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Development of an Ecological Model to Predict Risk for Acquisition of *Clostridium difficile*-Associated Diarrhea During Acute Care Hospitalization

by

Susan Elaine Steele

A dissertation submitted in partial fulfillment
of the requirements for the degree of
Doctor of Philosophy
College of Nursing
University of South Florida

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Dedicated to the memory of three individuals who fueled my desire for scholarship:

Edward Leroy Steele

1918-2007

Imogene M. King, Ed.D., RN

1923-2007

Rena Mae Lawrence, Ph.D., RN

1933-2005

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ABSTRACT

Background: The traditional model of infection control has failed to stop the spread of emerging infectious diseases such as *Clostridium difficile*-associated diarrhea (CDAD) in the acute care environment. Ecological models, which rely upon identification of susceptible hosts, offer an alternative to the prevention of deadly outbreaks. Previous epidemiological research has identified a number of risk factors associated with CDAD. Utilization of this body of research by nurses is limited due to methodological issues that introduce bias and confounding, and use of variables that have limited meaning to the practicing clinical nurse.

Aim: The aim of this study was to develop an ecological model useful for nurses in predicting the susceptibility of individuals to CDAD during an acute care hospital stay.

Method: A case-control study compared 66 cases with CDAD to 66 controls matched for the temporal and spatial risk factors of hospital admission date and geographic nursing care unit within the institution. The two subject groups were compared on variables of age, antibiotic burden, laxative or bowel preparation exposure, nutritional status, gastric acid suppression therapy, enteral feeding exposure, and severity of illness as measured on the Horn Severity of Illness index. All subjects were hospitalized between January 1, 2000 and December 31, 2006.

Results: On univariate analysis, age, severity of illness, serum albumin levels, length of exposure, and proton pump inhibitor drug burden were significantly associated with CDAD status. Following multivariate analysis, only severity of illness, length of exposure, and decreased antibiotic drug burden were significantly associated with the development of hospital-acquired CDAD.

Conclusions: This study supports the use of an ecological perspective in identifying risk factors and interventions to prevent the future spread of this infectious disease.

Chapter One

Introduction

Clostridium difficile (*C. difficile*) is a gram-positive, spore-forming anaerobe, first identified in 1935 in the normal enteric flora of infants (Hall & O'Toole, 1935). A case-series analysis published in the early 1960's discounted a serious health threat from *C. difficile*, concluding that either the toxin was not produced within the human body or that humans did not exhibit a marked sensitivity to the toxins produced by the bacteria (Smith & King, 1962). However, discovery of a link between antibiotic-associated pseudomembranous colitis and a toxin-producing strain of a *Clostridium* species in the 1970's clearly established the organism as a human pathogen and triggered a series of investigations associating various antibiotics with the development of the disease (Bartlett, 2004).

By 1980, *C. difficile*- associated diarrhea (CDAD) was identified as a serious nosocomial infection (Mulligan, George, Rolfe, & Finegold, 1980; Peikin, Galdibini, & Bartlett, 1980). Since the 1980's, research has expanded understanding that *C. difficile* infection occurs in a continuum extending from asymptomatic carriage to fulminating pseudomembranous colitis. Toxigenic strains of *C. difficile* have been isolated from feces of healthy adults not recently exposed to antibiotics, with reported prevalence ranging from 4.2% to 53.3% (Iizuka et al., 2004; Kato et al., 2001; Nakamura et al., 1981), indicating that transmission and carriage of the organism may occur undetected

among the general population, as well as among institutionalized individuals. Although such colonization is believed to be transient in most cases, there is evidence that persistent *C. difficile* colonization does occur in some individuals (Ozaki et al., 2003). Among hospital patients, colonization followed by an IgG antibody response has been associated with a decreased risk of CDAD acquisition after adjustment for age, sex and disease severity (Kyne, Warny, Qamar & Kelly, 2000), and an acquired immune response to toxin A following an episode of CDAD is believed to be protective against recurrence of diarrhea (Kyne, Warny, Qamar & Kelly, 2001).

There is indication that increasing numbers of persons are experiencing the most severe outcomes of *C. difficile* infection including pseudomembranous colitis and death (Dallal et al., 2002; Frost, Craun, & Calderon, 1998; Morris et al., 2002; Pepin et al., 2004). Most recently, a change in the epidemiologic pattern of the disease has been noted, including geographically dispersed outbreaks (Krausz et al., 2005; Loo et al., 2005; McDonald et al., 2005; Warny et al., 2005), an increasingly virulent strain of the organism (Morris et al., 2002; Pepin et al., 2004; Pepin, Valiquette, & Cossette, 2005), and susceptibility of populations previously believed to be low risk for acquisition of CDAD (Centers et al., 2005). Because persons who develop nosocomial CDAD experience a more than 50% increase in hospital costs, increased length of stay and a significantly higher mortality rate, the burden of this disease threatens the health care system (Kyne, Hamel, Polavaram, & Kelly, 2002; Miller, Hyland, Ofner-Agostini, Gourdeau, & Ishak, 2002).

Investigations in prevention of CDAD have included antibiotic restriction policies, vaccine development (Kotloff et al., 2001; Sougioultzis et al., 2005), and dietary

modulation of intestinal microflora. Dietary interventions have included use of *prebiotics*, non-digestible fiber agents that ferment and foster growth of normal flora, *probiotics*, live organisms found in healthy flora, and *synbiotics*, a combination of a prebiotic and probiotic agent (Bengmark, 2003; Collins & Gibson, 1999). These research efforts, while somewhat promising, currently lack sufficient evidence to support their use, and prevention of CDAD is dependent upon traditional infection control processes such as hand hygiene, isolation, and environmental decontamination.

Epidemiological and biomedical models for the prevention of infectious diseases are based upon identification, and destruction or elimination of the causative organism. Such an approach ignores the evolutionary potential of an enormously diverse prokaryotic domain (Galvani, 2003; Purssell, 2005a, , 2005b) as well as the environmental and social factors in the modern age that have altered the relationship between man and microbe (de Albuquerque Possas, 2001; McMichael, 2004; Waldvogel, 2004). In contrast, an ecological model of infectious disease offers an understanding of the way temporal and spatial relationships between host and pathogen can be altered to reduce the risk of disease (Koren & Crawford-Brown, 2004).

The human intestine can be described as a complex ecosystem, comprised of a constantly resurfacing organ playing host to both resident and transient microbes existing in a state of mutualism (Xu & Gordon, 2003). In healthy adults, the indigenous bacteria within the intestine control pathogenic invasion via colonization resistance (van der Waaij, 1989). In return, the host provides mucus, shed epithelial cells, and ingested food particles which allow the bacteria to thrive in a physiologic bioreactor (Backhed, Ley, Sonnenburg, Peterson, & Gordon, 2005).

CDAD represents a disruption of the intestinal ecosystem, manifested by an increase in enterococci (Hopkins & MacFarlane, 2002; Ozaki et al., 2003) a decline in *Bifidobacteria* and *Bacteroides* colonization (Hopkins & MacFarlane, 2002), and proliferation of *C. difficile*. A number of case series reported successful treatment of recurrent CDAD by rectal or nasogastric administration of donor stool, further supporting the importance of a healthy intestinal ecosystem in combating the disease (Aas, Gessert, & Bakken, 2003; Bowden, Mansberger, & Lykins, 1981; Eiseman, Silen, Bascom, & Kauvar, 1958; Persky & Brandt, 2000; Schwan, Sjolín, Trottestam, & Aronsson, 1984; Tvede & Rask-Madsen, 1989). Restoration of the normal colonic flora has been the focus of increasing research regarding a number of diseases including *C. difficile*, driven by advances in genomics and global awareness of emerging infectious diseases (McMichael, 2004; Rastall et al., 2005). However, there remains a lack of clinical trial data to help target populations most likely to benefit and to substantiate the specific agents appropriate for prevention and treatment of CDAD.

Alteration in intestinal ecology can effect diarrhea through disturbances in motility, defects in the immune system, and failure of colonization resistance (Hawrelak & Myers, 2004). The development and maintenance of the normal intestinal ecosystem is influenced by several factors, including age, dietary intake, and systemic drug therapy.

Indigenous bacteria begin to form colonies at the time of birth as a result of exposure to maternal bacteria in the vagina, and within the first few days of life, the diversity of the fecal flora changes rapidly (Park et al., 2005). Early in life, *C. difficile* colonization is common, and *C. difficile* has been demonstrated within the intestinal microbiota of healthy infants in hospital, home, and day care environments (Larson,

Barclay, Honour, & Hill, 1982; Matsuki et al., 2005; Park et al., 2005; Penders et al., 2005; Stark, Lee, & Parsonage, 1982). As an individual matures, changes in bacterial composition result in greater species diversity accompanied by a decline in *C. difficile* (Hopkins & MacFarlane, 2002; Hopkins, Sharp, & Macfarlane, 2001). However, as an individual approaches advanced age, a decline in *Bifidobacteria* and an increase in enterococci occurs (Hebuterne, 2003; Hopkins & MacFarlane, 2002; Hopkins, Sharp, & Macfarlane, 2001).

Dietary intake plays a significant role in fecal bacterial colonization. Ingestion of non-digestible carbohydrates provides nutrition to support the production of various bacterial species. Both naturally occurring and experimental-supplement induced dietary intake of oligosaccharides results in an increase in fecal *Bifidobacteria* levels (Boehm et al., 2004; Gibson, Beatty, Wang, & Cummings, 1995; Langlands, Hopkins, Coleman, & Cummings, 2004; Penders et al., 2005; Stark, Lee, & Parsonage, 1982) and the increase is dose-dependent (Bouhnik et al., 1999). Oligosaccharide supplementation resulted in a decrease in *C. difficile* colonization both in vitro (Hopkins & MacFarlane, 2003) and in healthy adults receiving the supplement during a course of an oral cephalosporin antibiotic (Orrhage, Svante, & Nord, 2000).

Hospitalization for illness or injury often requires nutritional support via enteral nutrition (tube feeding). A common gastrointestinal complication of enteral feeding is diarrhea, with reported incidence ranging from 2.3% to 38% among general hospital populations and as high as 68% among critically ill adults (Cataldi-Betcher, Seltzer, Slocum, & Jones, 1983; Elpern, Stutz, Peterson, Gurka, & Skipper, 2004; Heimbürger, Sockwell, & Geels, 1994; Homann, Kemen, Fuessenich, Senkal, & Zumtobel, 1994;

Montejo, 1999; Pancorbo-Hidalgo, Garcia-Fernandez, & Ramirez-Perez, 2001; C. Smith et al., 1990). Suggested causes for tube-feeding-associated diarrhea include hypoalbuminemia, formula intolerance, concomitant use of sorbitol-containing elixir medications (Eisenberg, 2002) and colonic response to a non-physiologic form of feeding (Bowling & Silk, 1998). However, enteral nutrition also results in a decrease in anaerobic and an increase in aerobic bacteria in feces (Schneider et al., 2000; Whelan, Judd, Preedy, & Taylor, 2004), a trend that may be altered by formulas supplemented with fiber (Nakao et al., 2002). Enteral feeding and the presence of nasogastric or percutaneously inserted enteral feeding tubes have been found to have significant association with the acquisition of CDAD (D. Z. Bliss et al., 1998; Komatsu et al., 2003; Kyne et al., 1999; Lai, Melvin, Menard, Kotilainen, & Baker, 1997).

Illness and medical treatment also is associated with alteration in the gut ecosystem. Although antibiotics have received the most attention in investigations, both acid suppressive and laxative medications also have been implicated in altering colonic flora. The effect of antibiotics upon intestinal microflora varies both between and within drug categories. A structured research review of papers published between 1991 and 2000 identified a decrease in obligate anaerobes such as *Bacteroides*, *Bifidobacterium*, and *Lactobacillus*, an increase in facultative anaerobes such as *Enterococcus*, and *Streptococcus*, and an increase in aerobic organisms such as *Pseudomonas* (Sullivan, Edlund, & Nord, 2001) as common antibiotic effects. This same review identified a large number of cephalosporin agents associated with overgrowth of *C. difficile*. More recent publications report similar findings of floral suppression and proliferation of competing bacteria (Bartosch, Fite, Macfarlane, & McMurdo, 2004; Buhling, Radun, Muller, &

Malfertheiner, 2001; Hawrelak & Myers, 2004; Madden et al., 2005; Monreal, Pereira, & Lopes, 2005; Takesue et al., 2002). The ecosystem changes which occur as a result of antibiotic usage are not immediately reversible with cessation of antibiotic therapy, and it may take more than a month to return to pretreatment levels of dominant microbial species (Buhling, Radun, Muller, & Malfertheiner, 2001; De La Cochetiere et al., 2005).

The use of gastric acid suppressive medications has been associated with the development of CDAD in residents of long-term care facilities, as well as hospitalized and community-based individuals (Al-Tureihi, Hassoun, Wolf-Klein, & Isenberg, 2005; Cunningham, Dale, Undy, & Gaunt, 2003; Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004; Dial, Delaney, Barkun, & Suissa, 2005; L.V. McFarland, Surawicz, & Stamm, 1990). The exact effect of acid suppressive agents upon intestinal microflora is not known. It is hypothesized that suppression of gastric acid allows an increased number of potential pathogens to survive transition from stomach to intestine (Donskey, 2004).

Laxative drugs also may alter intestinal flora. Chronic constipation in adults is associated with alterations in colonic ecology as compared to healthy controls. Treatment with both bisacodyl and lactulose result in normalization of the flora (Bouhnik et al., 2004; Khalif, Quigley, Konovitch, & Maximova, 2005; Zoppi et al., 1998), but polyethylene glycol-4000 does not exert the same ecological effect (Bouhnik et al., 2004). Lactulose in particular has been found to promote the growth of lactic acid bacteria and *Bifidobacteria* (Salminen & Salminen, 1997), and to suppress the proliferation of potential pathogens such as *Clostridium difficile* (Ballongue, Schumann, & Quignon, 1997; Ito et al., 1997). Laxative usage is a potential confounder in research

regarding CDAD, since both the disease and the drug may alter stool consistency and frequency.

Problem Statement

Through previous research, a number of ecological risk factors pertinent to hospitalized adults have been identified for the development of CDAD. These risk factors include advanced age, use of tube feeding, malnutrition, severity of illness, and the use of medications that alter the intestinal flora, including gastric acid suppressant agents, antibiotics, and cathartics.

The purpose of this study was to develop and test an ecological model useful to nurses in predicting risk for the development of *Clostridium difficile*-associated diarrhea. It was anticipated that the model would serve three purposes: (1) serve as a foundation for development of a valid and reliable risk assessment tool (2) aid in the design of future clinical trials of nurse-directed prevention strategies and (3) assist clinicians in modifying infection control practices in institutional settings when caring for high-risk individuals.

Hypotheses

This study is designed to test a hypothesis about the risk factors for development of hospital-acquired CDAD:

H1: Severity of illness, length of exposure, and malnutrition, are significant predictors for the development of CDAD among cases as compared to *C. difficile*-negative controls with diarrhea when matched for admission date, and geographic unit of hospital, and after controlling for the effects of age, tube feeding, cathartic administration, antibiotic therapy, and acid suppression therapy.

Definition of Terms

Diarrhea.

For purposes of this study, diarrhea was defined as at least three stools within a 24 hour period documented within the medical record as “liquid”, “loose”, “unformed”, or “diarrhea”, or 250 milliliters of liquid stool collected via a colostomy, adhesive fecal incontinence collection pouch, or a rectally inserted bowel management tube, or at least 1500 milliliters of liquid stool from an ileostomy pouch.

Clostridium difficile-associated diarrhea.

Diarrhea occurring concurrently with or within 7 days of a positive cytotoxin assay, stool culture, or endoscopic examination consistent with pseudomembranous colitis.

Antibiotic burden.

Antibiotic burden was defined as the sum of the average daily maintenance doses of oral or intravenously administered antibiotics received by the patient during the risk period. These average daily doses were determined by use of the World Health Organization Defined Daily Dose system (WHO Collaborating Centre for Drug Statistics Methodology, 2007)

Length of exposure/risk period.

The length of exposure, or risk period, for this study commenced on the date of admission to the acute care hospital and ended on the date that a stool specimen was submitted for *Clostridium difficile* testing.

Significance to Nursing

Nurses spend the most time in the physical proximity of infected and potentially vulnerable patients within the health care system. Because nursing role functions include assistance with toileting, they are often able to predict a positive *Clostridium difficile* cytotoxin test result based upon knowledge of a patient's prior antibiotic use and the presence of a distinctive fecal odor (Johansen, Vasishta, Edison, & Hosein, 2002).

Nurses have been implicated as possible agents in the spread of the disease in hospitals (Chang & Nelson, 2000; Perry, Marshall, & Jones, 2001), and are responsible in institutional settings for supervising the environmental decontamination of individual patient rooms and geographic units, critical to the prevention of disease transmission (Chang & Nelson, 2000; Kroker, Bower, & Azadian, 2001; Mayfield, Leet, Miller, & Mundy, 2000; Verity, Wilcox, Fawley, & Parnell, 2001; Wilcox et al., 2003).

Diarrhea is a frequent cause of fecal incontinence in acutely-ill hospitalized adults (Bliss, Johnson, Savik, Clabots, & Gerding, 2000), requiring increased nursing care hours and supplies for skin cleansing, treatment of skin breakdown, and linen changes. CDAD is now recognized as the most common type of infectious nosocomial diarrhea (McFarland, 1995). Outbreaks of CDAD have been well-documented (Blot et al., 2003; Johnson et al., 1999; Kuijper et al., 2001) and reported incidence rates of nosocomial CDAD range from 0.19 to 6.8 per 100 hospital admissions (Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004; Thomas, Stevenson, Williamson, & Riley, 2002). A global shortage of qualified professional nurses compromises containment of emerging infectious diseases in health care settings (Stone, Clarke, Cimiotti, & Correa-de-Araujo, 2004).

Traditional methods of preventing the spread of infectious diseases within hospitals have not proven sufficient to combat emerging infections such as CDAD. An alternative ecological approach to disease prevention requires change in nursing practice to decrease the impact of the organism upon human hosts. By strengthening the natural flora of the hospitalized individual and increasing the spatial and temporal distance between susceptible persons and potentially infectious organisms, the degree of harm caused by the organism can be reduced (Pursell, 2005a). Table 1 compares and contrasts the biomedical and ecological approaches to infectious disease prevention.

Table 1
Prevention and Treatment of Infectious Diseases: Traditional Versus Ecological Models

Aspect of Care	Model	
	Traditional	Ecological
Detection	Identification of persons with disease	Identification of persons susceptible to disease
Symptom treatment	Treat symptoms	Treat only symptoms that have no benefit for the host
Chemoprophylaxis	Prophylactic administration of antimicrobial agents	Administration of biotherapeutic agents to maintain or restore host flora
Isolation	Isolate persons with disease	Isolate persons with symptoms and susceptible persons based on level of susceptibility and degree of immunosuppression
Environmental Controls	Uniform environmental cleaning protocols to remove pathogens	In addition to environmental cleaning protocols, increase spatial and temporal intervals between susceptible host and pathogen contact

Note. Adapted from Pursell, 2005a and Pursell, 2005b.

Key differences in the two approaches lie in the detection of susceptibility to disease and the modulation of host flora to combat the growth of potentially pathogenic microbial species. To utilize such an approach, nurses require a simple and reliable risk assessment tool for estimating susceptibility to CDAD. The development of a predictive model would enable further development of such a tool.

Summary

CDAD is a serious infection that can be understood as a disturbance of the intestinal ecology. Identification of the factors that make an individual most susceptible to CDAD would enable the design of infection control interventions based on ecological principles to decrease the extent of harm caused by this microorganism. Advancing age, severity of illness, diet, and use of antibiotic, acid suppressive and cathartic medications have been identified as factors that disrupt the normal intestinal ecology and may promote the development of CDAD.

Chapter Two

Review of the Literature

By virtue of its increasing incidence, virulence and global geographic range, CDAD is considered an emerging infectious disease. Changes within the environment and in human ecology are precipitating factors which account for the emergence of most such diseases (de Albuquerque Possas, 2001; Lederberg, Shope, & Oaks, 1992; McMichael, 2004; Morse, 1995). Changes in human demographics and behavior increase both exposure to infectious agents and susceptibility to their deleterious effects. This chapter will present a review of the analytic epidemiology research literature identifying human demographic and behavioral risk factors for the development of *C. difficile*-associated diarrhea. An ecological model to guide the study of CDAD will be developed.

A literature search was conducted in both Medline and CINAHL electronic databases for the years 1995 through March 2006. The keywords “*Clostridium difficile*” was paired using the Boolean connector “AND” with each of the following keywords: “risk factors”, “age”, “antibiotic”, “tube feeding”, “laxative OR bowel preparation”, “severity of illness” and “acid suppression”. Articles were limited to those published in the English language, and studies concerning adult subjects in a hospital or institutional setting, designed to understand risk factors for an initial episode of *Clostridium difficile*-associated diarrhea. The electronic search was supplemented with ancestral retrievals for a final yield of 73 publications.

The sample included one meta-analysis and one structured literature review. The remaining 71 original research reports included 36 case-control studies, 13 prospective and nine retrospective cohort studies, three clinical trials, and ten descriptive studies. The majority of the studies in the review were conducted in North America and Europe; however, the sample did include studies conducted in Asia and Oceania-Australasia, indicating potential for some genetic diversity among the study populations. Subjects were being treated both medically and surgically for a variety of conditions including cancer, pneumonia, complications of human immunodeficiency virus infection, and cardiac disease.

Qualitative Assessment

To systematically assess the quality of the clinical trial, case-control and cohort study publications, a criterion-based checklist was adapted (Downs & Black, 1998). The original checklist was modified based upon important domains and elements for systematic review advocated by the Agency for Healthcare Research and Quality (West et al., 2002).

The checklist items were entered into an Excel spreadsheet and each publication was subjected to the same questions regarding quality of the published report, internal validity and external validity of the research methods described, and disclosure of funding source. Publications were assigned a code of one if the element was present within the publication and a code of zero if the element was absent. Thus, the both the number and the percentage of publications that met each of the specified criteria could be calculated. The ten descriptive studies were not subjected to this checklist assessment, as the conduct of the studies and the method of reporting were incompatible with this tool.

These publications, along with the two systematic reviews, were used to help further shape understanding of the ecological concepts. Table 2 summarizes the results of this qualitative review of the publications.

Table 2
Number and Percent of Studies Meeting Evaluation Criteria

Checklist Item	Number	%
Reporting Quality		
Was a hypothesis, aim, or study objective stated?	52	85.25
Were the main findings of the study clearly described?	55	90.16
External Validity		
Were inclusion and exclusion criteria used to determine eligibility for participation in the study?	20	32.79
Were only inclusion criteria, and not exclusion criteria, identified in the paper?	20	32.79
Were the subjects representative of the entire population from which they were recruited?	40	65.57
Internal Validity (bias and confounding)		
Was an attempt made to blind subjects to the intervention they have received (clinical trials, n=3)?	1	33.33
Was an attempt made to blind those measuring the main outcomes of the intervention (clinical trials, n=3)?	1	33.33
Were the statistical tests used to assess the main outcomes appropriate to the research question?	55	90.16
Were the measurements used to assess the main outcome accurate (valid and reliable)?	52	85.25
Were subjects in different intervention groups (trials and cohort studies) or were cases and controls (case-control studies) recruited from the same population?	32	52.46
Were study subjects randomized to intervention groups (clinical trials)?	1	33.33
Was there adequate adjustment for confounding in the statistical analysis from which the main findings were drawn?	35	57.38
Disclosure		
Was the funding source for the study identified?	13	21.31

Report quality

Within this body of literature, reporting quality was high for two of the criteria considered. The majority of the published reports included an explicit or implied

hypothesis, aim or objective for the study (85%), and clearly described the main findings of the study (90%). All three interventional studies clearly described the intervention. However, only 16 (26%) of the observational studies clearly reported a method of measurement for the exposures or risk factors of interest.

Internal validity

Assessment of internal validity included factors such as blinding and randomization for the interventional studies, statistical analysis methods, and presence or absence of bias.

Of the three interventional studies, only one blinded both subjects and those measuring outcomes to the group assignment (Cleary et al., 1998). The other interventional studies used no blinding in the study design (Rao, Rao, & Starke, 2003; Settle, Wilcox, Fawley, Corrado, & Hawkey, 1998). Cleary et al. also was the only study to include randomization in the assignment of subjects to intervention groups.

Statistical analysis methods used to assess study data were appropriate to the research question and design in most studies (86%). Although a number of the observational studies considered more than one risk factor for the development of CDAD, only 35 studies (57%) described adequate adjustment for the effects of confounding in discussing the main findings of the studies. Twenty-five studies (41%) used multivariate statistical techniques in an effort to control the effects of confounding during data analysis. The most frequently reported type of multivariate data analysis strategy was stepwise logistic regression. Results of stepwise procedures in multivariate analysis are easily misinterpreted because predictors may be excluded which are, in fact, highly correlated with the outcome of interest (Tabachnick & Fidell, 2001). To minimize

this problem, more liberal criterion for inclusion of predictors following univariate analysis ($p \leq .15$ or $.20$) is advised. Only four of the multivariate studies in this review specifically identified variable selection criteria with a univariate significance level greater than $p < .05$ (Komatsu et al., 2003; Kyne, Sougioultzis, McFarland, & Kelly, 2002; Muto et al., 2005; Pulvirenti et al., 2002). The remainder of the studies eliminated variables that might have had an independent association with the development of CDAD following multivariate testing, and indeed limited inclusion of variables that may have clinical relevance. Stepwise procedures hold the allure of statistical criteria for decisions about inclusion of predictors, rather than the tedium of formulating specific hypotheses about the role of multiple variables in association with CDAD. However, such a strategy limits the applicability of the predictive model to clinical practice.

A limitation common to most of the studies within this review is the lack of a consistent case definition for CDAD among studies. Clinically, both the presentation of diarrhea and confirmatory laboratory stool testing or endoscopic examination are considered essential for a diagnosis of CDAD. Although 73% of the studies include some type of change in bowel function as a part of the case definition, the definition of diarrhea varied greatly between studies. This problem of defining diarrhea and its impact on the interpretation of diarrhea study findings has been previously discussed (Bliss, Guenter, & Settel, 1992; Steele, 2006). Diarrhea defined by the number of bowel movements, consistency of stool, and duration of the symptom varies among studies, making meta-analysis unrealistic. Reliable instruments exist for classification of stool characteristics (Bliss, Larson, Burr, & Savik, 2001; Whelan, Judd, & Taylor, 2004). However, their use in retrospective designs that rely upon medical record documentation

is impractical. More than half of the studies in this sample were of a retrospective cohort or case-control design. Confirmatory testing was part of the case definition for 83% of the original studies. However, a variety of testing methods were used, making meta-analysis, as advocated by Bignardi, questionable. Two studies assigned subjects to case status based only on the diagnosis recorded in the medical record by a clinician (Buchner & Sonnenberg, 2001; Harbarth, Samore, & Carmeli, 2001) and a third publication, a letter to the editor reporting research findings, did not include details regarding case definition (Iizuka et al., 2004).

A large portion of the studies relied upon laboratory records of diagnostic stool testing for the selection of subjects and cases. This strategy assumes that all specimens submitted for *C. difficile* testing are from symptomatic individuals, and that all persons without stool testing are both asymptomatic and would produce negative stool testing results. Thus, a type of diagnostic bias was inherent in the design of these studies.

Comparison of studies is also complicated by the variety of ways in which specific risk factor exposure was measured. This is especially evident in assessing antibiotic exposure and severity of illness. Antibiotic exposure was measured by number of drugs, number of doses, number of days on a specific agent, appropriate versus inappropriate use, specific drug regimen, and by a simple binary measure of exposure. Only one study (Al-Eidan, McElnay, Scott, & Kearney, 2000) attempted to use a standardized measure of antibiotic exposure, the Defined Daily Doses established by the World Health Organization for drug utilization research.

Similarly, the concept of severity of illness responsible for hospitalization was measured in a number of ways. The number of medical or surgical procedures, number

of organ systems affected, critical care or ventilator use, prolonged hospital stay and disease staging were all forms of severity assessment. Only two instruments were used for categorization of this variable in a consistent manner. The Horn's Severity of Illness index was used in three studies (Kyne, Sougioultzis, McFarland, & Kelly, 2002; L.McFarland, 1995; Vesta, Wells, Gentry, & Stipek, 2005), while the Charlson Index of Comorbidity was used in two investigations (Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004; Loo et al., 2005). Only Kyne et al. addressed the sensitivity, specificity, and predictive value of the tool in discussion of the findings.

External validity

Subjects chosen for the studies within this review were described as representative of the target population in 40 (66%) publications. Twenty studies (33%) described inclusion criteria for study eligibility and an additional 20 described both inclusion and exclusion criteria for participation. For this reason, generalization of study findings to a specific target population is rather limited.

Funding disclosure

Only 13 (21%) publications acknowledged a funding source or study sponsorship either within the body of the paper or the notes and acknowledgements. This may be due to lack of sponsorship, journal standards and restrictions regarding acknowledgements, or simply reflective of the period of time in which the studies were published. In the checklist proposed by Downs and Black (1998), there is no question regarding sponsorship. However in the 2002 report by West et al. for the Agency for Healthcare Research and Quality, sponsorship is considered an essential criterion for evaluating quality. Of the 42 studies published before 2003, only 14% include funding information,

but 37% of those published in 2003 or later include this information. It is anticipated that this percent will increase over time as qualitative assessment of research findings becomes more refined.

Quantitative Assessment of Research Findings

In order to evaluate the strength of the association between specific ecological variables and the development of CDAD, published odds ratio (O.R.) and relative risk (R.R.) data were systematically examined similar to the method used in the qualitative portion of the meta-analysis publication (Bignardi, 1998). When O.R. or R.R. data was not provided, it was calculated, if possible, from data within the published article. Evidence supportive of the association of a risk factor with the development of CDAD was defined either of the following: (1) a statistically significant ($p < 0.05$) positive univariate O.R. or R.R. in the majority of studies using univariate analysis to evaluate the risk factor or (2) a statistically significant ($p < 0.05$), positive multivariate O.R. Evidence was considered non-supportive of an association if it failed to meet either of the above criteria or if two or more studies using multivariate analysis failed to identify a statistically significant positive association for the risk factor.

Age

The effect of age upon the development of CDAD in hospitalized populations is uncertain. Within this body of literature, age has been studied as a confounder, a risk factor, and an effect modifier. Age is often considered a confounding variable in epidemiological investigations. Eight studies used matching to control for potential confounding effects of age, thus eliminating it for consideration as a risk factor for the development of CDAD (Bliss et al., 1998; Changela et al., 2004; Cunningham, Dale,

Undy, & Gaunt, 2003; Kreisel, Thomas, Silver, & Cunningham, 1995; Loo et al., 2005; MacGowan et al., 1997; Thamlikitkul, Danpakdi, & Chokloikaew, 1996; Vesta, Wells, Gentry, & Stipek, 2005; Yip, Loeb, Salama, Moss, & Olde, 2001). An additional study did not use matching for age, but selected controls within the same age-range as case patients, thereby minimizing the possibility of differences in age between cases and controls (Changela et al., 2004).

Twenty-eight publications included age as a variable in the statistical analysis of study findings. Eight publications reported statistically significant differences in mean age between CDAD subjects and non-CDAD comparison subjects within the presentation of demographic data (Ackermann et al., 2005; Barbut et al., 2005; Buchner & Sonnenberg, 2002; Climo et al., 1998; Harbarth, Samore, & Carmeli, 2001; Kent, Rubin, Wroblewski, Hanff, & Silen, 1998; McFarland, 1995; Svenungsson, Lagergren, & Lundberg, 2001).

Age was evaluated as a specific risk factor for CDAD in 13 studies. Of the five studies reporting only univariate analysis, four reported statistically significant positive associations with CDAD (Ackermann et al., 2005; Karlstrom, Fryklund, Tullus, Burman, & Group, 1998; Kyne, Sougioultzis, McFarland, & Kelly, 2002; Watanakunakorn, Watanakunakorn, & Hazy, 1996). Increasing age was found to have a statistically significant positive association in five additional studies using multivariate data analysis (Andrews, Raboud, Kassen, & Enns, 2003; Chang & Nelson, 2000; Modena, Bearely, Swartz, & Friedenber, 2005; Muto et al., 2005; Starr, Martin, McCoubrey, Gibson, & Poxton, 2003). Although Harbarth et al. (2001) reported a univariate O.R. of 2.5 with a significance level sufficient for entry into multivariate analysis ($p = .08$), age was not

entered as a variable in the logistic regression analysis. Only one publication within this review reported a non-significant association (Kyne et al., 1999). Therefore, the findings of this review are consistent with those of Bignardi (1998) who reported substantial evidence for increasing age as a risk factor for CDAD.

A problem noted in comparing research findings regarding this variable is the disparity in ages considered reflective of advanced age. While some investigations considered subjects 60 years and older to be geriatric, others used 70 or 75 years as a cut-point. None of the published studies included information about why a particular age range was selected as a risk factor for data collection and analysis.

It is likely that age is also an effect modifier for development of CDAD. As age increases, the likelihood of exposure to toxigenic strains of the organism increases, principally through institutionalization in a hospital, rehabilitation, or long-term care setting. Institutionalization will not be investigated as a potential risk factor for CDAD in the present study because all subjects will be inpatients within an acute care hospital. Several investigations within this review reported significant differences in CDAD rates for nursing home versus community-dwelling adults (Al-Eidan, :McElnay, Scott, & Kearney, 2000; Cooper, Lederman, & Salata, 1995; Kent, Rubin, Wroblewski, Hanff, & Silen, 1998) . A descriptive study demonstrating a reduction in CDAD within geriatric wards of several acute care hospitals following implementation of an enhanced infection control program suggests that infection exposure may play a larger role than age in disease development (Stone, Beric, Quick, Balestrini, & Kibbler, 1998). Treatment of chronic illnesses is more common in old age, and acute exacerbations and complications increase the likelihood of multiple drug exposures, including antibiotics. A significant

difference has been demonstrated in the mean age of subjects exposed to antibiotics prior to the development of CDAD and those with no previous antibiotic exposure (Svenungsson, Lagergren, & Lundberg, 2001). Aging also makes an individual more likely to be exposed to tube feeding as a substitute or supplement to oral feeding. This type of artificial nutrition is associated with major changes in fecal flora (Schneider et al., 2000) and has been identified as a significant risk factor for both acquisition of the organism and development of CDAD (Bliss et al., 1998; Komatsu et al., 2003).

Acid Suppression

The administration of drugs to suppress or neutralize gastric acid production has been explored as a biologically plausible risk factor for the development of CDAD. Bignardi (1998) found substantial evidence for use of “anti-ulcer medication including antacids and H2 blockers” (p.5). A total of 16 studies were identified which considered alteration of gastric acid as a risk factor. However, within this review, acid suppressive medications were examined in three separate categories: antacids, histamine-2 blocking agents (H2), and proton pump inhibitors (PPI), as each class of drugs has a distinctly different physiologic mechanism of action.

Antacid medications were studied in six publications. Two were analyzed using univariate techniques only, while four described multivariate data analysis. Non-significant findings were reported for five studies (Aziz, Ayis, Gould, & Rawlins, 2001; Barbut et al., 1997; Hornbuckle et al., 1998; Kyne et al., 1999; Watanakunakorn, Watanakunakorn, & Hazy, 1996). Only one of these publications reported statistically significant positive associations for antacid medication use (Tacconelli et al., 1998).

Of the nine studies measuring exposure to H2 blocking agents, three reported non-significant univariate test results (Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004; Loo et al., 2005; Watanakunakorn, Watanakunakorn, & Hazy, 1996) and another reported non-significant multivariate results (Kyne et al., 1999). A high-magnitude statistically significant positive association was identified in one univariate study (Yip, Loeb, Salama, Moss, & Olde, 2001) and a more moderate significant multivariate association was reported by Muto (2005).

The use of proton pump inhibitor (PPI) agents has generated the most consistent associations for acid suppressive exposure across studies. Ten publications in this review assessed the relationship between PPI drugs and CDAD. Three univariate analyses produced statistically significant positive associations (Al-Tureihi, Hassoun, Wolf-Klein, & Isenberg, 2005; Cunningham, Dale, Undy, & Gaunt, 2003; Yearsley et al., 2006), but Kyne (1999) and Loo (2005) both reported non-significance. Two reports indicated statistically significant multivariate O.R. for PPI exposure (Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004; Muto et al., 2005).

All of the studies investigating acid suppression as a risk factor for CDAD measured exposure dichotomously; therefore, a dose response could not be assessed from this review. No evidence was found to suggest that antacids, which neutralize the pH level of gastric secretions, are associated with the development of CDAD. However, H2 blockers and PPI agents, which exert an effect at the cellular level and result in a decrease in acid production, have some evidence to suggest their influence. Future research should consider these as separate drug categories for data collection, and an attempt should be made to determine if there is a dose-response for either type of agent.

Antibiotic Administration

The risk factor for CDAD development investigated the most extensively over the past thirty years is antibiotic exposure. The scientific literature includes studies of both the amount of antibiotic exposure associated with CDAD and the specific antibiotics most likely to be associated with the disease. Although one meta-analysis reported a significant association between antibiotic exposure and CDAD (Bignardi, 1998), a systematic review of published epidemiologic studies between 1978 and 2001 identified threats to internal validity in a majority of the studies due to incorrect control group selection, inadequate sample sizes, inadequate control of confounders, and misclassification bias in case identification (Thomas, Stevenson, & Riley, 2003).

The original literature search identified 66 publications in which antibiotic exposure was considered as a risk factor for the development of CDAD. In addition to a meta-analysis and structured review, there were 64 original research publications identified. Of these studies, seven were descriptive and provided no information about the disease frequency, the role of chance, or the strength of the association (Bulstrode et al., 1997; Crabtree, Pellitier, Gleason, Pruett, & Sawyer, 1999; Gorecki, Schein, Rucinski, & Wise, 1999; Impallomeni, Galletly, Wort, Staff, & Rogers, 1995; Khan & Cheesbrough, 2003; Shek, Stacey, Rendell, Hellier, & Hanson, 2000; Stone, Beric, Quick, Balestrini, & Kibbler, 1998).

A major difficulty in assessing the strength of evidence regarding antibiotic exposure as a CDAD risk factor is the multiple ways in which exposure has been operationalized and measured. More than 25% of the studies measured antibiotic exposure via more than one method. The most frequently described measure, reported in

41 papers, and was a simple binary assessment of exposure to a specific antibiotic drug classification. Twenty-two papers measured any antibiotic exposure, while 12 measured the number of different drug agents used. Three studies examined specific antibiotic regimens and two classified antibiotic use as “appropriate” or “inappropriate”.

Surprisingly, only four of the publications attempted to quantify antibiotic exposure by measuring the number of doses, and only one did so through the use of a standardized dose measure. A unit of measure, the defined daily dose (DDD), represents the average daily adult dose for a drug when used for its primary indication (Pelle, Gilchrist, Lawson, Jacklin, & Franklin, 2006). This measure has been advocated for drug exposure research by the World Health Organization to facilitate comparisons of drug usage despite international variations in clinical practice patterns. Only one of the studies included in this review used the DDD unit for measuring antibiotic exposure, although the system has been in effect for over 20 years (Al-eidan).

In considering all antibiotic exposure as a whole, study results present conflicting evidence. Bignardi (1998) reported a pooled O.R. of 5.9 for a binary measure of antibiotic exposure. One multivariate study published after 1998 (Cunningham, Dale, Undy, & Gaunt, 2003) and five univariate studies reported statistically significant positive associations for antibiotic exposure (Blot et al., 2003; Loo et al., 2005; Starr, Martin, McCoubrey, Gibson, & Poxton, 2003; Svenungsson, Lagergren, & Lundberg, 2001; Yearsley et al., 2006). This provides strong evidence for antibiotic exposure as a consistent and independent risk factor for CDAD acquisition.

In contrast, four studies published after the Bignardi paper reported non-significant multivariate O.R. for this same variable (Chang & Nelson, 2000; Harbarth,

Samore, & Carmeli, 2001; Loo et al., 2005; McCusker, Harris, Perencevich, & Roghmann, 2003). Two case-control (Komatsu et al., 2003; Vesta, Wells, Gentry, & Stipek, 2005) and two prospective cohort studies (Cleary et al., 1998; Kyne, Sougioultzis, McFarland, & Kelly, 2002) did not enter antibiotic exposure into multivariate analysis based upon overly stringent bivariate test results. Barbut et al. (2005) did not include antibiotic exposure in the multivariate analysis due to both low magnitude and non-significance on univariate testing. In a sample of oncology patients, antibiotic exposure was associated with CDAD only in patients also receiving chemotherapy (Blot et al., 2003). Both treatment with any antibiotic and treatment with specific classes of antibiotics were entered into logistic regression models in a prospective cohort study, and both failed to demonstrate statistical significance (Bliss et al., 1998). This discrepancy in findings indicates the need for a more accurate and quantitative means of measuring antibiotic exposure in future research.

A number of studies did attempt to operationalize the burden of antibiotic exposure through measurement of the number of different antibiotic agents to which subjects were exposed. Three studies reported statistically significant differences between cases and comparison subjects in the mean number of antibiotic agents used, but these studies did not report measures of association or provide sufficient information within the publication to calculate these measures (Climo et al., 1998; Kyne, Sougioultzis, McFarland, & Kelly, 2002; Loo et al., 2005). Pulvirenti et al. (2002A) reported a statistically significant, positive multivariate O.R. for the number of antibiotics. Dial et al. (2004) also reported a statistically significant, positive multivariate O.R.s for the number of antibiotics in a cohort study, but a case-control study reported in

the same paper did not have statistically significant findings. Statistically significant positive univariate associations were reported by five other studies (Andrews, Raboud, Kassen, & Enns, 2003; Harbarth, Samore, & Carmeli, 2001; Lai, Melvin, Menard, Kotilainen, & Baker, 1997; L. V. McFarland, 1995; J.J. Pulvirenti et al., 2002).

Assessment of antibiotic burden measured based on a simple count of the number of doses has not yielded clinically important information. No significant difference was found in the mean number of antibiotic doses received between cases and comparison subjects in one investigation (MacGowan et al., 1997), and Kyne et al. (1999) reported a positive, but not statistically significant multivariate O.R. for the number of antibiotic doses. It would appear that a count of doses administered is not useful in quantifying antibiotic burden. Al-Eidan et al. (2000) attempted to quantify doses using the Defined Daily Dose system. However, the number of doses used for each drug were calculated based upon Pharmacy purchasing data, and were not obtained from individual patient records. Therefore the actual degree of antibiotic exposure for individual subjects was not accurately measured.

Within this review, researchers identified a number of antimicrobial drug categories as exposures associated with CDAD. These findings are summarized in Table 3. Antibiotic agents are grouped by their pharmacologic drug category in column 1. Studies reporting non-significant associations are listed in column 2, those reporting statistically significant univariate associations are listed in column 3, and studies reporting statistically significant multivariate associations are listed in column 4.

Table 3

Reported Significance of Test Results for Antibiotic Exposure by Drug Category

Significance of the Association with CDAD			
Drug Category	Non-significant	+ Univariate	+ Multivariate
Aminoglycosides	Johnson et al., 1999; Komatsu et al., 2003 Muto et al., 2005 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005 Shah, Lewis, Leopold, Dunstan, & Woodhouse Vesta, Wells, Gentry, & Stipek, 2005 Watanakunakorn, Watanakunakorn, & Hazy, 1996 Yip, Loeb, Salama, Moss, & Olde, 2001	Barbut et al., 1997 Bliss et al., 1998 Changela et al., 2004 Cooper, Lederman, & Salata, 1995 Halim, Peterson, Friesen, & Ott, 1997 Harbarth, Samore, & Carmeli, 2001 Kyne, 2002 Schwaber et al., 2000* Talon et al., 1995	Loo et al., 2005*
Penicillins	Aziz, Gould, & Rawlins, 2001 Harbarth, Samore, & Carmeli, 2001 Johnson Komatsu et al., 2003 Kyne et al., 1998 Kyne et al., 1999 Loo et al., 2005 McCusker, Harris, Perencevich, & Roghmann, 2003 Mody, Smith, & dever, 2001 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005 Shah, Lewis, Leopold, Dunstan, & Woodhouse Vesta, Wells, Gentry, & Stipek, 2005 Watanakunakorn, Watanakunakorn, & Hazy, 1996 Yip, Loeb, Salama, Moss, & Olde, 2001	Al-Eidan, McElnay, Scott, & Kearney, 2000 et al., 2000 Changela Cooper, Lederman, & Salata, 1995 Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 Rao, Rao, & Starke, 2003 Schwaber et al., 2000 Starr, Martin, McCoubrey, Gibson, & Poxton, 2003 Talon et al., 1995	Barbut et al, 1997 Talon et al., 1995
Cephalosporins	Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 Johnson et al., 1999 Komatsu et al., 2003 Kyne et al., 199	Al-Eidan, McElnay, Scott, & Kearney, 2000; Arrango et al., 2006 Barbut et al., 1997 Changela et al., 2004 Cooper, Lederman, &	Aziz, Gould, & Rawlins, 2001 Bliss et al., 1998 Loo et al., 2005 Yip, Loeb, Salama, Moss, & Olde, 2001

Table 3 (Continued)

Drug Category	Significance of the Association with CDAD		
	Non-significant	+ Univariate	+ Multivariate
Cephalosporins - continued	<p>McCusker, Harris, Perencevich, & Roghmann, 2003</p> <p>Mody, Smith, & dever, 2001 (1st generation)</p> <p>Muto et al., 2005 (1st and 2nd generation)</p> <p>Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005</p> <p>Vesta, Wells, Gentry, & Stipek, 2005</p>	<p>Salata, 1995</p> <p>Halim, Peterson, Friesen, & Ott, 1997</p> <p>Harbarth, Samore, & Carmeli, 2001</p> <p>Kyne et al., 1998</p> <p>Mody, Smith, & dever, 2001 (3rd generation)</p> <p>Muto et al., 2005 (3rd and 4th generation)</p> <p>Rao, Rao, & Starke, 2003</p> <p>Schwaber et al., 2000</p> <p>Settle, Wilcox, Fawley, Corrado, & Hawkey, 1998</p> <p>Shah, Lewis, Leopold, Dunstan, & Woodhouse Starr, Martin, McCoubrey, Gibson, & Poxton, 2003</p> <p>Talon et al., 1995'</p> <p>Tumbarello, Tacconelli, Leone, Cauda, & Ortona, 1995</p> <p>Watanakunakorn, Watanakunakorn, & Hazy, 1996</p>	
Quinolones	<p>Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 (case control)</p> <p>Komatsu et al., 2003</p> <p>Kyne et al, 1998</p> <p>Kyne, Sougioultzis, McFarland, & Kelly, 2002</p> <p>Mody, Smith, & dever, 2001</p> <p>Muto et al., 2005 (ciprofloxacin)</p> <p>Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005</p> <p>Rao, Rao, & Starke, 2003</p> <p>Shah, Lewis, Leopold, Dunstan, & Woodhouse</p> <p>Talon et al., 1995</p> <p>Watanakunakorn, Watanakunakorn, & Hazy, 1996</p>	<p>Barbut et al., 1997*</p> <p>Bilgrami et al., 1999*</p> <p>Changela</p> <p>Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 (cohort)</p> <p>Harbarth, Samore, & Carmeli, 2001</p> <p>Muto et al., 2005 (levofloxacin)</p> <p>Pulvirenti, 2002A</p> <p>Schwaber et al., 2000</p>	<p>Loo et al., 2005</p> <p>McCusker, Harris, Perencevich, & Roghmann, 2003</p> <p>Yip, Loeb, Salama, Moss, & Olde, 2001</p>

Table 3 (Continued)

Drug Category	Significance of the Association with CDAD		
	Non-significant	+ Univariate	+ Multivariate
Macrolides	Harbarth, Samore, & Carmeli, 2001 Komatsu et al., 2003 Kyne, 1998 Loo et al., 2005 Mody, Smith, & dever, 2001 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005 Shah, Lewis, Leopold, Dunstan, & Woodhouse Watanakunakorn, Watanakunakorn, & Hazy, 1996	Changela et al., 2004 Muto et al., 2005 Pulvirenti et al., 2002A Schwaber et al., 2000	
Clindamycin	Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 (case-control study) Komatsu et al., 2003 Kyne et al., 1998 Kyne, Sougioultzis, McFarland, & Kelly, 2002 McCusker, Harris, Perencevich, & Roghmann, 2003 Watanakunakorn, Watanakunakorn, & Hazy, 1996 Yip, Loeb, Salama, Moss, & Olde, 2001	Changela Harbarth, Samore, & Carmeli, 2001 Johnson Mody, Smith, & dever, 2001 Muto et al., 2005 Rao, Rao, & Starke, 2003 Schwaber et al., 2000 Thamliktkul, Danpakdi, & Chokloikaew, 1996 Tumbarello, Tacconelli, Leone, Cauda, & Ortona, 1995	Barbut, 1997 Chang & Nelson, 2000 Loo et al., 2005 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005
Sulfonamides	Aziz, Gould, & Rawlins, 2001 Harbarth, Samore, & Carmeli, 2001 Kyne, 2002 Shah, Lewis, Leopold, Dunstan, & Woodhouse Yip, Loeb, Salama, Moss, & Olde, 2001	Barbut et al., 1997* Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 Muto et al., 2005 Schwaber et al., 2000* Tumbarello, Tacconelli, Leone, Cauda, & Ortona, 1995	
Tetracyclines	Changela et al., 2004 Kyne, Sougioultzis, McFarland, & Kelly, 2002 Muto et al., 2005		
Carbapenems	Komatsu et al., 2003 Loo et al., 2005 Muto et al., 2005	Changela et al., 2004 Harbarth, Samore, & Carmeli, 2001	

Table 3 (Continued)

Drug Category	Significance of the Association with CDAD		
	Non-significant	+ Univariate	+ Multivariate
Glycopeptides	Johnson et al., 1999 Komatsu et al., 2003 Loo et al., 2005 Mody, Smith, & dever, 2001 Muto et al., 2005 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005 Shah, Lewis, Leopold, Dunstan, & Woodhouse Watanakunakorn, Watanakunakorn, & Hazy, 1996	Arrango et al., 2006 Barbut et al., 1997 Changela et al., 2004 Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 Schwaber et al., 2000	
Metronidazole	Kyne, Sougioultzis, McFarland, & Kelly, 2002 Mody, Smith, & dever, 2001 Palmore, Sohn, Malak, Eagan, & Sepkowitz, 2005 Watanakunakorn, Watanakunakorn, & Hazy, 1996 Wren, Ahmed, Jamal, & Safadi, 2005	Barbut et al., 1997* Chakrabarti, Lees, Jones, & Milligan, 2000 Dial, Alrasadi, Manoukian, Huang, & Menzies, 2004 Pulvirenti et al., 2002A Pulvirenti et al., 2002B Schwaber et al., 2000	

Statistically significant positive O.R. with magnitude > 1.0

Within the penicillin class, results were very varied. Two studies demonstrated independence for penicillin exposure, and seven demonstrated consistency. The overall number of studies with non-significant findings, however, outnumbers those with significant findings. The penicillin class of drugs is one in which successive generations have been developed in an attempt to avoid drug resistance. For this reason, a simple grouping of all penicillin's may not be adequate for epidemiological purposes. Future research regarding drug classes and exposure should distinguish these groupings in addition to determining if there is a dose-response to sub-group penicillin.

Cephalosporin antibiotics were investigated in the largest number of studies. Four reported independent associations after multivariate analysis, and 17 reported significant univariate findings, considerably more than the nine that reported non-significant findings. There is strong evidence to support cephalosporin drug exposure as a risk factor for CDAD.

Within the quinolone class of antibiotics, there were three studies reporting independent association with quinolone exposure after multivariate analysis to control for confounders. There were also seven univariate studies with statistically significant results. However, the author also reviewed 12 studies that reported negative findings for quinolone as a risk factor. Future research needs to examine this exposure more fully. The quinolones represent a clinically important category of antibiotic, and their use as a risk factor has been investigated in several hospital outbreaks.

Macrolide antibiotics did not have sufficient evidence in this review to consider them a substantial risk factor in the development of CDAD. There were no studies that met the criterion of independence, and only four that met the criterion of consistency. There were seven studies that reported non-significant results for this risk factor.

Not surprisingly, evidence continues to support clindamycin as a risk factor for CDAD. Although there were seven studies reporting non-significant results for clindamycin exposure within this body of literature, these studies were overshadowed by those with significant findings. Four studies reported significant multivariate results and nine reported significant univariate results for clindamycin exposure.

The category of carbapenems had no reports with multivariate support, and an even number of reports with significant and non-significant findings. There is not

sufficient evidence at this time to consider the carbapenems a serious risk factor for the development of CDAD.

Less than half of the studies that investigated glycopeptide antibiotic use as a risk factor reported statistically significant univariate measures of association, and there were no studies with significant multivariate findings. This is not surprising, since vancomycin, a major glycopeptide used to treat recurrent or resistant CDAD is in this category of drug. However, the assessment of evidence regarding metronidazole was unexpected. Metronidazole is considered by many clinicians to be the first line drug used to treat CDAD cases that fail to respond to withdrawal of antibiotic therapy. Studies with significant and non-significant measures of association for metronidazole use were almost equally divided.

The most difficult aspect of evaluating the association of CDAD with antibiotic exposure is the difficulty in establishing a dose response. Inconsistent methods of measuring antibiotic exposure complicate comparisons between studies. Median number of doses given to a specific subject, daily average antibiotic doses dispensed by an institutional pharmacy, and simple frequency counts of the number of subjects receiving a particular antibiotic agent have all been used to measure antibiotic exposure. Although biologic markers, such as serum drug levels, are not available for all antimicrobial drugs, even when such testing is available, it is not included in published descriptions of antibiotic exposure, and is not possible in any type of retrospective study.

Diet

Tube feeding has been investigated as a risk factor for the development of CDAD, as has malnutrition and hypoalbuminemia. Within this review, 12 studies were located that examined tube feeding and six considering nutritional deficits as a risk factor.

Tube Feeding. Twelve analytical epidemiology studies were located that considered tube feeding via either nasogastric or percutaneous abdominal routes as a potential risk factor for the acquisition of CDAD. A letter to the editor describing data supporting elemental diets as a risk factor for CDAD was excluded due to lack of details (Iizuka et al., 2004).

Of the two studies which analyzed data using only univariate analysis, one reported data sufficient to calculate an O.R. of 2.38 ($p < 0.05$) (Halim, Peterson, Friesen, & Ott, 1997), and the other reported a non-significant positive association (Watanakunakorn, Watanakunakorn, & Hazy, 1996). Therefore, satisfaction of the consistency criterion was unclear.

Ten studies used multivariate data analysis methods. Of these studies, three evaluated enteral feeding as a variable and determined that it was sufficiently related to other variables to eliminate for consideration as an independent risk factor (Kyne, Sougioultzis, McFarland, & Kelly, 2002; Loo et al., 2005; Shah, Lewis, Leopold, Dunstan, & Woodhouse, 2000). Two studies reported non-significant O.R. (Kent, Rubin, Wroblewski, Hanff, & Silen, 1998; Yip, Loeb, Salama, Moss, & Olde, 2001), although the specific values were not published. Four studies found enteral feeding to be an independent risk factor for CDAD (D. Bliss, Guenter, & Settel, 1992; Buchner & Sonnenberg, 2001; Komatsu et al., 2003; Talon et al., 1995), and a fifth found enteral

feeding by nasogastric tube, but not via percutaneously inserted gastrostomy tube (Kyne et al., 1999) to be independently associated with the disease. Lai reported a statistically significant negative association between the presence of a nasogastric tube and CDAD. Enteral feeding appears to be a substantial risk factor for CDAD. However, since this mode of feeding cannot exist without the presence of the administration tube, the possibility that enteral feeding is a confounder, rather than a risk factor, should be considered. Bignardi (1998) identified “nasogastric tubes” as a substantial risk factor, but did not address enteral feeding. An alternative explanation for the apparent association between enteral feeding and CDAD has been proposed as “feeding tubes are frequently given to immune compromised patients with dietary deficiencies and multiple other comorbid conditions who have spent appreciable amount of time in the hospital. Most importantly, all percutaneous tubes are inserted under antibiotic prophylaxis”(Sonnenberg, 2005).

Malnutrition. Of the studies that examined nutritional deficits in association with CDAD, four used univariate analysis methods only. Of these studies, three reported statistically significant positive associations between CDAD and malnutrition (Al-Tureihi, Hassoun, Wolf-Klein, & Isenberg, 2005; Andrews, Raboud, Kassen, & Enns, 2003; Rubin, Bodenstein, & Kent, 1995) as measured by serum hypoalbuminemia. The other report measured malnutrition via the Prognostic Nutritional Index, a calculation based on a number of laboratory and clinical factors (Dansinger et al., 1996) and identified an association between protein-losing enteropathy and CDAD.

Four studies also reported multivariate testing, however the Modena et al. and Andrews et al. studies omitted entry of the nutritional variable into multivariate analysis

due to excess missing data points. Buchner and Sonnenberg reported a statistically significant O.R. of 2.01 for the variable of malnutrition as an independent predictor of CDAD, but Shah did not report the multivariate O.R., indicating that it was no longer statistically significant.

The loss of serum protein identified by Dansinger et al. raises important questions about the relationship between malnutrition, susceptibility to infection, and diarrhea. This variable should be considered a substantial risk factor for inclusion in future research.

Cathartic Administration

The impact of laxatives and bowel preparation regimens upon development of CDAD has not been extensively studied. Among the publications reviewed, six included data regarding laxative use and one regarding bowel preparation regimens.

A statistically significant R.R. of 3.2 was reported in association with mechanical bowel preparation among a cohort of surgical patients (McCarter, Abularrage, Velasco, Davis, & Daly, 1996). This risk factor was not included in the Bignardi list, but most likely warrants inclusion in future studies.

Six studies investigated laxative use in association with the development of CDAD. Watanakunakorn et al. (1996) reported a non-significant negative association in a study of a general hospital population. However, an investigation of CDAD generated from an HIV-infected population identified a statistically significant positive association (Pulverenti et al., 2002). Four publications included laxative use in multivariate studies. Barbut et al. found a negative association between CDAD and laxative use on univariate testing, but did not include the variable in multivariate analysis, although the p value of

0.20 would have been sufficient for inclusion in a conditional logistic regression analysis. Kent et al. (1998) also reported a non-significant p value, and therefore did not include laxative use in the regression analysis. Kyne et al. (1999) reported a non-significant negative association. Only one paper reported statistical significance after multivariate analysis, but it was a negative association for both the general drug category of “laxatives” as well as the specific agent lactulose (Shah, Lewis, Leopold, Dunstan, & Woodhouse, 2000).

Laxative use does not appear to increase the risk of CDAD. It is possible that laxative use may exert a protective effect upon the bowel by facilitating the elimination of potentially toxigenic organisms. It may, however, serve as a confounder in studies in which subjects are selected based upon hospitalized persons who have stools submitted for laboratory testing due to loose or diarrheal stools. Bowel preparation is associated with CDAD in surgical patients, but this needs more study to determine if the association is consistent across more populations.

Severity of Illness

The immune response to infectious agent exposure can be impaired by a variety of mechanisms, including disease and injury. A number of investigations have explored the concept of severity of illness in association with the development of CDAD. A total of 22 studies were included in the review that explored the association between concomitant illness or injury and CDAD. The concept was operationalized in a variety of ways, including comorbidity, stage of illness, hospital length of stay, intensive care unit stay, and a severity of illness index (Horn & Horn, 1986).

The most frequently used method used to measure severity of illness within this group of studies was comorbidity. Five studies considered the presence or absence of specific diseases as a measure of comorbidity. Significant positive univariate associations were reported for chronic renal failure (Cunney, Magee, McNamara, Smyth, & Walshe, 1998; Harbarth, Samore, & Carmeli, 2001) and renal insufficiency (Rubin, Bodenstein, & Kent, 1995). Although Yip et al. (2001) reported a non-significant univariate O.R. for renal failure, the magnitude association was 7.0. Dial et al. also reported a significant positive multivariate O.R. of 4.3 for chronic renal failure. Other statistically significant positive univariate associations have been reported for gastrointestinal illness and anemia (Harbarth, Samore, & Carmeli, 2001), chronic obstructive pulmonary disease (Rubin, Bodenstein, & Kent, 1995) and malignancy (Rubin, Bodenstein, & Kent, 1995; Yip, Loeb, Salama, Moss, & Olde, 2001).

Andrews et al. (2003) reported a significant positive multivariate O.R. for increasing number of organ systems affected by comorbid illness. Two reports indicated a statistically significant difference in the number of diagnoses between CDAD cases and controls (Buchner & Sonnenberg, 2001; Mody, Smith, & Dever, 2001), but measures of association were not reported. Likewise, Buchner and Sonnenberg reported statistically significant differences in the number of medical and surgical procedures performed on cases versus controls.

A problem common to all of these studies is the fact that there between subjects, there may be a great deal of variation in the impact in which a medical diagnosis makes on the overall health of the individual. Therefore, a diagnosis category, or a count of diagnoses, is not a valid measure of comorbidity. When a standardized measure, the

Charlson Index of comorbidity, was used to assess differences between CDAD cases and a comparison group, no significant differences were found (Loo et al., 2005).

The stage of illness for Human Immunodeficiency Virus (HIV)-infected adults was examined in two separate studies. Although Pulverenti et al. (2005) found positive associations through progressive stages of HIV infection, these associations were not statistically significant on univariate analysis. However, low CD4 cell count has been significantly associated with CDAD in univariate analysis (Tacconelli et al., 1998), and both a history of opportunistic infection (Pulverenti et al., 2005) and a CD4 count below 50 mm^3 (Barbut et al., 1997) have been reported to have statistically significant positive associations following multivariate analysis. These findings provide biologically plausible support for immunosuppression, manifested by both opportunistic infection and a decline in CD4, as a factor in the causation of CDAD.

The length of hospitalization is significantly longer for persons who develop CDAD (Buchner & Sonnenberg, 2002; MacGowan et al., 1997; Pulverenti et al., 2002). However, length of stay data does not indicate whether the prolonged hospitalization is a risk factor or a consequence of CDAD. Three studies suggest that a prolonged hospital stay increases risk for the disease. Significant positive multivariate associations have been reported for prolonged post-operative hospital stay (Harbarth, Samore, & Carmeli, 2001), hospitalization greater than or equal to eight days during a risk period (Pulvirenti et al., 2002), and hospitalization more than ten days in the month preceding infection (Pulvirenti et al., 2002). Although an increasing length of stay increases exposure to microbes in the environment, its sensitivity and specificity as a measure of CDAD risk is uncertain. It may represent a confounder, rather than a risk factor.

Modena et al. defined severity of illness in terms of both mechanical ventilation and the need for care within an intensive care unit (ICU) (2005). Mechanical ventilation use had a significant positive association with CDAD in univariate analysis and an ICU stay was statistically significant in multivariate analysis. This categorical data lacks precision. Like length of hospital stay, it may represent a confounding variable rather than a risk factor.

McFarland (1995) provided the earliest publication in this review to use the Horn Severity of Illness Index as a measure. Statistically significant differences in severity index score were reported within the publication. Relative risks calculated from published data found a statistically significant positive association for the catastrophic level of illness, but not for the severe level. Kyne (2002) reported statistically significant positive associations for both the catastrophic and severe levels of illness. Kyne also examined the predictive value of the instrument with a score of three or more and reported sensitivity 79-87, specificity 39-73, positive predictive value 11-27 and negative predictive value 96-97. More recently, Vesta (2005) published study data that enabled calculation of a statistically significant O.R. of 7.67 for catastrophic illness. There is strong evidence that illness that weakens the body's immune system increases the risk of CDAD. Although still a categorical measure of severity of illness, the Horn Index presents a more precise measure of risk than a simple binary variable.

Use of a measure such as the Horn index is more appropriate in the development of an ecological model for the study of CDAD that is relevant to nursing practice. Such an instrument attempts to quantify the human response to illness, rather than labeling the disease process.

Ecological Model for Study of CDAD

The conceptual model for this study is derived from a model created for the study of ecosystems as they affect human health (Koren & Crawford-Brown, 2004). This model considers the macroenvironment, the external environment within which a potential host functions, and the microenvironment, the physiological factors that alter the susceptibility of the host.

The movement of human beings within the healthcare setting creates environmental stressors that can lead to contact, or exposure, between a potential host and a potentially infectious microorganism. Exposure, however, does not automatically result in disease for several reasons. First, the amount of exposure an individual sustains is influenced by the degree of temporal and spatial distance between the area of contamination and the human being. Extrinsic risk factors, such as antibiotic usage policies, infection control compliance, and the use of standardized cleaning procedures are designed to increase the temporal and spatial distance between microorganisms and humans. Second, and perhaps more importantly, the host response is dependent upon intrinsic factors that modulate the immune response through changes in the microenvironment. In the case of CDAD, specific factors known to have substantial evidence for an association with CDAD are severity of illness, changes in the microflora of the gut due to aging, antibiotic or chemotherapy usage, use of enteral feeding systems, malnutrition, gastric acid suppression, and preoperative bowel preparation regimens. An exposed individual will respond to *Clostridium difficile* exposure by forming antibodies, becoming an unprotected carrier, or by developing active Cd infection in varying degrees.

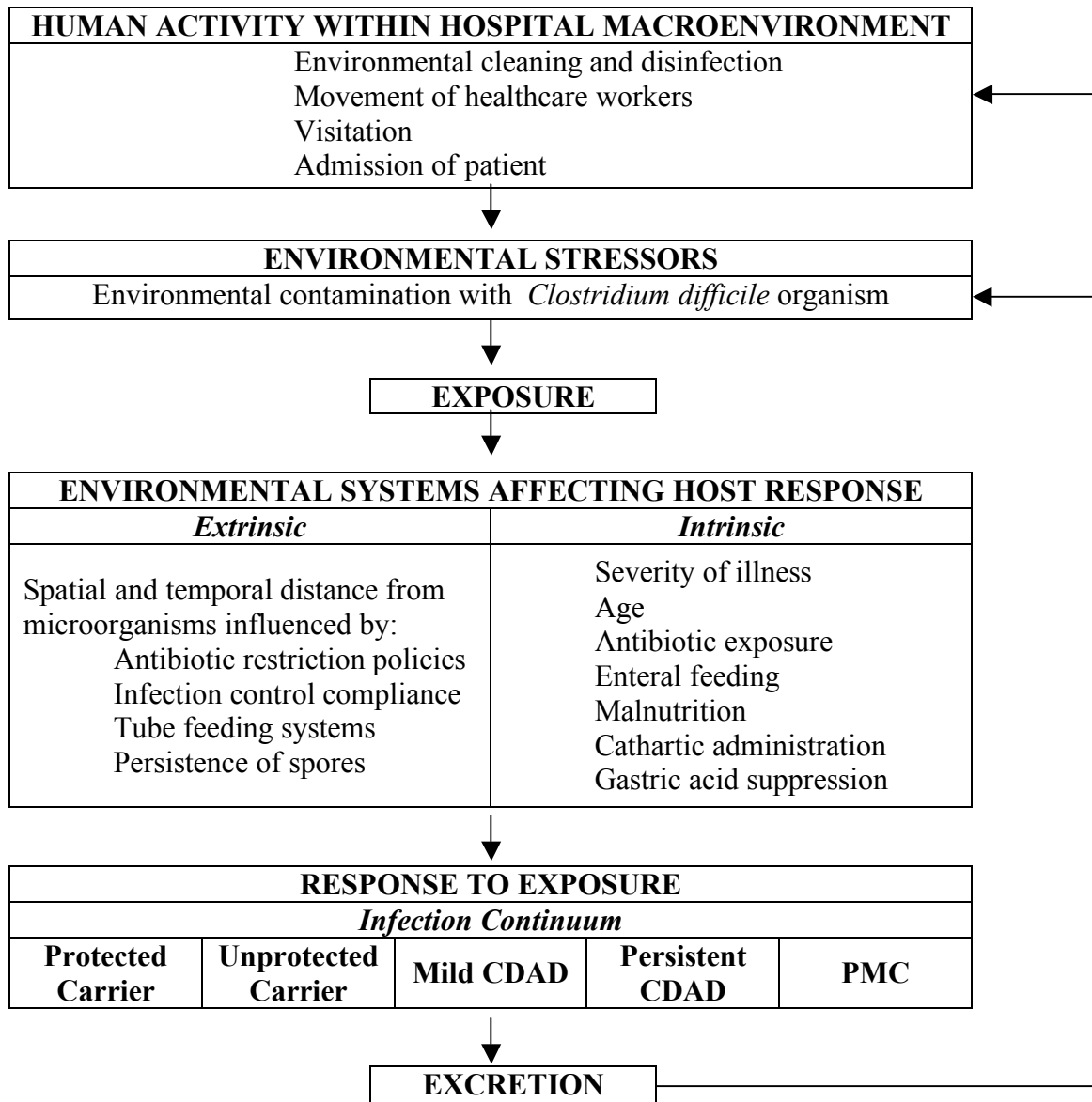


Figure I. Model for the study of ecological variables affecting development of *Clostridium difficile*-associated diarrhea.

This study will focus only on the portion of the model that considers the intrinsic and extrinsic factors that affect a host response to Cd exposure. Because the Cd organism can persist in an environment for prolonged periods of time in its spore state, reliance solely upon extrinsic environmental factor to stop the infection may not be sufficient.

Despite nearly two decades of widespread use in the developed world of personal

protective equipment for the handling of all potentially infectious material, as well as national campaigns to promote handwashing and judicious antibiotic usage, outbreaks of CDAD continue. Therefore, an alternative means of identifying the most susceptible hosts and providing ecologically based care may present of more effective means of controlling this disease.

Summary

This review of literature has identified ecological variables that affect the response of a hospitalized adult exposed to the Cd organism. These variables will be used in the design of a case-control study to identify an ecological model useful for nurses. These variables include age, antibiotic exposure, bowel preparation, enteral feeding, malnutrition, gastric acid suppression, and severity of illness.

Chapter Three

Research Methods

Research Design

The purpose of this study was to develop and test a predictive model for the development of *Clostridium difficile*-associated diarrhea. It is anticipated that such a model can serve three purposes: (1) serve as a foundation for development of a valid and reliable risk assessment tool (2) aid in the design of future clinical trials of nurse-directed prevention strategies, and (3) assist clinicians in modifying infection control practices in institutional settings when caring for high-risk individuals. The study used a case-control design to develop a predictive model to identify ecological risk factors most closely associated with the development of hospital-acquired *Clostridium difficile* associated diarrhea.

Setting

The setting was a Veteran's Administration Medical Center located in the state of Florida. The Medical Center is a 327 bed tertiary care teaching hospital, with 300 additional authorized nursing home care beds and serves an estimated population of 435,442 veterans. The full range of inpatient and outpatient care is provided including Medicine (111 beds), Surgical (61 beds), Psychiatry (50 beds), Neurology (7 beds), as well as a 60-bed Spinal Cord Injury Service, and a 38-bed Comprehensive Rehabilitation Center. The center also provides a number of outpatient clinics on site and at satellite locations.

The Center is affiliated with a University College of Medicine and provides residency training programs in the following specialties: Internal Medicine, Orthopedics, Hematology, Pathology, Radiosurgery, Ophthalmology, Dermatology, Podiatry, Neurology, Neurosurgery, Urology, Nephrology, General Surgery, Otolaryngology, Psychiatry, Radiology.

Sample

The study sample consisted of 66 cases and 66 matched controls for a total of 132 subjects. Cases and controls were selected from the electronic medical record database of the study site medical center. Electronic medical records generated between January 1, 2000 and December 31, 2006 were utilized. Figure 2 illustrates the sequential steps used in selecting the cases, selecting potential controls, and matching controls to cases.

A total of 1,739 medical records were selected by data mining the medical records database. Criteria specified for the data mining were

(1) inpatient hospital admission between January 1, 2000 and December 31, 2006 and

(2) laboratory studies for *Clostridium difficile* cytotoxin assay, total lymphocyte count, serum albumin, and serum pre-albumin level documented during this time frame.

Of the 1,739 records, 330 were selected as potential cases from the original group because they also had a medical record diagnostic coding for *Clostridium difficile* associated diarrhea. Within this set, 155 records were identified which met the inclusion criteria of hospital acquired infection as defined by a diagnosis made ≥ 2 days after hospital admission and ≤ 31 days after hospital discharge, the cut-points selected for determination of a hospital-acquired CDAD.

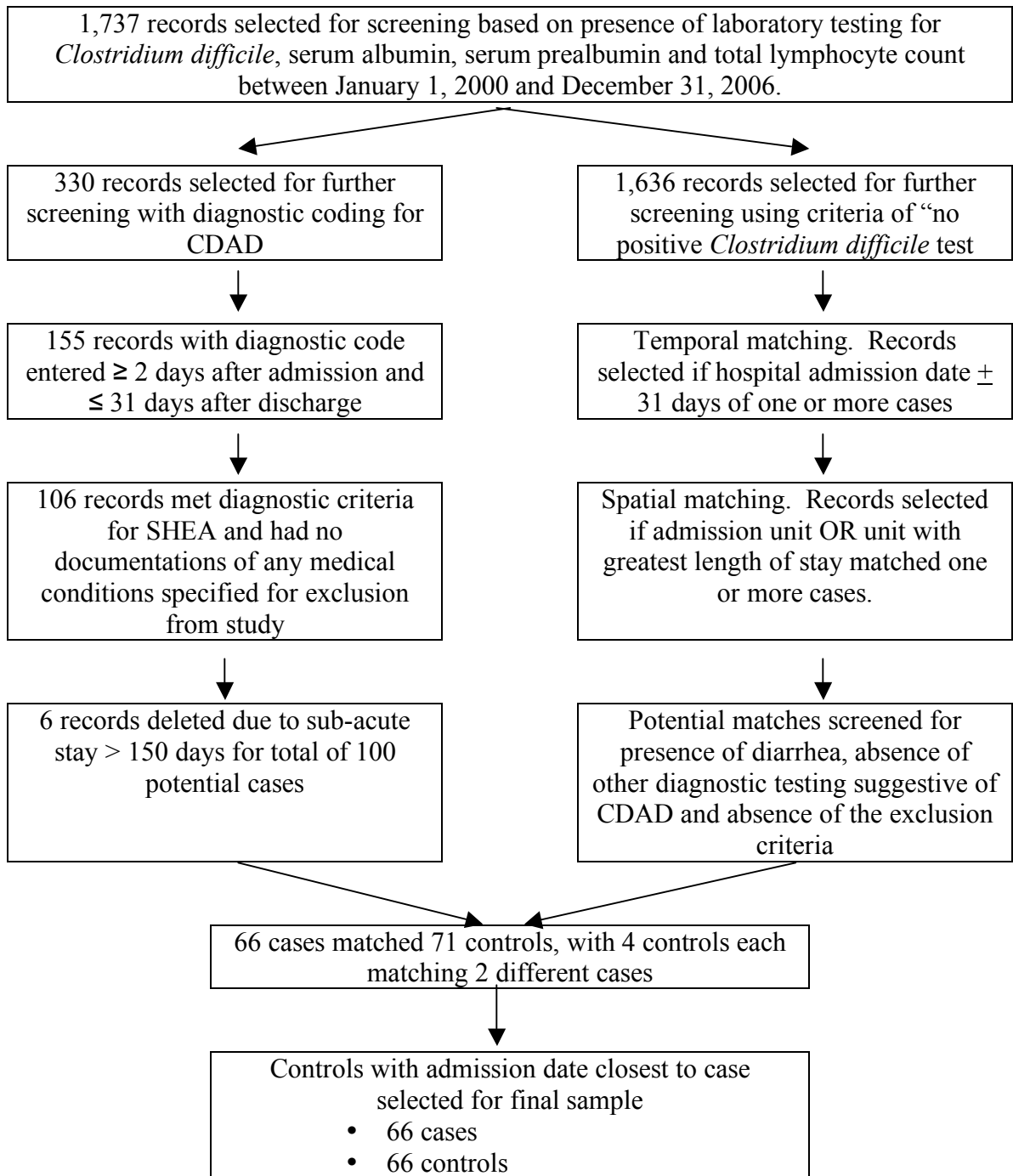


Figure 2. Sample selection process.

The set of 155 medical records was examined to determine if the record was consistent with criteria for CDAD case definition outlined in the Shea Position Paper

(Gerding, Johnson, Peterson, Mulligan, & Silva, 1995). These criteria were (1) diarrhea (2) documented concurrently or within 7 days of a positive result on one or more of the following diagnostic tests:

- Endoscopy with confirmation of pseudomembranous colitis
- Stool culture for toxigenic strain of *C. difficile*
- Cell culture cytotoxin test
- EIA toxin test

To reduce selection bias that might occur in persons tested for *C. difficile* due to recurrence of diarrhea following a previously diagnosed infection, cases were excluded if the database included a diagnosis of CDAD within 12 months preceding the positive test result. Because persons with chronic diarrhea referred for stool testing may experience colonization with *C. difficile* and might be easily misclassified, cases were also excluded if there was a history of any of the following: surgical removal of sufficient bowel to result in chronic diarrhea, chronic ulcerative colitis not previously subjected to surgical cure, Crohn's disease, diarrhea-predominant irritable bowel syndrome, or diabetic neuropathy with chronic diarrhea.

The screening process identified 106 medical records that qualified for inclusion in the study. Because the focus of the study was acquisition of CDAD during acute care hospitalization, records also were examined for length of stay in sub-acute areas of the institution. Six records were discarded due to sub-acute length of stay in excess of 150 days. This yielded a final set of 100 cases.

A total of 1,636 records in the Access database were screened for selection of controls. These 1,636 records were based on a query to identify records that never

recorded a positive *C. difficile* assay. Because ecological disease models consider both temporal and spatial distance to be a factor in the spread of communicable disease, controls were matched to cases by hospital admission date and geographic unit of hospital. For purposes of matching, when a record indicated that the subject was treated in more than one unit, the unit with the longest length of stay was used for spatial matching. To select the potential cases, records were first matched based on admission date \pm 31 days using an Access query. Records were then manually inspected to match potential controls to cases by admission unit or unit with the longest length of stay. Records that matched a case for both admission date and geographic unit of the hospital were reviewed to ensure presence of the following: (1) diarrhea documented within 7 days of the *Clostridium difficile* assay test, and (2) absence of both endoscopy and stool culture results indicative of CDAD. A total of 71 records were identified as matches. Within the 71 records, there were 4 duplicate matches. To enable a 1:1 matching, the control record that most closely matched the case record admission date was selected for the study.

Measurement of Variables

Age

Age of subjects was recorded as the chronological age in years, on the date of hospital admission.

Nutritional Status

Nutritional status was measured through use of three continuous variables: serum albumin, serum prealbumin, and lymphocyte count. Laboratory values were recorded

only for specimens collected within seven days of hospital admission. Laboratory reference ranges for these tests at the research site are identified in table 4.

Table 4
Laboratory Reference Ranges for Nutritional Indicators

Test	Low	High
Pre-Albumin (mg/100 ml)	18	45
Albumin (mg/100 ml)	3.8	4.8
Lymphocyte count ($10^9/L$)	1.1	3.4

Antibiotic Burden

Antibiotic burden was calculated for each subject by antibiotic agent, as a numerical sum of all Defined Daily Doses. The Defined Daily Dose is the assumed average maintenance dose per day for a drug when it is used for its main indication in adults with normal organ function. This measure was developed for the World Health Organization (WHO) Collaborative Centre for Drug Statistics Methodology to enable measurement of drug consumption independent of formulation or price. All DDD calculations were calculated using the WHO index on the world-wide web at <http://www.whocc.no/atcddd/>. For each antibiotic agent administered, data was recorded from the electronic record regarding the prescribed dose of the drug and the number of doses administered during the length of the exposure period (hospital admission to date of CDAD testing). These prescribed doses were then converted to DDD units, and a sum of the DDD units was computed for each drug. The drug burden for each subject was the sum total of all DDD's for all antibiotic agents.

Laxatives and Bowel Preparation Medications

Use of any cathartics administered orally or via a feeding tube for either (a) bowel cleansing regimens prior to surgical or endoscopic procedures or (b) prevention or treatment of constipation were recorded in a manner similar to antibiotics, and a DDD was calculated for each subject. Bowel preparations were excluded if there was documentation in the medical record indicating administration for a diagnostic procedure to identify pseudomembranous colitis in an individual believed infected with Cd.

Gastric Acid Suppression

Gastric acid suppression was measured for each subject by drug, dosage, dosage units, and number of dosages administered between the date of hospital admission and the date a stool sample was submitted for CDAD testing. This information was then converted to Defined Daily Doses (DDD), and a sum of the DDD units was computed. For comparison purposes, drugs classified as proton pump inhibitors and those classified as histamine-2 blocking agents were recorded separately and DDD units were calculated as separate categories.

Enteral Feeding

Enteral feeding was measured by the number of days in which a nasogastric, nasoenteral, or percutaneously inserted enteral feeding tube was used to provide nutritional support between the date of admission and the date a stool sample was submitted for CDAD testing.

Severity of Illness

Severity of illness level was assigned by the principal investigator using the Severity of Illness index (Horn & Horn, 1986). This index is an instrument designed to

create a generic measure, independent of clinician practice patterns, which quantifies a patient's illness severity based on retrospective review of records at the time of discharge. The index groups patients within a four-level ordinal scale, with one representing the least severe and four the most severe illness. The scale is applied to seven different dimensions: stage of principal diagnosis, complications, interactions, dependency, non-operative procedures, the rate of response to therapy, and the degree to which acute symptoms are resolved at the time of discharge and the overall severity score is the modal score for the seven categories. Data from 18 different hospitals found an overall weighted interrater reliability of 93.5%. The index has been found to explain 69-87% of the variability in resource use within Diagnostic Related Groups (DRGs) and when used to adjust for DRGs predicts 61% of the variability in cost per case. Kyne et al. (2002) reported a sensitivity of 87, specificity of 39, positive predictive value of 11 and negative predictive value of 97 for a Horn's index of 3 or more (severe or catastrophic illness) when used with a cohort of hospitalized adult patients receiving antibiotics. In a second cohort used to validate the model, the team reported sensitivity of 79, specificity of 73, positive predictive value of 27 and negative predictive value of 96.

Procedures

Approvals

Approval to conduct this study was obtained from the University of South Florida Institutional Review Board and the James A. Haley VAMC Human Subjects Research and Development Committee. Request was obtained from both boards to waive informed consent. To protect the privacy of all subjects, electronic medical records were de-identified prior to the start of data collection. A member of the medical staff of the

VAMC, who served on the dissertation committee, performed downloading of potential records into an Access database prior to accession by the principal investigator. Each record was then assigned a unique pointer number that was a computer-assigned sequential number, having no association with identifying data. A code-breaking log of subjects was retained by the VAMC employee within a locked office in a secured area of the medical center until data collection and analysis was completed. The log was then destroyed according to VAMC procedures.

Validation of Data Collection Procedure

At the start of data collection, a sample of ten electronic medical records was selected and data recorded using an Excel workbook developed by the principal investigator. To validate the accuracy of the principal investigator (PI) in use of the electronic medical record system, the VAMC co-investigator also reviewed the same records. The workbook of the PI and co-investigator were compared for concordance of data elements. All discrepancies between the data were clarified and reviewed until 100% concordance was obtained between the two sets of data. The PI then developed data collection reference guidelines to ensure consistency during the remainder of the data collection process.

Data Collection

All data were retrieved from the electronic medical record and recorded in an Excel workbook developed by the principal investigator based upon a paper worksheet previously approved by the dissertation committee. The worksheet listing all data elements collected is in Appendix A. The written data collection reference guide is found

in Appendix B. All medical record data was recorded under the unique identifier number developed in the initial data-mining operation.

Data Analysis

The Statistical Package for Social Sciences (SPSS), version 15 software was used for all data analysis. Initial inspection of data revealed missing values for the variables prealbumin and albumin. Because more than one-third of the subjects were missing prealbumin data, this variable was not utilized in the statistical analysis. Missing values for albumin were imputed by use of the mean for each group.

Cross tabulation revealed that 25% of the cells for the variable severity of illness included fewer than 5 subjects. To increase power of the analysis, the four categories of the severity of illness variable were collapsed into two: mild/moderate and severe/catastrophic. However, this process resulted in 31 uninformative pairs, and the decision was made to retain all four categories of severity of illness for the remainder of the data analysis.

Data were entered into SPSS as a sample of 66 pairs, with the cases coded as 1 for outcome and controls coded as 0. For each covariate, the difference between case and control values was computed, and the difference scores were used as the covariates in performing logistic regression analysis (Hosmer & Lemeshow, 2000). Conditional logistic regression was then performed using the Cox regression procedure, with data stratified by the matched pairs. Both univariate and multivariate logistic regression analysis were performed for all covariates using a forced entry method.

Chapter Four

Results

This chapter presents the findings from the study. Following description of the cases and controls, results of analytical testing, including the logistic regression modeling, is presented.

Sample

The data mining and screening process yielded a sample of 66 pairs matched for admission date and geographic location within the hospital. The sample was 98% male (64 cases and 65 controls), consistent with the veteran population from which the sample was selected. The age range for cases was 46.48 to 88.6 years, while control age range was 31.4 to 90.7 . The majority of the sample had exposure to antibiotic (83%) and proton pump inhibitor (65%) drugs, however there was limited exposure to tube feeding (17%) and histamine-2 blocking agents (14%) in this sample. Subjects were treated in a variety of patient care units within the institution, with the majority of the matched pairs treated in medical-surgical units, rather than critical care or rehabilitation oriented settings.

Subjects were classified in all categories of severity of illness, with the largest portion (45%) classified as having moderate illness and a third of the sample (33%) experiencing major illness. Although only 6 subjects (16%) experienced catastrophic illness, 4 of these 6 subjects were classified as cases.

Approximately half the sample had lymphocytopenia, present in a slightly higher percentage of cases (59%) than controls (45%). Hypoalbuminemia was present for 58% of both groups. Lymphocyte count and serum albumin values correlated significantly (Spearman's rho= .409, p<.01).

A comparison of means was computed for all continuous variables via paired t-tests. There was a statistically significant difference between means (p<.01) for all variables except histamine 2 burden and tube feeding days. Tube feeding days was significant at p<.05, and histamine 2 burden was non-significant. Table 5 identifies the variables, means and results of the paired testing for each of these variables.

Table 5
Comparison of Case and Control Means Scores on Continuous Predictors

Predictor	Cases (n=66)	Controls (n=66)	t	p
Length of exposure/risk period	17.65	24.77	-9.80	.000
Laxative/bowel prep burden	6.68	6.12	-3.43	.001
Proton Pump Inhibitor burden	8.81	15.56	-7.53	.000
Histamine 2 burden	2.02	1.29	-2.94	.769
Antibiotic burden	16.95	15.42	-6.70	.000
Tube feeding days	4.12	4.97	-2.17	.032
Serum albumin	3.72	3.58	-30.93	.000
Total lymphocyte count	1.24	1.12	4.25	.000
Age	68.47	71.27	-66.77	.000

Temporal and Spatial Clustering

Examination of the number of cases by unit by month revealed no patterns suggestive of an institution-wide outbreak during the time frame of the study. There

were 7 instances of temporal and spatial clustering of cases identified. Four of the 7 clusters occurred on the same medical nursing care unit. All of the clustering occurred during the months of March through August in different years. This temporal and spatial clustering underscores the importance of the macroenvironment in the spread of this disease.

Univariate Analysis

Table 6 illustrates results of the univariate logistic regression for host, microenvironmental and macroenvironmental characteristics for both cases and controls.

Table 6
Sample Description and Predictors of Hospital Acquired CDAD

Characteristic	Total (N=1 32)	Group		Univariate logistic regression			
		Controls (n=66)	Cases (n=66)	OR	95% CI		<i>p</i>
					Lower	Upper	
Host characteristics							
Age (years)							
M	69.87	71.27	68.47	1.013	.997	1.029	.124
SD	11.69	10.28	12.88				
Severity of illness							
% minor illness	16.67	25.76	7.58	1.845	1.322	2.574	.000
% moderate illness	45.45	54.55	36.36				
% major illness	33.33	16.67	50.00				
% catastrophic illness	4.54	3.03	6.06				
Serum albumin (mg/dl)							
M	3.65	3.65	3.65	0.863	.752	.989	.034
SD	0.73	0.72	0.74				
% hypoalbuminemic	57.58	57.58	57.58				
Lymphocytes							
M	1.18	1.12	1.24	0.915	.735	1.139	.424
SD	0.74	0.77	0.72				
% lymphocytopenic	52.27	45.45	59.09				
Macroenvironmental characteristic							
Length of exposure (days)							
M	21.21	24.77	17.65	1.008	.999	1.018	.085
SD	23.03	26.21	18.87				

Table 6 (Continued)

Characteristic	Total (N=1 32)	Group		Univariate logistic regression			
		Controls (n=66)	Cases (n=66)	OR	95% CI		<i>p</i>
					Lower	Upper	
Microenvironmental exposure characteristics							
Laxatives/preps							
M	6.40	6.12	6.68	.999	.989	1.009	.851
SD	16.44	15.98	17.01				
% exposed (≥ 1 DDD)	31.82	31.82	31.82				
Proton pump inhibitors							
M	12.18	15.56	8.81	1.017	1.002	1.032	.023
SD	18.20	21.72	13.13				
% exposed (≥ 1 DDD)	65.15	65.15	65.15				
Histamine 2 blockers							
M	1.65	1.29	2.02	.990	.961	1.020	.516
SD	5.98	4.22	7.35				
% exposed (≥ 1 DDD)	14.39	15.15	13.64				
Antibiotic burden							
M	16.18	15.418	16.95	1.002	.994	1.010	.684
SD	25.14	24.642	25.804				
% exposed (≥ 1 DDD)	78.03	72.73	83.33				
Tube feeding (days)							
M	4.55	4.97	4.12	1.002	.991	1.012	.768
SD	16.10	17.37	14.85				
% exposed (≥ 1 day)	17.42	16.67	15.15				

This analysis identified age, severity of illness, serum albumin level, length of exposure, and proton pump inhibitor drug burden as statistically significant predictors of case status ($p < .25$).

Multivariate Analysis and Model Evaluation

Conditional logistic regression analysis via a Cox regression procedure was used to identify predictor variables significantly associated with classification of CDAD case status. Following an initial modeling process, only three variables remained as significant predictors of case status: severity of illness, antibiotic exposure, and length of exposure/risk period. Table 7 presents the results of the conditional logistic regression

modeling. Increased severity of illness, a decrease in the number of Defined Daily Doses of antibiotic, and an increase in the length of hospitalization prior to symptom development were all significantly associated with CDAD case status. The overall model was significant at $p=.000$.

The calculated Hosmer and Lemeshow estimation of R^2 was 25.274, indicating that the model explains approximately 25% of the variance in CDAD outcome. It should be noted, however, that “low R^2 values in logistic are the norm” (Hosmer & Lemeshow, 2000, p.167).

Table 7
Conditional Logistic Regression of Variables Predictive of CDAD Case Status

Variable	B	Wald	p	OR	95% CI
Severity of illness	.739	13.506	.000	2.094	1.412, 3.106
Antibiotic exposure	-.021	6.007	.014	.979	.962, .996
Length of exposure	.022	5.933	.015	1.022	1.004, 1.040

Note: $R^2 = 25.274$ (Hosmer & Lemeshow). Model $\chi^2 = 23.123$, $p=.000$. OR= Odds Ratio, CI=Confidence Interval.

As covariates were removed from the model, the Odds Ratios (OR) for all other covariates remaining in the model were examined to determine if there was a large magnitude in the change of the OR which might indicate confounding. Only one clinically significant change was noted. Both albumin and lymphocyte count appeared to serve as slight confounders for the measure of severity of illness, although the magnitude of the change in the severity coefficient was small for each variable (5% for serum albumin, 3% for lymphocyte count).

Following the identification of the initial model, a number of interactions were tested via forced entry. Potential interactions were identified for numerous combinations that might be clinically important related to age, serum albumin, antibiotic drug burden, and the length of exposure/risk period. Table 8 lists the interactions tested and the outcome of each test. None of these interactions was found statistically significant.

Table 8
Conditional Logistic Regression of Interaction Effects as Predictors of Case Status

Interaction	B	Wald	p	OR	95% CI
Age x Antibiotics	-.021	.076	.783	.980	.846, 1.135
Age x Albumin	.156	.018	.893	1.169	.120, 11.414
Age x Lymphocytes	.116	.008	.927	1.123	.094, 13.432
Age x Laxatives	.000	.034	.853	1.00	1.00, 1.00
Age x Proton pump inhibitor	.031	.076	.783	1.031	.8289, 1.283
Age x H2 blocker					
Age x Length of exposure	-.015	.038	.846	.985	.844, 1.149
Albumin x Lymphocyte	2.520	.018	.893	12.431	.000, 1E+017
Albumin x Proton pump inhibitor	.426	.376	.540	1.532	.392, 5.984
Albumin x Length of exposure	-.194	.076	.783	.824	.207, 3.275
Antibiotics x Albumin	-.425	.250	.617	.654	.123, 3.460
Antibiotics x H2 blockers	-.076	.403	.526	.926	.732, 1.173
Length of exposure x Lymphocytes	-.195	.038	.846	.823	.116, 5.859

DFBeta residuals were examined to identify data points that may have been unduly influential for a particular covariate or which did not fit the model well. All of these residuals were very small (≤ 0.06).

Collinearity was assessed via generation of a matrix of correlations, using the non-parametric Spearman rho. Statistically significant correlations were all of low magnitude ($<.50$) except for the correlations between length of exposure with antibiotic burden (.541, $p=.000$) and tube feeding days with antibiotic burden (.513, $p=.000$). Collinearity diagnostics in the linear regression function revealed no tolerance values < 0.1 and no VIF values > 10 , indicating that there were no serious problems with collinearity caused by these correlations.

Receiver Operator Characteristic (ROC) curves were generated for each of the predictors in the model. Figure 3 illustrates the plot of sensitivity versus 1-specificity for the Severity of Illness index for all possible cut points. The area under the curve is .776, indicating an acceptable level of discrimination. The area under the curve for length of exposure was less acceptable at .620. The plot for antibiotic burden was nearly non-discriminatory with an area under the curve of .537.

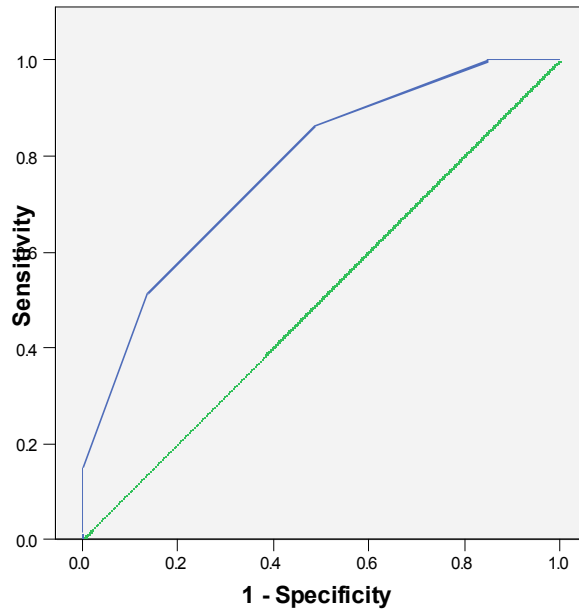


Figure 3. ROC curve for Severity of Illness index. Area under the curve = .776.

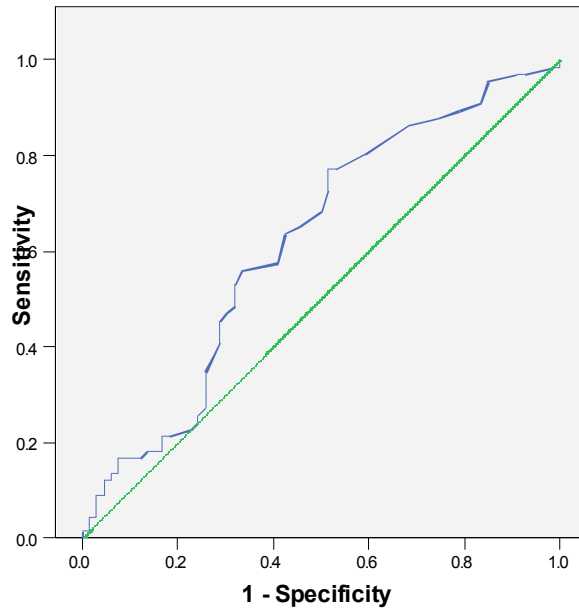


Figure 4. ROC curve for length of exposure. Area under curve = .620.

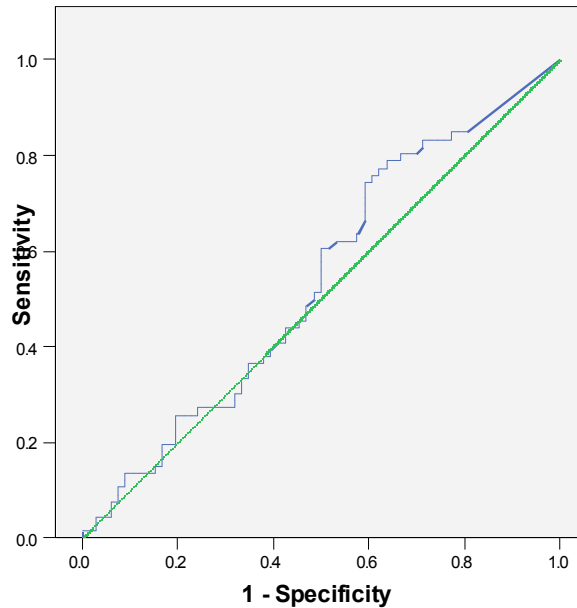


Figure 5. ROC curve for antibiotic burden. Area under curve = .537.

Hypothesis

The study originally hypothesized that severity of illness, length of exposure and malnutrition are significant predictors of CDAD cases status when cases and controls are matched for admission date, and geographic unit of hospital, and after controlling for the effects of age, tube feeding, cathartic administration, antibiotic therapy, and acid suppression therapy. This sample suggests that both severity of illness and length of exposure are significant predictors, but that malnutrition estimated by two laboratory indicators, serum albumin and lymphocyte count, is not an accurate predictor. It was anticipated that antibiotic exposure might represent an effect modifier, rather than a predictor. However, antibiotic exposure, quantified by cumulative Defined Daily Doses was identified as a significant independent predictor of CDAD case status and failed to demonstrate any interaction effects when tested in combination with a number of biologically plausible covariates.

Chapter Five Discussion

The purpose of this study was to test an ecological model for its usefulness in predicting the development of *Clostridium difficile*-associated diarrhea in hospitalized adults. This chapter discusses the findings of the study, conclusions about the findings, limitations of the study, and implications for nursing practice and education, as well as future nursing research.

Based on an ecological model, it was hypothesized that severity of illness, length of exposure and malnutrition would significantly predict the development of CDAD case status when cases and controls are matched for admission date, and geographic unit of hospital, and after controlling for the effects of age, tube feeding, cathartic administration, antibiotic therapy, and acid suppression therapy.

Severity of Illness

Although the portion of the sample rated as catastrophic on the Severity of Illness Index was less than 20%, the portion of cases rated as catastrophic was double that of controls. An even greater disparity was observed in the category of major illness, with 50% of cases and only 16.67% of controls exhibiting characteristics of major illness. More than 80% of controls fell into either moderate or minor severity of illness categories, but only less than 45% of cases were similarly ranked.

Severity of illness was identified as a statistically significant independent predictor of CDAD case status for this sample following conditional logistic regression analysis.

This finding is consistent with previous studies using the Horn Severity of Illness index (McFarland, 1995; Kyne, 2002; Vesta, 2005). Likewise, it is consistent with the work of Modena (2005) who characterized severity in relation to events such as mechanical ventilation and ICU utilization, criteria that are part of the Horn Severity index.

Length of Exposure

Length of exposure was defined as the number of days which elapsed from the date of hospital admission until the submission of a stool sample for *Clostridium difficile* laboratory testing. This variable was derived for this study from the ecological model specifying extrinsic ecologic factors affecting infectious disease exposure. Based on the model, both temporal and spatial distance must be increased to prevent the transmission of microbial contaminants to susceptible hosts. Previous researchers have considered length of total hospital stay as a variable, but this was studied primarily as a proxy for severity of illness. The author is unaware of previous studies considering temporal distance as a risk factor for acquisition of this disease.

In this study, there was a statistically significant difference between the mean length of exposure for cases and controls (17.65 versus 24.77 days). Length of exposure was also identified as a statistically significant independent predictor of CDAD case status following multivariate modeling. Specifically, the longer a subject had been hospitalized prior to the development of diarrhea and subsequent stool sample collection, the more likely that the diarrhea was caused by *Clostridium difficile*.

Malnutrition

This study attempted to operationalize malnutrition by means of three commonly available laboratory tests: serum albumin, lymphocyte count, and serum prealbumin.

Since immunocompetence is partially dependent upon positive nitrogen balance and intake of essential vitamins and minerals, it was hypothesized that malnutrition, evidenced by values below the laboratory's reference values, would be associated with CDAD status. Although the original data mining process used prealbumin test results as one of the criteria for selection of a record for screening, only a small number of the records included prealbumin test results during the same hospitalization as the diarrhea episode. Therefore, this variable could not be included in the analysis.

Between cases and controls, identical proportions of subjects demonstrated hypoalbuminemia, while a greater proportion of cases than controls exhibited lymphocytopenia based on the laboratory's reference values for each test. However, in this sample, cases demonstrated significantly higher mean albumin and lymphocyte values than cases..

Lymphocyte count was not significantly associated with CDAD status in either univariate or multivariate analysis. Serum albumin was associated on univariate analysis with CDAD case status, but failed to demonstrate a significant association during multivariate analysis. The univariate association is consistent with other reported univariate associations between serum albumin and CDAD (Al-Tureihi et al., 2005, Andrews et al., 2003, Raveh, Rabinowitz, Breuer, Rudensky, & Yinnon, 2006; Rubin et al., 1995). Recently, a significant multivariate association was reported between hypoalbuminemia and CDAD (Peled et al., 2007). In that study, a significant difference existed between mean albumin scores of cases and controls, and subjects were categorized for the logistic regression analysis based on a selected cut-point to indicate hypoalbuminemia. The data group in this present study also exhibited a statistically

significant difference in mean albumin scores between cases and controls on univariate testing with a paired t-test. It should be noted that in this sample, identical proportions of cases and controls had serum albumin levels lower than the laboratory reference low value of 3.8 mg/100 ml. Only 26% of the entire sample had albumin levels lower than a value of 3.4 mg/100 ml.

Age

This sample demonstrated a statistically significant difference between the mean age of cases and controls, but failed to produce an independent association between age and CDAD status on multivariate analysis. This finding is in contrast to the substantial support in the research literature for advancing age as a risk factor for CDAD (Bignardi, 1998). The sample in this study was relatively heterogeneous in terms of age, and this represents a departure from samples in other studies. This broader range of subject ages may be a reflection of the specific Veteran's Administration Medical Center population from which the sample was drawn.

Microenvironmental Exposure Variables

This study included a number of exposures previously identified in the literature as affecting the bacterial ecology of the intestinal tract. These exposures included laxative or cathartic drugs, acid suppression drugs, antibiotics, and tube feeding. It was hypothesized that these variables would not be significant predictors of CDAD status in a study design that carefully considered criteria for case and control selection and matched for both temporal and spatial exposure data.

For purposes of this study, an attempt was made to quantify the dose response phenomenon for microenvironmental exposure through use of standardized measures.

For drug exposure, the World Health Organization's Defined Daily Doses were computed for each class of drug and summed to produce a continuous measure of exposure. For tube feeding, the measure was the number of days of tube feeding prior to the onset of diarrhea and stool testing. Although a number of studies have considered these same variables, exposure has often been measured strictly as a dichotomous variable.

Tube Feeding

This data failed to demonstrate tube feeding exposure as a significant predictor of CDAD. Although nearly all of the subjects tube fed during the period experienced enteral nutrition for more than 2 weeks, there were simply not enough subjects in the sample among either cases or controls who received this therapy. Therefore, there was not sufficient power to detect a difference between the two groups.

Cathartic Burden

When measured as drug burden based on the total number of Defined Daily Doses ingested during the exposure period, cathartic drugs failed to demonstrate a significant exposure risk. Although there was a statistically significant difference between mean cathartic drug burdens of the two groups, both univariate and multivariate logistic regression revealed non-significant O.R.'s for this variable.

Acid Suppression Burden

Acid suppression burden considered the DDD's for both proton pump inhibitor drugs and histamine 2 blocking agents. Neither category of drug was found to be a statistically significant predictor of CDAD when measured this way. In this sample identical or nearly identical portions of both cases and controls had exposure to acid suppression therapy, so that even if this variable had been measured as binary data,

similar to some previous studies, it is unlikely that it would have proven to be a predictor. Although two prospective case-studies have recently reported significant multivariate associations for acid suppression, both measured exposure based on subject's reporting of prescribed drugs prior to hospitalization (Peled et al., 2007; Yearsley et al., 2006) and thus were subject to recall bias. Likewise, neither study focused on hospital administered acid suppressive drugs, the most meaningful information available to the bedside clinician.

Antibiotic Burden

This study demonstrated that an increase in antibiotic drug burden actually provided a slight decrease in risk of the development of hospital-acquired CDAD. This finding contrasts sharply with other studies, which measure exposure in other terms such as the number of drugs, number of prescribed doses, number of days of therapy. In previous research, a dose-response relationship has been demonstrated for antibiotic exposure. The decreased dose-response in this study may have occurred for several reasons. The combined DDD's of all categories of antibiotics were considered together as a group, regardless of the therapeutic or chemical classification of the antibiotic, which may account for some of the differences. Analysis of data by specific drug categories may have resulted in some demonstration of increased risk. Case-control studies, which examine specific drugs or drug categories are subject to considerable selection bias, particularly in prospective studies in which the drug exposure of interest is used as a criterion for study participation. Drug prescribing patterns vary geographically and between different health systems, and the antibiotic formulary available to clinicians varies between institutions, making cross comparisons difficult. It also is possible that

antibiotic exposure in some studies may actually represent the confounding factor of severity of illness. The more severely ill individual remains hospitalized longer, thus is exposed to more drug therapy overall, and more environmental stressors.

Limitations of the Study

A serious limitation of this study was the small sample size obtained for the number of variables screened. A lack of statistical power may have influenced the failure to obtain statistical significance for some predictors. Although the original data pool was very large, the screening criteria and matching criteria were very strict to decrease bias identified in earlier research. As a result of this screening and matching process, only 66 pairs could be identified from the initial 1, 737 records.

The homogeneity of gender for this sample is also a limitation. The predominance of male subjects is not representative of the general acute care hospital population. The data collection process made no attempt to determine ethnicity of the subjects. Although previous research has not identified any ethnic differences in the development of CDAD, it is possible that differences in response to exposures, particularly drug exposures may vary based on genomic differences.

The large number of missing values for the prealbumin variable necessitated removal of this variable from any type of statistical modeling. Elimination of this variable, combined with the small number of tube fed subjects, makes it extremely difficult to determine the impact of nutritional status on development of CDAD among this sample. Although lymphocyte count provides some information about nutritional status, serum albumin is influenced by many biologic factors in addition to nutrition and cannot be considered an accurate measure of nutritional status.

This study used a retrospective case-control design. Although an attempt was made to establish a temporal relationship between exposure and disease by excluding subjects hospitalized less than 2 days or discharged more than 31 days before the onset of diarrhea symptoms, it is still possible that actual exposure to *Clostridium difficile* microorganisms occurred prior to or after hospitalization. The design was also vulnerable to selection bias, since cases were initially identified via data mining medical records to identify those with available data points for four specific laboratory tests, one being the toxin assay for the disease. Therefore, the clinicians caring for the subjects at the time diarrhea symptoms were identified, introduced a form of diagnostic bias into the study, which was beyond the control of the researcher. If a clinician believed that CDAD was a possible differential diagnosis, a specimen was likely submitted. There may have been additional potential subjects with diarrhea symptoms who would have tested either positive or negative for CDAD cytotoxin.

Generalizability

Given the limitations of the sample and the retrospective case-control design, caution should be used in any attempts to generalize these findings to other acute care hospital settings. The lack of female subjects and the constricted age range indicate these findings may not be applicable to other settings. The study site is located in a major metropolitan area, with numerous other comprehensive hospitals available. Given the mean age of study participants, Medicare insurance coverage would qualify many of the subjects for treatment at any of these other institutions. It is impossible to know why an individual might be hospitalized at a particular institution. Therefore, it is possible that

other host-related variables exist inherently within the veteran's population, which were not measured in this study.

Implications for Nursing Education and Practice

This study presents some important implications for nursing education and clinical practice. The framework used to organize the study suggests that if clinicians can identify intrinsic host characteristics that make them more susceptible to disease, the clinician can alter the external environmental conditions to increase spatial and temporal distance between the vulnerable individual and the harmful environmental exposure. This study identified increased length of exposure, increased severity of illness, and a decrease in antibiotic drug burden as independent predictors of CDAD development among hospitalized adults. As more virulent strains of *Clostridium difficile* continue to emerge, clinicians must develop new strategies for increasing temporal and spatial distance beyond the currently employed system of universal precautions combined with isolation of infected persons. Some means of "isolating" will need to occur for the most severely ill persons.

This is particularly evident in the critical care environment, where most patients have extensive organ involvement, require multiple means of life support, and have complex interrelated pathophysiologic derangements. Unfortunately, this is also the same environment in which traditional spatial distance is limited by design of the unit and temporal distance is rarely possible due to the high demand for critical care bed placement in modern hospitals. Therefore, new strategies may be necessary to achieve spatial distance such as strict stool containment and disposal, limitation of caregivers, and assignment of infected and non-infected patients to totally different care teams.

Nurse educators working in both pre-licensure and continuing education settings face a number of challenges. Through contamination of hands, equipment, and uniforms, nurses can serve as a vector for transmission of *Clostridium difficile* spores. Continuing to teach students and practitioners prevention of infectious disease through traditional mechanisms of identification and isolation does not fully enable them to appreciate the importance of identifying the vulnerable, at-risk population to protect them from exposure. Nurses working in practice settings with clients at high risk for infection, such as neonatal and bone marrow transplant units, have demonstrated that limitation of visitation, restriction of access to the care unit, and adherence to use of handwashing and barrier products can reduce the incidence of infections among high-risk persons. Educators teaching about infectious disease need to include content about identification of at-risk clients and increasing spatial and temporal distance between at-risk individuals and infected individuals.

Nurse administrators may also need to consider changes in work assignment, visitation policies, and the spatial assignment of individual patients. This will be problematic given a global nursing shortage. However, nosocomial infections may prove to be an economic burden that threatens the continued viability of institutions, particularly in an era when third party payers are choosing to withhold compensation for problems that develop during an individual patient's stay within an acute care hospital.

Recommendations for Future Research

Nurses must become increasingly involved in research concerning the epidemiology and prevention of all types of hospital acquired infections to ensure that predictors and prevention measures are well grounded and clinically useful.

Future research regarding CDAD should include measures to determine if other commonly used measures of disease severity or patient acuity might be as useful in predicting risk as the Horn Severity of Illness index, particularly tools that are already widely used by nurses such as the APACHE II scoring system.

More research is needed to understand the relationship between drug exposure and CDAD. A prospective, multi-center case-control study would enable accumulation of sufficient data points to determine multivariate associations. There is a definite need for a standardized measure of drug exposure. Although somewhat tedious, this study demonstrates that the DDD system is one way in which standardized measures of drug exposure could be compared across studies.

Two recent publications have identified colonization pressure, the sum of daily point prevalence rates of disease for each day spent in the environment of exposure, as significant predictors of CDAD development (Dubberke et al., 2007; Lawrence et al., 2007). Colonization pressure is a quantifiable way to measure the variable of environmental stressors identified in the conceptual framework for this study. Intervention studies to determine strategies for disease prevention could use this concept as an outcome variable. As a continuous variable, this would offer benefits in study design and data analysis over traditional binary outcomes.

Conclusions

This study confirmed severity of illness and length of exposure as independent predictors of the acquisition of hospital-acquired *Clostridium difficile*-associated diarrhea. It did not confirm malnutrition as a risk factor as hypothesized. An increase in the number of Defined Daily Doses of antibiotics was found to indicate a slight decrease

in disease risk, a departure from previous research measuring drug exposure in other ways.

The study supports the use of an ecologic model to help explain the phenomenon of hospital-acquired CDAD and indicates a framework for future research regarding intrinsic host characteristics suggesting increased risk, and intervention strategies to decrease environmental stressors on the vulnerable host.

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Appendix A: Data Collection Worksheet

Data Collection Worksheet

Exclusion Criteria

NONE of the following located in medical record:

- Diagnosis of CDAD within 12 months preceding hospitalization
- History of surgically created chronic diarrhea
- History of Crohn's disease or untreated chronic ulcerative colitis
- History of diarrhea-predominant irritable bowel syndrome
- Diabetic neuropathy with chronic diarrhea

Inclusion Criteria

<input type="checkbox"/> Case	<input type="checkbox"/> Diarrhea Control
Diarrhea:	Diarrhea:
<input type="checkbox"/> >2 stools in 24 hours	<input type="checkbox"/> >2 stools in 24 hours
<input type="checkbox"/> loose and unformed or liquid consistency	<input type="checkbox"/> loose and unformed or liquid consistency
<input type="checkbox"/> concurrent or within 7 days of positive finding(s) for:	<input type="checkbox"/> concurrent or within 7 days of negative finding(s) for:
<input type="checkbox"/> Stool culture for toxin-producing strain of <i>Cd</i> Date:	<input type="checkbox"/> Stool culture for toxin-producing strain of <i>Cd</i> Date:
<input type="checkbox"/> Cell culture cytotoxin assay Date:	<input type="checkbox"/> Cell culture cytotoxin assay Date:
<input type="checkbox"/> EIA toxin test Date:	<input type="checkbox"/> EIA toxin test Date:
<input type="checkbox"/> Endoscopy with pseudomembranous colitis Date:	<input type="checkbox"/> Endoscopy with pseudomembranous colitis Date:

Demographic Data

Age on date of admission: _____

Gender: Male Female

Matching Information

	Date	Unit		Date	Unit		Date	Unit
Admit			Transfer			Transfer		
Transfer			Transfer			Transfer		
Transfer			Transfer			Discharge		

Severity of Illness Index

Characteristic	LEVELS				
	1	2	3	4	
Stage of Principal Diagnosis	Asymptomatic	Moderate Manifestations	Major Manifestations	Catastrophic Manifestations	
Complications	None or very minor	Moderate – less important than principal diagnosis	Major – as or more important than principal Dx	Catastrophic – death or major permanent disability	
Interactions	None or minor	Moderate	Major	Catastrophic	
Dependency	Low	Moderate	Major	Extreme	
Procedures (Non-O.R.)	Noninvasive Diagnostic or Minor Therapeutic	Therapeutic or Invasive Diagnostic	Nonemergency Life Support	Emergency Life Support	
Response to therapy	Rate	Prompt	Moderate Delay	Serious Delay	No Response
	Resolution of Acute Symptoms	Complete	Extensive but incomplete	Incomplete and Disabling	No Resolution

Bowel preparation regimen

<input type="checkbox"/> No laxatives or bowel preparation documented	
<input type="checkbox"/> Enema <input type="checkbox"/> Colostomy irrigation <input type="checkbox"/> Cathartic: <hr/> <input type="checkbox"/> Cathartic: <hr/>	<input type="checkbox"/> NO EVIDENCE that prep being administered for colonoscopy to diagnose Pseudomembranous colitis

Tube feeding

<input type="checkbox"/> No tube feeding documented	
<i>Type of tube</i>	<i>Number of days used</i>
<input type="checkbox"/> percutaneously inserted tube <input type="checkbox"/> gastric <input type="checkbox"/> enteral	
<input type="checkbox"/> nasoenteral tube	
<input type="checkbox"/> nasogastric or orogastric tube	

Nutritional indices within first 7 days of hospitalization

Laboratory indicator	Lab normal	Value	Date	Hospital Day #
<i>Serum albumin</i>				
<i>Pre-albumin</i>				
<i>Lymphocyte count</i>				

Gastric acid suppression

A Drug name	B Category	C Dosage	D DDD	E DDD Units	F # Doses	G (E) x (F) Burden
	<input type="checkbox"/> PPI <input type="checkbox"/> H2 blocker					
	<input type="checkbox"/> PPI <input type="checkbox"/> H2 blocker					
	<input type="checkbox"/> PPI <input type="checkbox"/> H2 blocker					
	<input type="checkbox"/> PPI <input type="checkbox"/> H2 blocker					

Appendix B: Record Review Protocol

Electronic Medical Record Review Protocol

Data Collection Step	Location of Information
1. Verify that potential subject does not meet exclusion criteria	History and physical Admitting diagnosis
2. Verify that potential subject meets criteria for inclusion as either case or control	Bowel record Laboratory data Endoscopy data
3. Record age	Patient information
4. Record gender	Patient information
5. Record dates of admission and discharge	Patient information
6. Record dates of transfer within the facility by room number	Medication Record
7. Complete Severity of Illness Index:	
a. Stage of principal diagnosis at admission = peak extent of organ involvement as manifested by the patient at admission	History and physical examination findings
b. Complications = complications of the principal diagnosis, or complications that are a direct result of the therapy, hospitalization, or accidents that arise during hospitalization	Discharge summary
c. Interactions = conditions or problems, other than the principal diagnosis and complications, that have no	Discharge summary
d. Physiologic relationship to the principal diagnosis or that contributed to or caused the illness represented by the principal diagnosis	History and physical Discharge summary
e. Dependency = degree to which the patient requires more than the minimal level of direct care for the principal diagnosis. It includes dependency that is a consequence of the principal	Nursing acuity rating Nursing care records

Data Collection Step	Location of Information
<p>diagnosis, complications, and active interactions. The rating should be representative of the patient's "most of the time" dependency during the entire hospitalization and the degree to which "most-of-the-time" level of care is greater than the minimum expected for that principal diagnosis.</p>	
<p>f. Non-operating room procedures = peak procedural interventions undertaken EXCLUDING surgical procedures performed in an operating room. The TYPE of procedure, not the number of procedures performed, determines the level of this dimension</p>	<p>Cardiac arrest record Mechanical ventilation Laboratory Diagnostic imaging</p>
<p>g. Rate of response to therapy = response to the therapies instituted for the principal diagnosis and the impact of complications and active interactions on the hospitalization</p>	<p>Discharge summary</p>
<p>h. Remission of acute symptoms = extent to which a patient still shows evidence of the acute injury or illness related to the principal diagnosis, complications, or active interactions at the time of discharge, excluding preexisting conditions that did not change during the hospitalization and for which no change was expected</p>	<p>Discharge summary</p>
<p>8. Determine if bowel preparation was administered during hospital stay, and if so, record the type of preparation administered.</p>	<p>Medication administration record</p>
<p>9. Determine if tube feeding was used during hospitalization, and if so, record the number of days administered by the appropriate type of feeding tube</p>	<p>Diet record</p>
<p>10. Record the laboratory value for the <i>first</i> test completed during the hospitalization. Record the date of specimen collection. To determine the hospital day number, count the date of admission as day 1.</p>	<p>Laboratory record</p>
<p>11. Determine gastric acid suppression exposure:</p>	

Data Collection Step	Location of Information
a. Record drug name in column A	Medication Record
b. Check box for correct drug category in column B.	
c. Record the drug dosage in Column C	Medication Record
d. Record the Defined Daily Dose (DDD) in Column D	
e. Divide Column C by Column D to determine the number of DDD units and record value with two decimal places in Column E	Pocket calculator
f. Record the number of doses received prior to diagnostic testing for <i>Clostridium difficile</i> in Column F.	Medication Record
g. Multiply the values in Columns E and F with two decimal places to determine the drug burden and record in Column G.	Pocket calculator
h. Add the values of Column G and record the total Acid Suppressant burden	Pocket calculator
12. Determine the antibiotic exposure	
a. Record drug name in column A	Medication Record
b. Check box for correct drug category in column B.	Antibiotic Guide
c. Record the drug dosage in Column C	Medication Record
d. Record the Defined Daily Dose (DDD) in Column D	Antibiotic Guide
e. Divide Column C by Column D to determine the number of DDD units and record value with two decimal places in Column E	Pocket calculator
f. Record the number of doses received prior to diagnostic testing for <i>Clostridium difficile</i> in Column F. number of doses received during hospital days 1-30 only.	Medication Record

Data Collection Step	Location of Information
g. Multiply the values in Columns E and F with two decimal places to determine the drug burden and record in Column G.	Pocket calculator
h. Add the values of Column G and record the total Antibiotic burden	

Appendix C: Institutional Review Board Approval



UNIVERSITY OF
SOUTH FLORIDA

June 6, 2007

Susan E. Steele, M.S.
9780 Deepstep Road
Sanderville, GA 31082

Attn: Susan McMillan, Ph.D.

RE: **Expedited Approval** for Initial Review including your Application for Waiver/Alteration of HIPAA Authorization
IRB#: 105808d
Title: *Development of an Ecological Model to Predict Risk for Acquisition of Clostridium Difficile-Associated Diarrhea During Acute Care Hospitalization*
Study Approval Period: 05/29/07 to 05/27/08

Dear Ms. Steele:

On 05/29/07, Institutional Review Board (IRB) reviewed and **APPROVED** the above protocol **for the period indicated above**. It was the determination of the IRB that your study qualified for expedited review based on the federal expedited category number 5. This approval includes a Waiver of Informed Consent.

The IRB also reviewed and **APPROVED** your Application for Waiver/Alteration of HIPAA Authorization for the above protocol as outlined below:

Your HIPAA Waiver/Alteration application has been approved for you to conduct a retrospective medical chart review involving those patients who were admitted to the James A. Haley Veterans Hospital between January 1, 2000 and December 31, 2006 and who had one or more tests recorded for Clostridium difficile stool toxin, as stated in your application.

Please note: approval of your waiver/alteration of HIPAA authorization is based upon your having satisfied the following HIPAA Privacy Rule mandates:

1. The use or disclosure of protected health information (PHI) involves no more than minimal risk to the privacy of the individual, based on the presence of: a) an adequate plan to protect the identifiers from improper use and disclosure; b) an adequate plan to destroy the identifiers at the earliest opportunity consistent with the conduct of the research unless there is health or research justification for retaining the identifiers or such retention is otherwise required by law; and c) adequate written assurance that the PHI will not be reused or disclosed to any other person or entity, EXCEPT 1) as required by law, 2) for authorized oversight of the research study, or 3) for other research for which the use or disclosure of PHI would be permitted by the HIPAA Privacy Regulations.

OFFICE OF RESEARCH • DIVISION OF RESEARCH INTEGRITY & COMPLIANCE
INSTITUTIONAL REVIEW BOARDS, FWA NO. 00001669
University of South Florida • 12901 Bruce B. Downs Blvd., MDC035 • Tampa, FL 33612-4799
(813) 974-5638 • FAX (813) 974-5618

2. The research could not practicably be conducted without the waiver or alteration; and
3. The research could not practicably be conducted without access to and use of the PHI.

Please note, if applicable, the **enclosed informed consent/assent documents are valid during the period indicated by the official, IRB-Approval stamp located on page one of the form.** Valid consent must be documented on a copy of the most recently IRB-approved consent form. Make copies from the enclosed original.

Please reference the above IRB protocol number in all correspondence regarding this protocol with the IRB or the Division of Research Integrity and Compliance. In addition, we have enclosed an Institutional Review Board (IRB) Quick Reference Guide providing guidelines and resources to assist you in meeting your responsibilities in the conduction of human subjects research. Please read this guide carefully. It is your responsibility to conduct this study in accordance with IRB policies and procedures and as approved by the IRB.

We appreciate your dedication to the ethical conduct of human subject research at the University of South Florida and your continued commitment to the Human Research Protections Program. If you have any questions regarding this matter, please call 813-974-9343, or, for HIPAA-specific questions, please contact Vinita Witanachchi J.D., Research Privacy Officer, at 974-5478.

Sincerely,



Barry B. Bercu, M.D., Chairperson
USF Institutional Review Board

Enclosures: (If applicable) IRB-Approved, Stamped Informed Consent/Assent Documents(s)
IRB Quick Reference Guide

Cc: Sandra Partap, USF IRB Professional Staff
Vinita Witanachchi, J.D., Research Privacy Officer
JAH-VA
FAO

Appendix D: James A. Haley VAMC Research Committee Approval

APPROVAL - Initial Review (Minimal Risk)

Date: June 27, 2007
From: John A. Schinka, Ph.D., Chairperson *John A. Schinka Ph.D.*
Investigator: Philip R. Foulis, M.D., M.P.H.
Protocol: Development of an Ecological Model to Predict Risk for Acquisition of Clostridium difficile-Associated Diarrhea During Acute Care Hospitalization
ID: 05468 Prom#: N/A Protocol#: N/A

The following items were reviewed and approved through Expedited Review:

- Research Protocol (03/14/2007)
- Abstract
- Request for waiver of consent
- Request to Waive HIPAA Authorization

Expedited Approval was granted on 03/21/2007. This Expedited review will be reported to the fully convened Research & Development Committee on 04/06/2007.

1. Your proposal was reviewed both administratively and scientifically and approved by the Research and Development Committee.
2. Documentation from the USF Health Sciences IRB has been received granting approval. Having both R&D and IRB approval, the project is now fully approved and subjects may now be admitted to the study.

Approval by each of the following is required prior to study initiation:

- Institutional Review Board (IRB 01a)
- Research & Development Committee

About the Author

Susan Elaine Steele received a Bachelor of Science degree in Nursing from Albright College in Reading Pennsylvania in 1977. She completed the Master of Science degree in Adult Health Nursing at the University of South Florida, in Tampa, Florida in 1984. In a nursing career spanning more than 30 years, Ms. Steele has worked in community health and acute care hospital settings in the role of clinician, educator, and administrator. She served for 14 years as wound, ostomy and continence clinical nurse specialist at Bayfront Medical Center in St. Petersburg, Florida, including a portion of the time engaged in doctoral studies at the University of South Florida.

Ms. Steele has been actively involved in many nursing organizations, and served as the President of the Florida Nurses Association, District Thirteen, the President of the Southeast Region Wound, Ostomy and Continence Nurses Association (WOCN), and a member of Sigma Theta Tau, the international nursing honor society. She serves on the Center for Clinical Investigation board for the WOCN, and has published in the society's journal. She is a presenter at national conferences and currently serves as an Assistant Professor of Nursing at Georgia College and State University in Milledgeville, Georgia.