea accounted for between 47% -69% of all hospital-acquired acute gastroenteritis among hospitalized children in the studies covering Austria, Germany, Spain and Switzerland and also agree with **(Fordtran et al., 2006)** who found that 25 from 100 patients would have a bacterial cause. On the other hand, the frequency of community acquired rotavirus GE represented 21% among agroup of infants and children with acute diarrhea **(El-Mohamady et al ., 2006)** , while the frequency was higher in another Egyption study **(El-Hodhod et al ., 2008)**

Regarding The frequency of nosocomial diarrhea and clostridium difficle associated diarrhea :

The result of the present study showed that the frequency of ND was 1045/100, 000 Two studies from 1987 and 1991 followed large numbers of patients from admission to discharge, nosocomial diarrhea in 32% and 22% of patients, respectively **(McFarland et al., 1995)** (**Samore et al., 1994)**. More recently, a point prevalence study of 485 hospitalized patients found that 12% of patients had diarrhea (12000/100, 000) including 27% of those hospitalized for ≥3 weeks (**Garey et al., 2006**), the most recent study, in Northeast Brazil 8.7% infants developed ND with an incidence density of 1130 per 100, 000 patient days (**Sette et al., 2012).**

Clostridium difficile is the most common bacterial infectious cause of nosocomial diarrhea , The results of the present study showed that CD was found among 18% of hospital acquired diarrhea, these results are in agreement with those of **(Cohen et al ., 2010) (Polage et al., 2012**) who found that CDI is the most common cause of ND representing 10%–20% of cases . **Stanley (2013)** found that C. difficile is now the most common cause of diarrhea in the acute care setting , responsible for up to 30%of all cases. The incidence of *C. difficile* infection has been increasing rapidly since the early 2000s. The rate of *C. difficile* infection nearly tripled between 1996 and 2005.The number of severe cases of *C. difficile* infection is also rising, The increasing of disease may be explained by a rise in an epidemic strain (NAP1/BI/027) which produces toxin A and B in significantly greater quantity compared to the normally occurring strain**(Dineen et al., 2013)**

The results of the present study showed that the frequency of clostridium difficile associated diarrhea was 188/100, 000 among hospitalized children , there was a wide variation in incidence of CDI among countries.

A prospective survey of oncology patients <18 y with gastroenteritis was performed in Egypt, Fifteen cases (14%) were due to C. difficile, confirmed by toxin testing. Infection with C. difficile had the highest mortality rate **(El-Mahallawy et al., 2004).**

A study done in Kuwait over a 3-year period found that the prevalence of hospital-acquired CDI amongst patients with diarrhea in 2003, 2004 and 2005were , 17/175 (9.7%), 17/218 (7.8%) and 22/304 (7.2%), respectively(**Jamal et al., 2010)**, while the incidence was much low in a study done in Dhahran, Saudi Arabia where the incidence of CDAD was 24 and 17 per 100, 000 patient in 2007 and 2008, respectively. The incidence in these hospital is low and this could be related to under detection due to the lack of the availability of cell cytotoxicity assay or toxigenic culture **(Al-Tawfiq , Abed., 2010)**

In a recent study from Jordan , CDI was found to be 13.7% of 300 stool Specimens (**Nasereddin et al., 2009).** in a Spanish study, the mean annual incidence rate was 41.2 per 100, 000 discharges **(Soler et al., 2008).** in a Brazilian study of both inpatients and outpatients, C. difficile was cultured from 14 (6.7%) of 210 children aged 3 months to 7 years (**Pinto et al., 2003)**

This wide variation in incidence can be explained by differences in laboratory testing (toxin or culture) and patient factors e.g. age of the child , prior antibiotics, method of feeding (enteral , breast or formula) or parenteral) and duration of hospital stay **(Enoch et al., 2011)**

Some studies have suggested that CDI is emerging as an increasingly common cause of diarrhea in children, The largest study of CDI in children was in hospitalized patients and reported that the incidence of CDI increased substantially from 2001 to 2006, from 44 to 65 cases per 100, 000 patient-days**(Kim et al 2008)** and In a more recent study, Zilberberg and colleagues reported a increase in CDI-related hospitalisations in the US for the period 1997-2006, the almost 2-fold rise in incidence (from 72 to 128/100, 000 hospitalisations) **(Zilberberg et al., 2010)**

Contrary, some studies describe a decrease in incidence of CDI as Benson and colleagues who noted a significant decrease in the incidence of C. difficile infection among inpatients over a 5-year period (from 100 cases per 100, 000-patient days to 68 cases per 1000-patient days) (**Benson et al., 2007)** and in a study done in Finland found that The annual incidence rate of CDIs decreased by 24%, from 119 per 100, 000 population in 2008 to 90 per 100, 000 in 2010 **(Kanerva et al., 2013) ,** and this can be explained by increase the awareness of CDI and improvement in control measures.

Regarding the age, The median age in the present study is 11.5 (4.8 – 21.3) months where there was no significant difference between patients with CDI 9 (4.5 – 14) months and other causes of ND 12 (4.5 – 26.5) months. these results are in agreement with those of **(Zilberberg et al., 2008)** who found that CDI-related hospitalizations nearly doubled among infants younger than 12 months from 2000 through 2005, and this explained by their inability to mount a specific serum IgG immune response when first exposed to the toxins, which is also associated with a higher rate of recurrent disease in this subset of patients . also agree with A smaller prospective study of 226 children in a US paediatric outpatient setting detected C. difficile in 8 of 104 (7.7%) samples tested 54 with children under one year of age, more likely to have C. difficile detected than older children (16% versus 5%, P Z 0.09) **(Denno et al., 2005).**
However these results were disagree with those of **(Enoch et al., 2011**) who stated that The median age of children with CDI was 4 years, with 26% being <1 year and 5% were newborns.

Regarding gender , the present study showed that males accounted for 66.7% in CDI patients with no significant difference with other causes of ND 58.5%. these results Agree with those **(Svenungsson et al., 2003)**who found that patients did not differ from the total patient population with regard to gender (135 males and 169 females, *P* _ 0.9**)** but disagree with those of **(Vecchioa and Zacurb., 2012**) who stated that Male sex was more comman in CDI cases.

The presenting complaint:

The hallmark of CDI is new-onset diarrhoea **(Joshi et al., 2012)** .
Regarding the diarrhea, the result of the present study showed that the diarrhea of CDI was significantly more frequent 6 (4 – 6.5) motion/day , more delayed in onset 10 (7.5 – 14.5) days, and more prolonged in duration 12 (8 – 15) days than other causes of ND. These results are in agreement with those of **(Dubberke et al., 2011)** who found that C.difficle Patients had significantly more bowel movements per day compared to their baseline (median number of bowel movements was 3 versus 1 at baseline [*P* _ 0.001] . In a study among 45 Indian children the results found that moderate diarrhea (passage of 7-10 stools/day; 60%) and bloody diarrhea (24%) were relatively frequent CDI (**Gogate *et al*., 2005).**

Svenungsson et al (2003) found that The median number of days between admission and the date of microbiological diagnosis of *C. difficile* infection was 10 (range, 2 to 73 days) for hospital-associated cases also **Stanley (2013)** found that the median time to symptoms was 9 days after antibiotics and 13 days after admission to hospital . **(Forster et al., 2012)** found that The median number of days from admission to detection of *C. difficile* was 12 days(IQR 7–2400),

Polage et al (2012) stated that Most nosocomial diarrhea not due to CDI is mild or moderate and resolves after a few days while Diarrhea in patients with CDI usually occurs within several days to months after treatment with antibiotics.

The results of the present study disagree with a study done in Kuwait and found that There was no statistical significant difference (P > 0.05) between the duration of stay in the hospital before the onset of the disease and the development of CDI**. (Jamal et al., 2010)**

In the present study there was no significant difference regards consistency, presence of blood and mucus between patients with CDI and other causes of ND, clostridium difficile associated diarrhea was watery in 66.7% of patients, bloody in11.1% of patients, with mucus in55.6% of patients and these result agree with those of (***Goldstein et al., 2009)*** who found that Liquid and formed specimens had similar rates and also agree with those of **(McFarland ., 2008) (*Sammons et al* .,**

2013) who stated that diarrhea was rarely with blood or mucous and come in agreement with a study done Among 200 Canadian children with CDI and found that 79% presented with watery diarrhea and 12.5% with bloody diarrhea (**Morinville, McDonald., 2005)** But disagree with **(Badger et al., 2012)** who stated that *C difficile* associated diarrhea was usually watery**.**

Regarding the fever: The result in the present study showed that fever was present among 77.8% in patients with CDI which is significantly higher than other causes of ND (36.6%) and these results agree with those **(Jamal et al., 2010)** who found that a statistically significant difference was observed that patients who had fever of >38 C in CDI was 51.7% also **Gogate *et al* (2005**) found that Fever (84%) was relatively frequent CDI manifestations.

Regarding the associated GI symptoms:

Abdominal pain :

The result in the present study showed that abdominal pain was present among 100% in patients with CDI which is significantly higher than other causes of ND (36.6%) and these results are in agreement with those of (**Gogate *et al*., 2005**) who found that abdominal pain (60%) were relatively frequent CDI manifestations and also **Cohen et al (2010)** described cramping abdominal pain in the *Clostridium difficile* diarrhea . **Badger et al (2012)** stated that CDI usually manifests itself as mild to moderate diarrhea, often associated with crampy abdominal pain **.**

vomiting :

The result in the present study showed that vomiting was present among 55.6% in patients with CDI with no significant difference with other causes of ND (36.5%) and these results agree with **(Stanley ., 2013)** who stated that In addition to diarrhea, patients may present with other symptoms and signs such as nausea, vomiting.

Bleeding per rectum :

The results of the present study found that There was no significant difference between patients with CDI (11.1%) and other causes of ND (7.3%) regarding bleeding per rectum. these results agree with those of **(Knight, Surawicz ., 2013**) who stated that diarrhea of CDI is rarely bloody although it can be in severe cases, also agree with **(Dellinger et al., 2008)** who stated that CDAD not usually bloody. **Morinville, McDonald (2005)** reported 12.5% with bloody diarrhea Among 200 Canadian children with CDI. **Gogate *et al* (2005)** found that bloody diarrhea (24%) were relatively frequent CDI manifestations among 45 Indian children also **Fordtran (2006)** stated that Stools are usually not grossly bloody, but they can be Occult blood is present in 30% of patients.
On the other hand, Acute self-limiting infectious diarrhea proved to be the most common cause of BPR (37.1%), Bacterial causes represented 72.2% **(El-Khayat et al., 2006).**

Local abdominal examination :
The results of the present study showed that abdominal distention and tenderness were significantly more frequent in patients with CDI 88.9% , 77.8% than other causes of ND 24.4% , 19.5% respectively and these results are in agreement with those of **(Bouza et al., 2005) and (Bauer et al., 2009)** who stated that Other symptoms of mild to moderate disease can include abdominal distension. while **Knight, Surawicz (2013)** stated that abdominal distension is significant indicators of severe disease.

As regard the risk factors:

The use of antibiotics is considered The main risk factor for the development of CDI **(Rebeaud et al., 2012)** and this may be explained by that the bacterial gut residents suppress growth of *C. difficile* in the colon. Broad-spectrum antimicrobials have the potential to disrupt the balanced ecology of the stool microbiota creating an opportunity for *C. difficile* spores to germinate resulting in overgrowth and attendant production of toxins, which are responsible for most of the clinical symptoms of CDI and pseudomembraneous colitis**.(Hell et al ., 2013***)***and these increases the risk of symptomatic CDI by 2- to 16-fold **(Shah et al., 2012).**

Johal et al (2004) found that *C. difficile* diarrhea which followed antibiotic exposure in 96% started in the hospital in 72% of patients and in the community in 28% patients . In addition, 87% of those in whom the disease began in the community had been hospitalized during the preceding 12 months.

The results of the present study showed that the duration of antibiotic administration was significantly longer in patients with CDI 10 (7.5 – 14.5) than other causes of ND 6 (4.5 – 7) and these results agree with an Egyptian study found that There was a strong link with intravenous antibiotic use for more than a week prior to the occurrence of diarrhea (66% versus 10% in those without C. difficile infection) **(El-Mahallawy et al., 2004)**
In a case-control study done in the Netherlands found that a 7- to 10-fold increased risk for CDI during antibiotic therapy that persisted for the first month after stopping antibiotics. The risk gradually declined over the following 2 months, returning to baseline 3 months after stopping antibiotic therapy. There was a trend toward increased risk with longer overall duration of antibiotics **(Hensgens et al., 2012)** . **Kim et al (2010)** stated that the longer the duration of antibiotics use, the higher the risk of colonization by CD, also **Vecchioa and Zacurb (2012)** stated that Prolonged use of antibiotic was the main risk factor to develop CDI.

Regarding the type of antibiotic , Amoxicillin and third-generation cephalosporins were significantly more frequent in patients with CDI than other causes of ND and these results were in agreement with many studies as In one Turkish study, all patients with CDI were receiving antibiotics, mostly third generation cephalosporins or ampicillin-sulbactam with an aminoglycoside **(Oguz et al., 2001)** . The majority (74%) of patients in the Canadian case series had received antibiotics in the preceding 2 months, with cephalosporins being implicated most frequently; 59% received multiple antibiotics. **(Morinville, McDonald., 2005)** . **Jamal et al (2010)** found that the third-generation (ceftriaxone, cefotaxime or ceftazidime) and fourth-generation (cefepime) cephalosporins were the most common antimicrobial agent received by the patients with CDI. They were used alone (73%) or in combination with amikacin (5.4%). Also **Al-Tawfiq, Abed (2010)** found that Cephalosporins were the most common antimicrobial drugs (30.6%), fluroquinolone (15.3%) and a combination of these