UNIVERSIDADE FEDERAL DO RIO GRANDE DO SUL
CENTRO DE BIOTECNOLOGIA
PROGRAMA DE PÓS-GRADUAÇÃO EM BIOLOGIA CELULAR E MOLECULAR

Caracterização Molecular do Microbioma Hospitalar por Sequenciamento de Alto Desempenho

Tese de Doutorado

Pabulo Henrique Rampelotto

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Caracterização Molecular do Microbioma Hospitalar por Sequenciamento de Alto Desempenho

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“A tarefa não é tanto ver aquilo que ninguém viu, mas pensar o que ninguém ainda pensou sobre aquilo que todo mundo vê” (Arthur Schopenhauer)

SUMÁRIO

LISTA DE ABREVIATURAS

CCIRAS	Comissões de Controle de Infecção Relacionada à Assistência à Saúde
CDC	Centers for Disease Control and Prevention
ECU	Emergency Care Unit
HAIs	Hospital-Acquired Infections
ICU-A	Intensive Care Unit A
ICU-B	Intensive Care Unit B
IRAS	Infecções relacionadas à assistência à saúde
IU	Inpatient Unit
MRSA	*Staphylococcus aureus* resistente à meticilina
MU	Medical Unit
OTU	Operational Taxonomy Unit
PCoA	Principal Coordinates Analysis
SC	Surgery Center

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Capítulo I

RESUMO

Instituições de saúde são ecossistemas complexos que incluem potenciais patógenos responsáveis por infecções hospitalares, os quais representam um sério problema de saúde em todo o mundo. Assim, compreender o microbioma hospitalar pode ser essencial para melhor combater as infecções que ocorrem neste ambiente e ajudar na melhoria da assistência à saúde. Com o objetivo de identificar padrões que auxiliem na caracterização do microbioma hospitalar, o objetivo deste trabalho foi analisar e comparar as comunidades bacterianas de 663 amostras de um hospital brasileiro utilizando sequenciamento do gene 16S rRNA. Para aumentar o perfil taxonômico e a especificidade da identificação baseada no 16S rRNA, foi aplicado um processo rigoroso de filtragem da qualidade das sequências para a identificação precisa de táxons bacterianos clinicamente relevantes. Nossos resultados indicam que o ambiente hospitalar é predominantemente habitado por espécies intimamente relacionadas. Observou-se uma dominância massiva de alguns táxons em todos os níveis taxonômicos, onde os dez gêneros mais abundantes em cada unidade hospitalar representaram 64,4% de todos os táxons observados, com maior predominância de *Acinetobacter* e *Pseudomonas*. As análises de alfa e beta diversidade indicaram uma homogeneidade em relação ao agrupamento das amostras. A presença de vários patógenos nosocomiais foi observada. Alguns destes táxons patogênicos também foram diferencialmente abundantes entre as amostras. A análise de co-ocorrência indicou que a rede microbiana presente no ambiente hospitalar apresentou baixa conectividade, formando uma topologia agrupada, mas não estruturada entre grupos de nós (isto é, módulos). Além disso, foi possível detectar relações ecologicamente relevantes entre táxons microbianos específicos; em especial, potencial competição entre patógenos e não-patógenos. De modo geral, esses resultados fornecem novos *insights* sobre diferentes aspectos do microbioma hospitalar e indicam que o sequenciamento do 16S rRNA pode ser uma ferramenta robusta para a caracterização de uma ampla gama de táxons bacterianos clinicamente relevantes em ambientes hospitalares com alta resolução.

ABSTRACT

Healthcare institutions are complex ecosystems that include common potential pathogens responsible for hospital-acquired infections (HAIs), which are a serious problem worldwide. Thus, understanding the hospital microbiome contributes to infection control and management in hospitals and to the improvement of healthcare assistance. In order to identify patterns that help to characterize the hospital microbiome, the aim of this work was to analyze and compare the bacterial communities from 663 samples of a Brazilian hospital using high-throughput sequencing of the 16S rRNA gene. To increase taxonomic profiling and specificity of 16S based identification, a strict sequence quality filtering process was applied for the accurate identification of clinically relevant bacterial taxa. Our results indicate that the hospital environment is predominantly inhabited by closely related species. A massive dominance of a few taxa in all taxonomic levels down to the genera was observed, where the ten most abundant genera in each facility represented 64.4% of all observed taxa, with major predominance of *Acinetobacter* and *Pseudomonas*. Alpha and beta diversity analyzes indicated a general homogeneity regarding sample clustering. The presence of several nosocomial pathogens was revealed. Some of these pathogenic taxa were also differentially abundant among the samples. Co-occurrence analysis indicated that the present hospital microbial network was low connected, forming a clustered topology, but not structured among groups of nodes (i.e. modules). Furthermore, we were able to detect ecologically relevant relationships between specific microbial taxa, in especial, potential competition between pathogens and non-pathogens. Overall, these results provide new insights into different aspects of the hospital microbiome and indicate that 16S rRNA sequencing may serve as a robust one-step tool for microbiological identification and characterization of a wide range of clinically relevant bacterial taxa in hospital settings with high resolution.

1. INTRODUÇÃO

1.1. O AMBIENTE HOSPITALAR

O termo ambiente hospitalar inclui edifícios hospitalares e instituições de saúde com todos os componentes internos que os compõe: pessoas (incluindo pacientes, profissionais e visitantes), ar interior, superfícies, equipamentos médicos, medicamentos, dispositivos, alimentos e resíduos (Bottero et al. 2015; Capolongo et al. 2016). Todos esses componentes abrigam uma variedade de microrganismos, os quais constituem o microbioma hospitalar.

O microbioma encontrado no ambiente hospitalar pode ter características diferentes das de outros ambientes internos devido às suas peculiaridades, como conjunto de pessoas presente, regimes de limpeza, uso de antibióticos e projeto geral do edifício.

Por ser um ambiente dinâmico com alto fluxo de pessoas, incluindo um grupo particular (os pacientes), é provável que o microbioma interno de um hospital seja amplamente influenciado pelos indivíduos presentes e contenha um número desproporcional de patógenos em comparação com outros ambientes. De fato, os hospitais são ambientes únicos que podem levar a novos processos evolutivos como, por exemplo, o surgimento de enterococos resistentes à vancomicina (Davies e Davies 2010).

O interesse em microrganismos presentes no ambiente hospitalar surgiu em 1847, quando Ignaz Semmelweis, um médico húngaro-alemão, descobriu que estudantes de medicina em um hospital de Viena estavam transmitindo infecções pós-parto de salas de autópsia para maternidades, levando a altas taxas de mortalidade. Ignaz Semmelweis introduziu a lavagem compulsória das mãos, a primeira intervenção para uma infecção adquirida no hospital já registrada (Bencko e Schejbalová, 2006). Desde então, a maior

parte dos estudos envolvendo microrganismos em hospitais tem se concentrado, de forma semelhante, na prevenção de surtos causados por patógenos específicos, especialmente aqueles que são perigosos para pessoas que já estão doentes ou têm o sistema imunológico debilitado.

Com o advento das tecnologias de sequenciamento alto desempenho, novos grupos de pesquisa estão tentando caracterizar o microbioma hospitalar como um todo (Poza et al., 2012; Hewitt et al., 2013; Oberauner et al., 2013; Tang et al., 2015). Estes estudos pioneiros vêm demonstrando que a diversidade bacteriana é muito maior que a esperada. No entanto, a informação obtida a partir destes estudos ainda é esparsa e limitada, isto é, geralmente baseada em poucas amostras quando considerado o ambiente complexo de um hospital. Uma visão mais completa do microbioma hospitalar nos ajudará a entender melhor os diferentes aspectos relacionados à ecologia deste ambiente heterogêneo, sujeito a diferentes pressões seletivas, e ainda pouco explorado.

1.2. FONTES E DISPERSÃO DE MICRORGANISMOS NO AMBIENTE HOSPITALAR

A principal fonte de microrganismos no ambiente hospitalar são as pessoas, sendo que a via mais comum de transferência de patógenos ocorre entre as mãos de profissionais de saúde e pacientes. Por isso, acredita-se que as infecções causadas por microrganismos frequentemente advêm da contaminação cruzada entre indivíduos (Tajeddin et al., 2016; Price et al., 2017).

A microbiota humana normal contém várias bactérias com potencial de patogenicidade. A pneumonia, por exemplo, é mais comumente causada por *Streptococcus pneumoniae* e atualmente é responsável pela maioria das mortes e doenças relacionadas à

infecção respiratória na Inglaterra (Health Protection Agency, 2005). Esta bactéria está presente na nasofaringe em aproximadamente 60% das crianças saudáveis em pré-escola (Henriques-Normark e Normark 2010) e as taxas de colonização são importantes na disseminação deste microrganismo nas creches (Abut et al., 2008).

Junto com microrganismos que fazem parte da microbiota humana normal, o corpo pode servir como uma fonte de microbiota transitória, transportando espécies que normalmente não são residentes (WHO, 2009). As mãos são muitas vezes a principal fonte dessas espécies transitórias e podem transportá-las de um local para outro. No entanto, as superfícies inanimadas e os equipamentos hospitalares também são fontes de bactérias, incluindo as potencialmente patogênicas. Nesse sentido, o ambiente hospitalar pode contribuir significativamente para a disseminação destes patógenos. De fato, estudos recentes indicam que o ambiente pode servir de reservatório para microrganismos patogênicos que causam infecções hospitalares (Hota, 2004; Boyce, 2007; Weber et al., 2010; Otter et al., 2011; Faires et al., 2012; Weber et al., 2013; Zarpellon et al., 2015), e a maioria desses organismos pode sobreviver ou persistir por um longo período de tempo (Kramer et al., 2006; de Abreu et al., 2014).

Além disso, microrganismos patogênicos são comumente encontrados nos utensílios dos profissionais de saúde, como vestuário (Wiener-Well et al., 2011), telefones celulares (Brady et al., 2009; Datta et al., 2009), estetoscópios (Youngster et al., 2008), teclados de computadores (Doğan et al., 2011), entre outros.

A contaminação de superfícies e equipamentos aparentemente limpos indica que estes locais muitas vezes são ignorados durante procedimentos de limpeza. Tal premissa reforça a ideia de que muitas vezes os profissionais e visitantes, após tocar um paciente, não se atêm à importância da higienização das mãos e retomam atividades sem se dar conta da possibilidade de disseminar microrganismos. A presença de bactérias resistentes, a

exemplo de *Staphylococcus aureus* resistente à meticilina (MRSA), na microbiota das mãos dos profissionais de saúde e em superfícies inanimadas do ambiente hospitalar reforça a hipótese de contaminação entre indivíduo e ambiente (Haamann et al., 2011).

1.3. INFECÇÕES RELACIONADAS À ASSISTÊNCIA À SAÚDE

As infecções relacionadas à assistência à saúde (IRAS) são definidas como aquelas adquiridas após a admissão do paciente, com manifestação durante a internação ou após a alta, quando puderem ser relacionadas à internação ou procedimentos hospitalares (Brasil, 2016). Entretanto, a ampliação do foco não restrito exclusivamente ao ambiente hospitalar se refere ao fato de que as IRAS podem ocorrer em todos os níveis de atenção à saúde, a exemplo dos ambulatórios, clínicas especializadas e assistência domiciliar. Devido a esse aspecto, o Centro de Controle e Prevenção de Doenças (em inglês: Centers for Disease Control and Prevention - CDC), no *guideline* para precauções de isolamento de 2007, substitui o termo infecção hospitalar por infecções relacionadas à assistência à saúde (Siegel et al., 2007).

Também conhecidas como infecções nosocomiais, as IRAS representam um grave problema de saúde pública, afetando milhões de pessoas em todo o mundo, sendo a sexta causa de morte nos Estados Unidos e dados semelhantes são relatados na Europa (Peleg e Hooper, 2010).

A prevalência anual de infecções hospitalares em países desenvolvidos varia entre 3,5% e 12% (WHO, 2011). Nos EUA, anualmente, estas infecções têm um custo estimado de 36 a 45 bilhões de dólares e resultam em aproximadamente 100.000 mortes (Scott, 2009). Em hospitais europeus, a cada 18 pacientes, um apresenta algum tipo de infecção

hospitalar e estima-se que, a cada dia, cerca de 80.000 pacientes estejam com pelos menos um tipo de IRAS (ECDC, 2013). O total anual de pacientes com IRAS em hospitais europeus no período 2011-2012 foi estimado em 3,2 milhões, com um intervalo de confiança variando de 1,9 a 5,2 milhões de pacientes (ECDC, 2013).

Em países subdesenvolvidos ou em desenvolvimento, a prevalência anual de infecções hospitalares varia entre 5,7% e 19,1% (WHO, 2011), embora os dados sejam limitados e, muitas vezes, não confiáveis. A proporção de pacientes com infecção adquirida em UTIs varia de 4,4% a 88,9% (Allegranzi et al., 2011). De modo geral, aproximadamente 30% dos pacientes em Unidades de Terapia Intensiva (UTIs) são afetados por IRAS. Apesar dos avanços no âmbito legislativo e da implantação das Comissões de Controle de Infecção Relacionada à Assistência à Saúde (CCIRAS) nos hospitais brasileiros, até o presente momento não há dados oficiais publicados pelos órgãos competentes sobre a incidência e prevalência de IRAS no Brasil.

Até 2017, a única avaliação de amplitude nacional realizada no Brasil havia sido um estudo publicado em 1995 (Prade et al., 1995), que identificou uma taxa de prevalência de 15,0% de IRAS em 99 hospitais brasileiros, dados de 1994, ou seja, há mais de duas décadas. Em junho de 2017, Fortaleza e colaboradores publicaram um novo estudo da prevalência de IRAS no Brasil. O estudo contou com os dados de 152 hospitais de cinco macrorregiões brasileiras, referentes ao período de 2011 a 2013. A prevalência de IRAS no Brasil reportada no estudo foi de 10,8% (Fortaleza et al. 2017). De acordo com a WHO, a prevalência anual de infecções hospitalares no Brasil pode ser de 14% (WHO, 2011). Em outro estudo multicêntrico de UTIs brasileiras, 51,2% dos pacientes estavam infectados e 79,4% tinham pelo menos uma IRAS (Braga et al., 2018). Por sua vez, Pessoa-Silva e colaboradores (2004) demonstraram que o número de infecções em UTIs neonatais brasileiras chega a ser 9 vezes maior do que nos EUA (Pessoa-Silva et al., 2004).

Dentre os microrganismos causadores das IRAS, as bactérias contribuem com aproximadamente 95% das infecções, com um percentual considerável de isolados multirresistentes a antibióticos (Mayhall, 2013). O termo "multirresistente" refere-se às bactérias resistentes a três ou mais classes de antibióticos (Magiorakos et al., 2012). Em alguns casos, as cepas também adquiriram maior virulência e melhoramento dos meios de transmissão (Davies e Davies, 2010).

A manifestação de resistência a antibióticos pelos microrganismos, inicialmente, relaciona-se com a prescrição de agentes antimicrobianos para o tratamento das IRAS, ocasionando a pressão seletiva (Blair et al., 2015). O uso de antibióticos pode selecionar cepas naturalmente resistentes ou aquelas previamente sensíveis, que adquiriram mecanismos de resistência. Com o crescente uso inadequado destes fármacos nas últimas décadas, o surgimento de cepas bacterianas multirresistentes vem aumentando significativamente, constituindo um dos principais desafios atuais enfrentados por hospitais do país e do mundo (Livorsi et al., 2013). Bactérias multirresistentes, como *Acinetobacter baumannii*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae e Enterococcus* spp., tornam-se cada vez mais comuns nas instituições de saúde. São bactérias que podem causar infecções graves e frequentemente mortais, como infecções da corrente sanguínea, pneumonia e infecções do trato urinário.

Por isso, a prevenção da disseminação de bactérias potencialmente patogênicas nos estabelecimentos de saúde é uma grande necessidade no âmbito nacional e internacional.

1.4. ESTRATÉGIAS PARA PREVENÇÃO E CONTROLE DE MICRORGANISMOS PATOGÊNICOS

A persistência de bactérias em superfícies e equipamentos do ambiente hospitalar está relacionada à frequência na qual estes são limpos, à forma como é realizada a limpeza, e ao uso adequado dos desinfetantes conforme concentração indicada e técnica adequada de desinfecção de equipamentos de acordo com as indicações do fabricante, dentre outros fatores.

Atualmente, a identificação de possíveis reservatórios microbianos a fim de prevenir a disseminação de microrganismos causadores de infecções nos estabelecimentos de saúde é uma importante estratégia para o controle da resistência bacteriana e das IRAS, por favorecer a revisão e elaboração de medidas preventivas.

Pensando na importância do ambiente hospitalar no contexto das infeções hospitalares, o Ministério da Saúde elaborou o Programa Nacional de Prevenção e Controle de Infecções Relacionadas à Assistência à Saúde (Brasil, 2016), visando a segurança do paciente e a garantia, por parte das instituições, de oferecer aos pacientes um local limpo e um ambiente com menor carga de contaminação possível, contribuindo para a redução da possibilidade de transmissão de infecções oriundas de fontes inanimadas.

Entretanto, a persistência de patógenos em vários locais do ambiente hospitalar e as diversas possibilidades de dispersão indicam que as medidas de prevenção usualmente adotadas, bem como as práticas de esterilização e rotinas de limpeza em vigor, não são suficientes para proteger os pacientes. Isso ocorre porque, apesar do grande interesse no estabelecimento de hospitais com taxas reduzidas de infecções adquiridas, ainda não há nenhuma ferramenta de análise sistemática capaz de rastrear e identificar, com rapidez e precisão, a presença de microrganismos patogênicos nestes ambientes. A eficácia das medidas de limpeza e prevenção ainda é baseada principalmente em métodos de cultivo, embora tais metodologias tradicionais só possam detectar uma pequena fração da

diversidade microbiana, dependendo das condições de cultura que estão sendo utilizadas e da viabilidade dos microrganismos (Chen et al., 2017a).

Por isso, há uma grande necessidade de estudos baseados em novas tecnologias moleculares que venham a caracterizar em detalhes os diferentes aspectos da ecologia microbiana associada ao ambiente hospitalar. Este conhecimento será essencial para o desenvolvimento de novas práticas e medidas preventivas que reduzam significativamente as altas taxas de IRAS.

1.5. TECNOLOGIAS MOLECULARES PARA A CARACTERIZAÇÃO DO MICROBIOMA HOSPITALAR

As novas técnicas moleculares de sequenciamento de DNA vêm sendo utilizadas com sucesso para a caracterização de microrganismos com alto grau de precisão. Estes métodos apresentam uma grande vantagem sobre os métodos tradicionais de cultivo em meios de cultura pelo simples fato de que grande parte dos microrganismos presentes em amostras ambientais não são cultiváveis ou são de difícil cultivo (Hilton et al., 2016).

O sequenciamento do gene da unidade 16S do RNA ribossômico (16S rRNA) é particularmente relevante em estudos de caracterização microbiana, pois este gene é altamente conservado em todas as bactérias (Clarridge, 2004). Além disso, as regiões hipervariáveis do 16S rRNA são assinaturas únicas que permitem uma identificação específica de táxons, sendo amplamente utilizadas para a identificação de microrganismos em uma variedade de habitats (Caporaso et al., 2011; Rampelotto et al., 2013, 2015; Gilbert et al., 2014). Dessa forma, as sequências do gene que codifica o 16S rRNA podem ser usadas para identificar uma grande variedade de espécies dentro de uma comunidade

bacteriana mista e complexa. A Figura 1 apresenta um esquema do gene 16S rRNA e a localização dos *primers* mais comuns usados para estudos de taxonomia.

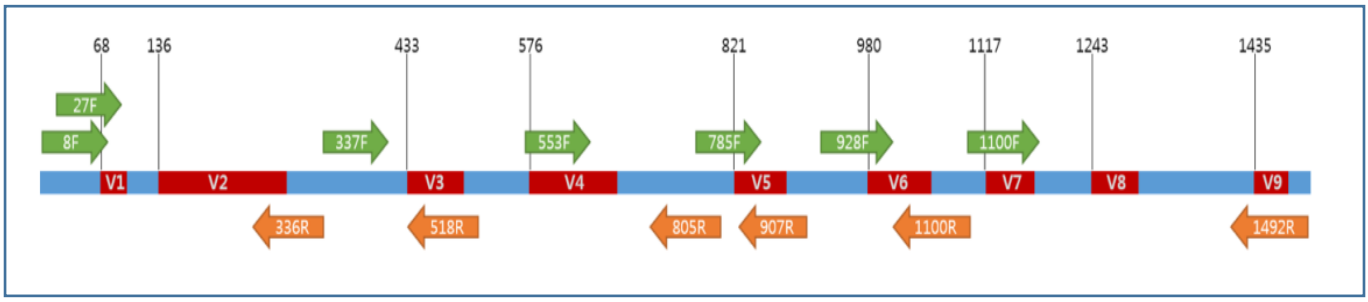

Figura 1. Esquema do gene 16S rRNA e localização dos primers mais comuns usados para estudos de taxonomia. Em azul estão representadas as regiões conservadas e em vermelho as regiões variáves (V1 à V9); em verde são os *primers forward* e em laranja os *primers reverse*.

Consequentemente, a aplicação destas tecnologias moleculares tem o potencial de revolucionar o panorama atual da microbiologia clínica (Srinivasan et al., 2015) e alterar profundamente nossa compreensão de como os microrganismos interagem uns com os outros e com o meio ambiente. Por estas razões, estudos metagenômicos são necessários para caracterizar em detalhes as comunidades microbianas associadas ao ambiente hospitalar. Esse conhecimento nos ajudará a entender melhor os diferentes aspectos relacionados à ecologia microbiana deste ambiente heterogêneo, o qual esta sujeito a diferentes pressões seletivas e é ainda pouco explorado.

Com o advento das tecnologias de sequenciamento de alto desempenho, a identificação e rastreamento de microrganismos em ambientes hospitalares tornou-se uma realidade (Tang et al., 2015; Chen et al., 2017b; Manaka et al., 2017). Esses estudos

pioneiros demonstraram o potencial uso do sequenciamento em larga escala de amplicons do gene 16S rRNA para identificar uma variedade de bactérias associadas ao desenvolvimento de IRAS. No entanto, a informação obtida por estes estudos ainda é limitada e esparsa, geralmente com base em poucas amostras, quando considerado o ambiente heterogêneo e complexo de um hospital.

2. JUSTIFICATIVA

Este trabalho pode contribuir para a compreensão de diversos aspectos relacionados à ecologia microbiana das bactérias presentes no ambiente hospitalar. Além disso, o conhecimento gerado neste trabalho pode servir de base para o desenvolvimento de novas práticas e medidas preventivas que reduzam significativamente os casos de IRAS e, consequentemente, a mortalidade decorrente dessas infecções e os gastos exorbitantes associados ao tratamento deste grave problema de saúde pública que atinge milhões de pessoas em todo o mundo.

3. FORMULAÇÃO DO PROBLEMA

As seguintes questões foram abordadas neste projeto:

- Existem diferenças entre as comunidades microbianas dos diferentes ambientes de um mesmo hospital?
- Existe um padrão que caracteriza o microbioma deste hospital?

4. OBJETIVOS

4.1. OBJETIVO GERAL

Identificar e caracterizar, por sequenciamento de alto desempenho, as comunidades bacterianas presentes em superfícies inanimadas de um hospital brasileiro.

4.2. OBJETIVOS ESPECÍFICOS

- Determinar e comparar os padrões de diversidade de cada ambiente.
- Identificar quais são as amostras que apresentam maior concentração de bactérias.
- Identificar quais bactérias são diferencialmente abundantes em cada ambiente.
- Determinar quais bactérias são mais prevalentes no ambiente hospitalar avaliado.
- Analisar padrões de interação entre as bactérias presentes no ambiente hospitalar avaliado.

5. PARTE EXPERIMENTAL E RESULTADOS

5.1 CAPÍTULO I

Exploring the hospital microbiome by high-resolution 16S rRNA profiling

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Exploring the Hospital Microbiome by High-Resolution 16S rRNA Profiling

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Abstract

The aim of this work was to analyze and compare the bacterial communities from 663 samples of a Brazilian hospital using high-throughput sequencing of 16S rRNA gene. To increase taxonomic profiling and specificity of 16S based identification, a strict sequence quality filtering process was applied for the accurate identification of clinically relevant bacterial taxa. Our results indicate that the hospital environment is predominantly inhabited by closely related species. A massive dominance of a few taxa in all taxonomic levels down to the genera was observed, where the ten most abundant genera in each facility represented 64.4% of all observed taxa, with major predominance of *Acinetobacter* and *Pseudomonas*. The presence of several nosocomial pathogens was revealed. Co-occurrence analysis indicated that the present hospital microbial network was low connected, forming a clustered topology, but not structured among groups of nodes (i.e. modules). Furthermore, we were able to detect ecologically relevant relationships between specific microbial taxa, in especial, potential competition between pathogens and non-pathogens. Overall, these results provide new insight into different aspects of the hospital microbiome and indicate that 16S rRNA sequencing may serve as a robust one-step tool for microbiological identification and characterization of a wide range of clinically relevant bacterial taxa in hospital settings with high resolution.

Keywords

Microbiota – Nosocomial pathogens – Hospital-acquired infections – 16S rRNA – Clinical microbiology– *Acinetobacter* – *Staphylococcus* – *Pseudomonas*

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Introduction

Hospital-acquired infections (HAIs) represent a serious public health problem, affecting millions of people worldwide [1]. Also known as nosocomial infections, they are the fifth leading cause of death in acute-care hospitals. In the United States, these infections cost several billions of dollars and result in approximately 90,000 deaths annually [2,3]. In developing countries, where the burden of endemic health-care-associated infection is significantly higher [4], the prevalence of HAIs varies between 5.7% and 19.1% [1].

One of the challenges in preventing HAIs is understanding the microbial diversity associated with the hospital environment, the sources of infectious agents and the routes of transmission. Recent studies suggest that environmental contamination plays a significant role in HAIs and several pathogens can persist for months in surfaces and serve as vehicles of transmission and dissemination in hospital facilities [5-7]. Thus, understanding the hospital microbiome could be essential to maintain low levels of HAI infections and to help improving healthcare assistance.

DNA sequencing of the 16S rRNA gene has been successfully used for characterization of microbial populations in a variety of habitats [8-11]. The advantage of this approach is that all microbial taxa may be detected and the limitations of culture conditions are easier to overcome [12]. Consequently, the application of such molecular methods has the potential to revolutionize the landscape of clinical microbiology and infectious diseases [13-15], and reveal which bacteria are present in hospitals and how they interact with each other and the environment. For these reasons, metagenomic studies involving both whole-genome sequencing and targeted gene sequencing are necessary to characterize in details the microbial communities associated with the hospital environment.

With the advent of next generation sequencing technologies, which allowed the massively parallel sequencing of the 16S rRNA gene, the identification and tracking of bacterial diversity in hospital environments has become feasible. These pioneer studies demonstrated the potential use of amplicon sequencing to identify a variety of pathogens associated with the development of HAIs [16-19]. Nevertheless, the information obtained from them is limited and sparse, i.e. usually based on few samples when considered the complex environments of a hospital. A comprehensive view of the hospital microbiome will help us better understand the different aspects concerning the microbial ecology of this heterogeneous environment, subject to different selective pressures, and still poorly

explored. Furthermore, it will be essential for the development of new practices and preventive measures able to significantly reduce the high rates of hospital infections.

In order to identify new patterns that help to better characterize the hospital microbiome, the aim of this work was to analyze and compare the bacterial communities from different inanimate surface environments of a Brazilian teaching hospital using high-throughput sequencing of the 16S rRNA gene. Overall, our results indicated that the hospital microbiome presents a homogeneous structure composed of a massive dominance of a few taxa in all taxonomic levels and a low connected microbial network forming a clustered topology.

Materials and Methods

2.1 Sampling site and collection

The study was carried out at a tertiary-level teaching hospital with 200 beds located in southern Brazil. 111 surface samples were monthly collected at six hospital facilities between April-September 2015, including two Intensive Care Units (ICU-A and ICU-B), one Surgery Center (SC), one Medical Unit (MU), one Inpatient Unit (IU), and one Emergency Care Unit (ECU). In total, 663 samples were collected during the six months period of the study (3 samples were discarded due to problems during the sequencing). The types of surface sampled within the facilities were chosen based on the frequency with which the surfaces were touched (here defined as high-touch surfaces), such as workstations, medical and surgery devices. All sampling locations and their characteristics are given in Supplementary Table S1. Beyond individual characterization, samples were also grouped in four categories, named Month, Facility, Room, and Sample-Type. For example, all samples collected in April were grouped in the "April" Month category; all samples collected in the Emergency Care Unit during the 6 months period of sampling were grouped in the "ECU" Facility category. Room and Sample-Type categories followed the same principle.

Sterile swabs and gloves were used for sampling collection. Swabs were moistened with sterile saline solution and streaked across the surface of each sample. After sampling, the swabs were transported back to the laboratory for DNA extraction, library preparation and DNA sequencing.

Environment measurements

Measurements of relative humidity and air temperature were carried out soon after the sample collection using a digital hygro-thermometer (Incoterm - TTH100). For surface temperature measurements, a digital laser infrared thermometer (GM300, Benetech) was used.

DNA extraction, PCR amplification, and amplicon sequencing

DNA was extracted following an optimized magnetic bead-based DNA extraction and purification protocol, owned by Neoprospecta Microbiome Technologies (Brazil). Barcoded PCR amplification was performed using the 341F and 806R primers (with 465 bp amplicons flanking the highly variable V3-V4 region of the 16S rRNA gene sequence) with the following conditions: the first PCR primers contain the Illumina sequences based on TruSeq structure adapter (Illumina, San Diego, CA), allowing the second PCR with indexing sequences. PCR was always carried out in triplicates using Platinum Taq (Invitrogen, USA) with the conditions: 95°C for 5 min, 25 cycles of 95°C for 45 s, 55°C for 30 s and 72°C for 45 s and a final extension of 72°C for 2 min for PCR 1. In PCR 2 the conditions were 95°C for 5 min, 10 cycles of 95°C for 45 s, 66°C for 30 s and 72°C for 45 s and a final extension of 72°C for 2 min. Taq Platinum was chosen due to its capacity to better amplify samples with low amounts of DNA (i.e. < 5ng) and PCR cycles for the amplicon PCR were reduced to 21 to diminish PCR bias. The final PCR was cleaned up using AMPureXP beads (Beckman Coulter, Brea, CA) and samples were pooled in the sequencing libraries for quantification. Library estimations were performed with Picogreen dsDNA assays (Invitrogen, USA), and then the libraries were diluted for accurate quantification by qPCR using KAPA Library Quantification Kit for Illumina platforms (KAPA Biosystems, Woburn, MA). The libraries were sequenced in a MiSeq system using V2 kit, with a single-end 300 nt run.

16S rRNA reads processing for downstream analyses

Sequencing raw data from MiSeq was processed using a customized python script. Briefly, all the reads were individually submitted to a quality filter, based on the sum of the

DNA bases probabilities errors, allowing a maximum of 1% of accumulated errors. Subsequently, the DNA sequences corresponding to the Illumina adapters were removed. Sequences that presented 100% identity were clustered and defined as Operational Taxonomy Unit (OTU). If any cluster was represented by fewer than 5 reads, it was not considered in further analysis. Each OTU was then aligned against a private reference alignment database (owned by Neoprospecta) at the 99% identity level, using Blast [20]. The taxonomy associated with each OTU was assigned as the taxonomy associated with the reference sequence defining the OTU. For all OTU-based analyses except alpha diversity and co-occurrence network, the original OTU table was normalized using cumulative sum scaling (CSS) method [21]; for alpha diversity, the original OTU table rarified to a depth of 1000 sequences per sample.

Community composition and diversity analysis

QIIME version 1.9.0 was used to estimate alpha and beta diversity [22]. OTU abundances were used to calculate the alpha diversity metrics, including OTU richness (unique OTUs), ChaoI richness estimation and Shannon's diversity indices. For overall comparison of significant differences among bacterial communities (i.e. beta diversity), principal coordinates analysis (PCoA) was performed. Samples were grouped in four categories, named Month, Facility, Room, and Sample-Type (Table S1). A matrix using Bray Curtis and Sørensen–Dice metrics for each pair of environments was calculated. The distances were turned into points in space with the number of dimensions one less than the number of samples. The first three principal dimensions were used to plot a three-dimensional graph that was visualized using EMPeror [23].

To achieve statistical confidence for the sample grouping observed by PCoA (Month, Facility, Room, and Sample-Type), we performed the ANOSIM multivariate test using the vegan package through the compare_category.py script of QIIME. The otu_category_significance.py script was run using the ANOVA to find OTUs whose members are differentially represented among the hospital facilities. Moreover, to analyze whether there is any significant relationship between samples and environmental parameters, we performed the Mantel test. The most prevalent OTUs across samples were analyzed with compute_core_microbiome.py at different cut-off values.

Co-occurrence network analysis

Non-random co-occurrence network analyses were performed using SparCC from the raw count OTU table [24]. Ten interactions were used to estimate the median correlation of each pairwise and the statistical significance of the correlations was calculated by bootstrapping with 100 iterations. SparCC correlations with a magnitude of 0.9 and statistical significance ($p<0.01$) were incorporated into network analyses. The nodes in the reconstructed networks represent the OTUs, whereas the edges (that is, connections) correspond to a strong and significant (positive or negative) correlation between nodes. In order to describe the topology of the resulting network, two centrality measures (i.e. Betweenness Centrality and Eigenvector Centrality) were calculated and the network was visualized using the interactive platform Gephi [25].

Results

Library characterization

For the bacterial community profiling at high-resolution and with high accuracy, we applied a rigorous filtering process at different depths/levels before any taxonomic analysis. The resulting library after all filtering steps is summarized in Table S1.

In total, 663 samples were collected during the six months. 502 (75.7%) samples contained classifiable sequences while 161 (24.3%) did not present sequences after the filtering process. Noteworthy, the library presented some particular features that were taken into account before further analysis. First, a relative low number of sequences in most libraries was observed, which can be partially related to the rigorous filtering process, but most importantly, due to the intrinsic feature of the hospital environment on presenting a few number of microorganisms (when compared to other environments) as a consequence of its constant cleaning and sterilization procedures. Second, a large variability in library sizes across samples was observed, which indicated that rarefying the libraries with a non-parametric test should be chosen as a method of normalization.

Composition of bacterial communities

After all trimming steps, the resulting library composed by the 502 samples contained 7,925,186 classifiable sequences grouped in 878 OTUs, which belong to 567 species and 203 genera. This dataset of high quality classifiable sequences was used to compute the final OTU table. Due to such high variability in library size, the raw OTU table was normalized using CSS.

The analysis of the sequences showed the presence of only five phyla (Figure 1). A major dominance of Proteobacteria was observed in all facilities (67.5%), with smaller proportions of Firmicutes (22.0%), Actinobacteria (5.0%), Bacteroidetes (3.4%), and Fusobacteria (1.9%). Such dominance of a few taxa was observed in all taxonomic levels. The ten most abundant genera in each Facility represented 64.4% of all observed taxa (Table 1), with major predominance of *Acinetobacter* and *Pseudomonas*.

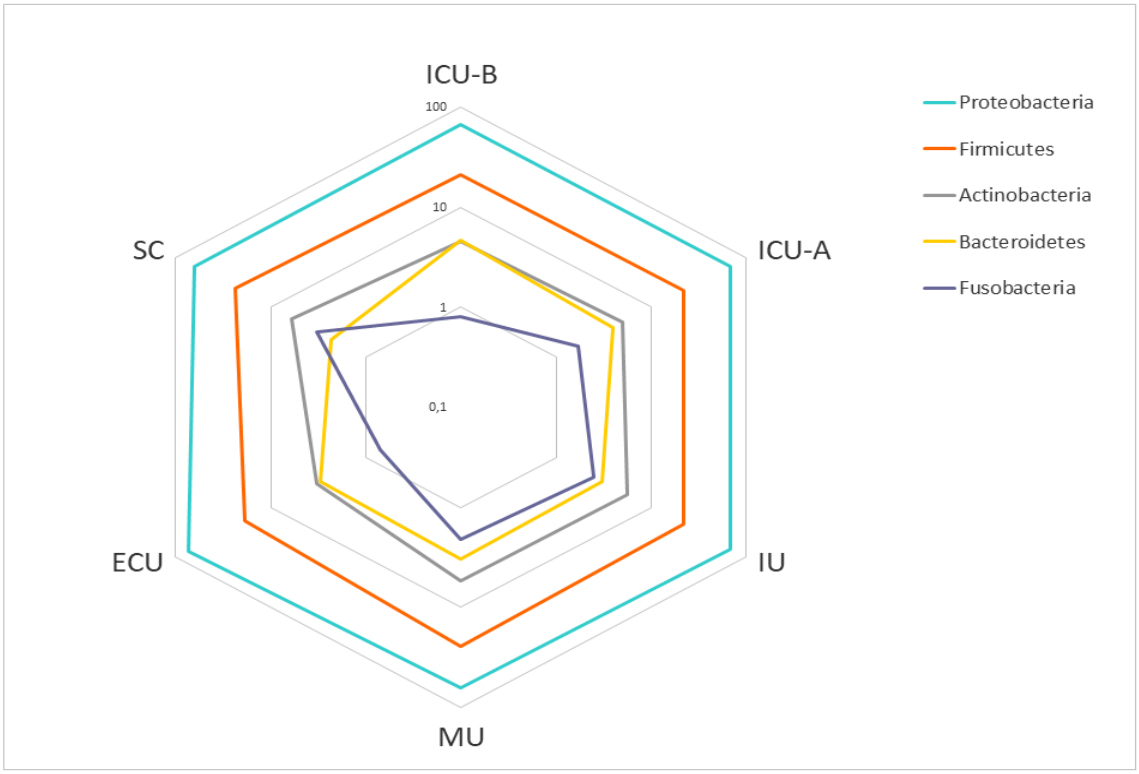

Figure 1. Abundance of the main phyla in the hospital facilities. Relative abundances are shown in percentage (%) on a log scale. Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Table 1. Most abundant genera in the hospital facilities. Relative abundances are shown in percentage (%). Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Phyla	**Genera**	**Total**	**ICU-B**	**ICU-A**	**IU**	**MU**	**ECU**	**SC**
Proteobacteria	*Acinetobacter*	17.2	17.8	15.9	18.1	14.9	21.6	15.0
Proteobacteria	*Pseudomonas*	16.2	17.9	16.7	13.1	16.5	18.4	14.4
Firmicutes	*Staphylococcus*	6.8	9.0	8.6	6.4	5.9	3.5	7.2
Proteobacteria	*Klebsiella*	6.6	8.5	9.0	6.5	7.2	2.2	6.4
Firmicutes	*Streptococcus*	3.5	2.3	3.8	3.5	4.5	1.4	5.7
Proteobacteria	*Pantoea*	3.4	3.5	4.9	5.0	1.0	3.4	2.9
Firmicutes	*Bacillus*	3.3	1.2	2.0	3.7	5.5	6.0	1.6
Proteobacteria	*Escherichia*	2.9	4.7	3.3	2.4	2.3	2.2	2.8
Proteobacteria	*Stenotrophomonas*	2.3	1.7	1.7	2.7	2.8	3.6	1.5
Proteobacteria	*Moraxella*	2.2	1.7	1.7	2.5	1.7	3.1	2.5

Structure and diversity of bacterial communities

To explore the relationship among the bacterial communities of all samples, PCoA analyses based on Sorensen-Dice and Bray Curtis indices were performed. Both analyses demonstrate that no pattern of clustering was observed (Figure 2a and b).

To investigate whether the sample grouping in different categories was statistically significant, the non-parametric multivariate statistical test ANOSIM (analysis of similarity) was performed on the distance matrices generated from the beta diversity step. The p-value observed indicate that there were significant differences for the four categories. However, the low correlation value of R suggests that the clustering of samples based on the categories is relatively weak (Table S2).

When attempting to correlate the sample grouping with environmental parameters, no significant relationship was observed for Surface Temperature, Ambient Temperature and Relative Humidity (Table S3).

We next attempted to study the bacterial diversity in each grouping category using Shannon and Simpson diversity metrics. The results are summarized in Figure 3. For Facility category, no significant difference was observed. For Room and SampleType category, differences were only evident in some cases.

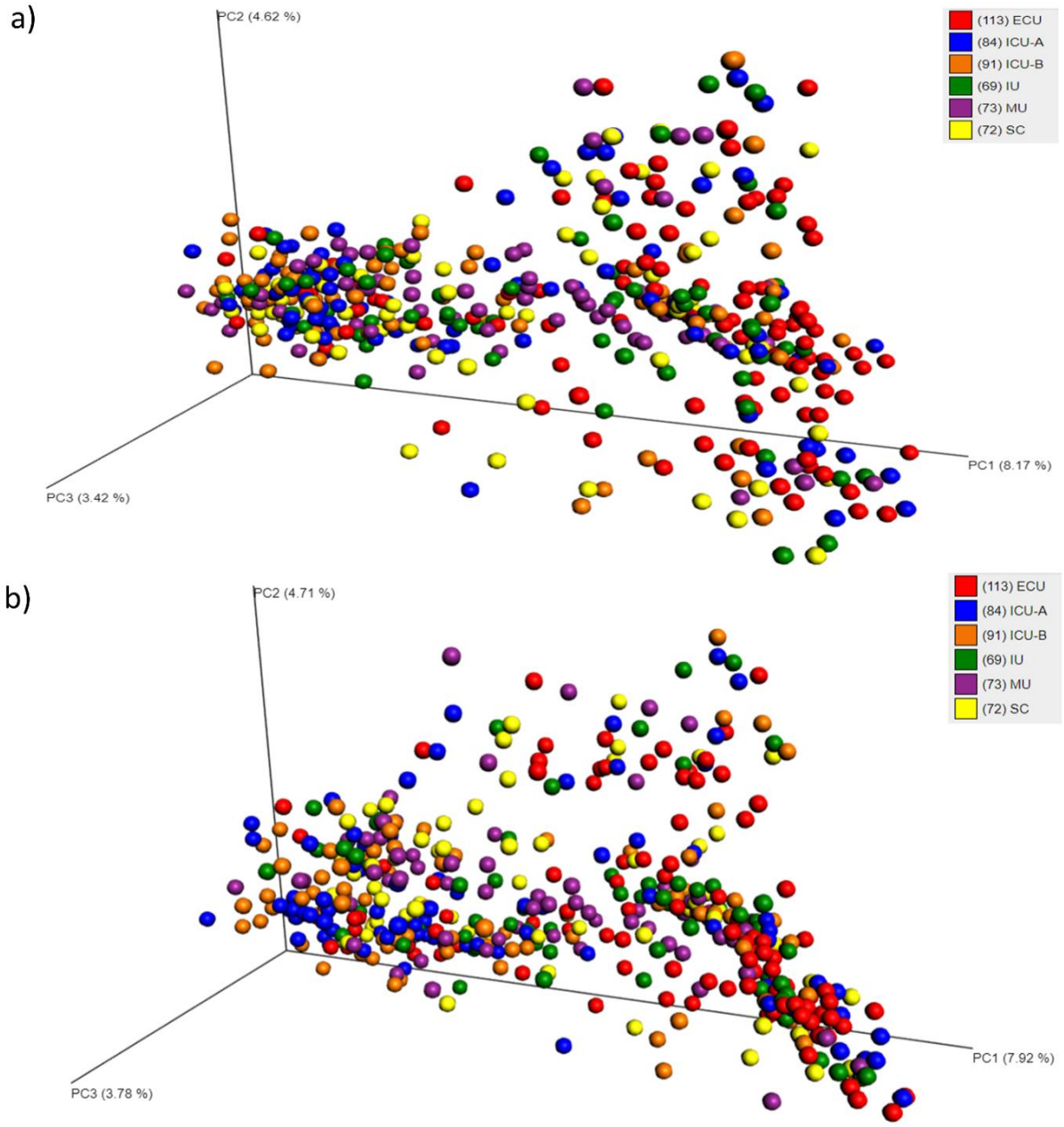

Figure 2. Principal coordinates analysis (PCoA) plot based on Bray-Curtis (a) or Sorensen-Dice (b) dissimilarity depicting the clusters of bacterial communities grouped according to the Facility category. Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

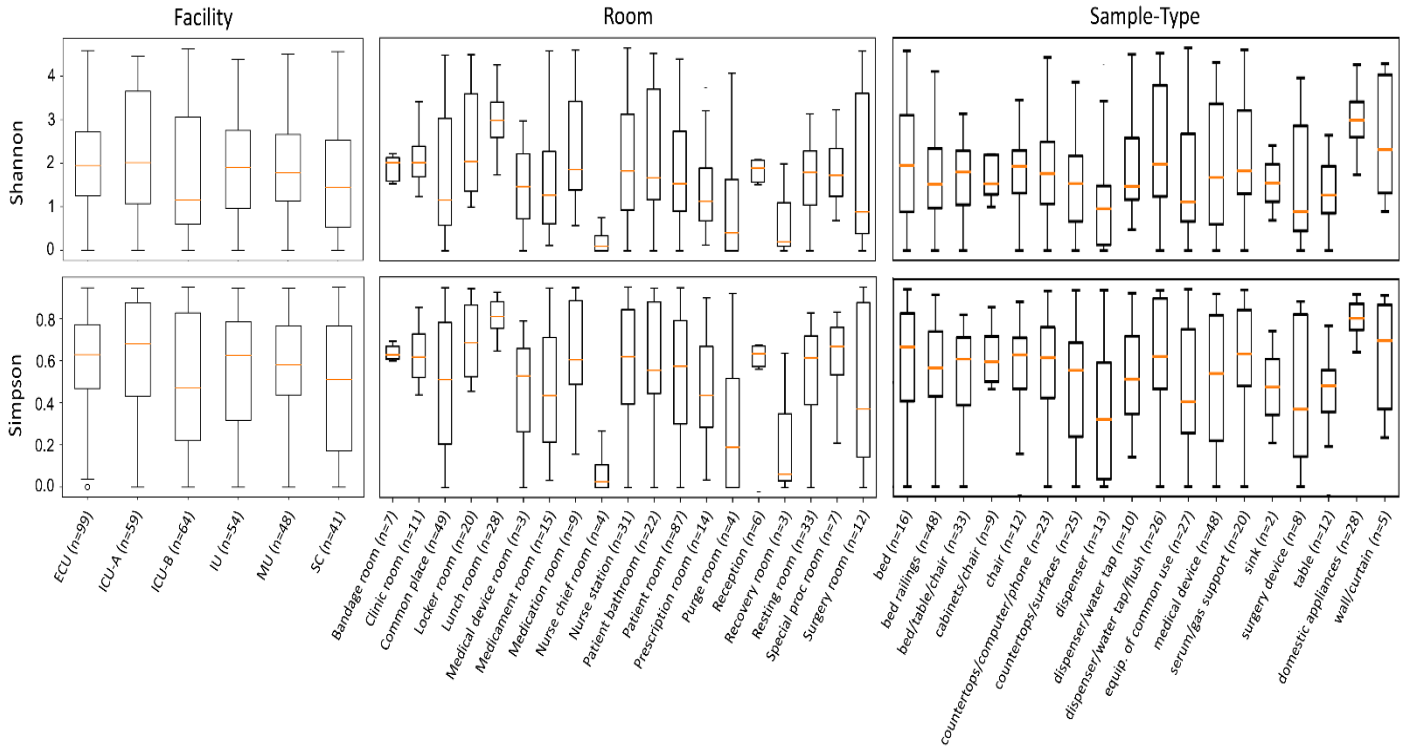

Figure 3. Boxplots showing the distribution of Shannon and Simpson diversity indexes for samples grouped in Facility, Room, and Sample-Type categories. Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Single OTUs

From the 878 OTUs, 347 (42.6%) were present only once (Table S4). With the exception of *Pseudomonas cremoricolorata*, which is one of the taxonomically and ecologically closely related species of the *Pseudomonas putida* species complex, the most prevalent species from these OTUs are usually not associated with the hospital environment nor considered potential pathogens.

In order to find any pattern among the single OTUs, their percentages in each grouping category were summarized and plotted in Figure 4. As observed in Figure 4b, ECU was the facility with the highest percentage of OTUs (25.7%), and ECU with the lowest (7.9%).

Although 19% of the single OTUs were found in SC, only 8% and 6% were found in Surgery room and Surgery device, respectively, which indicate that other non-strict SC places, i.e. those also found in other facilities (especially Lunch room, Common place, and Locker room) concentrated more of such transient, sporadic taxa. In other facilities, the prevalence of single OTUs was observed in Sample-Type (Figure 4d) related to Patient room and Lunch room (Figure 4c).

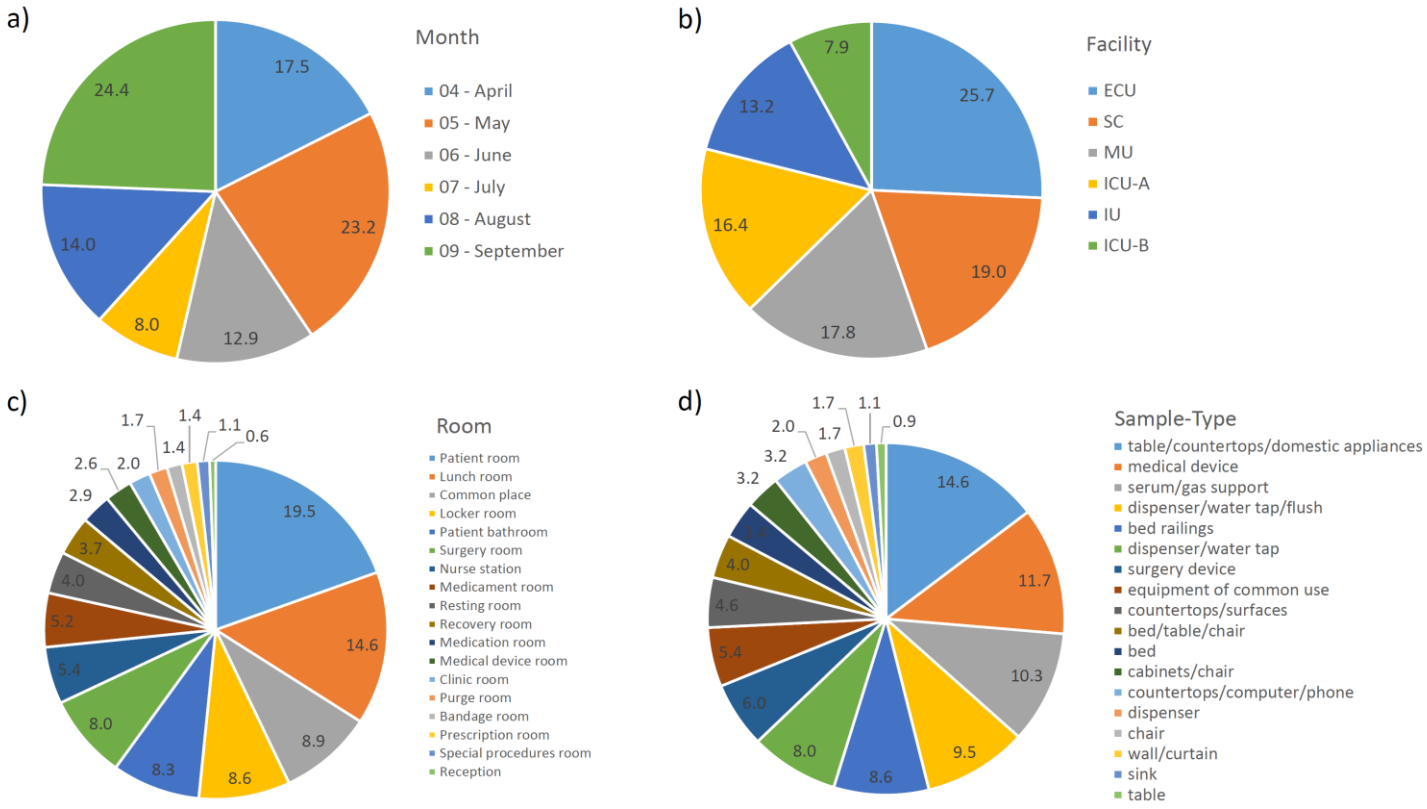

Figure 4. Percentage of single OTUs in each grouping category. (a) Month; (b) Facility; (c) Room; (d) Sample Type. Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Differential abundance

The QIIME Python script group_significance.py was used to calculate significant changes, using the Kruskal-Wallis analysis as the significance test. Differences were considered significant when Bonferroni adjusted p-values < 0.05. The Kruskal–Wallis test identified 12 OTUs showing differential abundances among the Facilities (Table 2). Most of them were associated with ICU-B and ICU-A. The presence of nosocomial pathogens

was also notable, including *Acinetobacter baumannii*, *Staphylococcus epidermidis*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Acinetobacter nosocomialis*.

Table 2. Differentially abundant OTUs in each hospital facility. Statistical confidence was accessed using the Krustal-Wallis test.

OTU	Species	Bonferroni p	ICU-B	ICU-A	IU	MU	ECU	SC
596827	*Pseudomonas stutzeri*	1.78e-10	4.41	2.34	0.55	2.92	0.51	1.20
624096	*Acinetobacter baumannii*	7.00e-06	4.50	4.42	1.87	3.65	1.22	3.07
623550	*Staphylococcus epidermidis*	2.72e-05	4.66	4.12	3.14	3.30	1.25	4.13
602646	*Aerococcus viridans*	0.01	0.09	0.30	0.40	0.11	0.23	1.48
617614	*Anaerococcus vaginalis*	0.01	1.33	0.74	0.00	0.00	0.44	0.13
587128	*Murdochiella asaccharolytica*	0.01	1.30	0.57	0.00	0.00	0.34	0.24
584857	*Porphyromonas bennonis*	0.01	1.30	0.85	0.00	0.00	0.41	0.12
624114	*Pseudomonas aeruginosa*	0.01	2.35	0.99	0.48	1.52	0.32	0.92
614684	*Acinetobacter lwoffii*	0.01	1.85	1.15	0.97	1.17	3.23	0.35
624449	*Escherichia coli*	0.01	5.46	4.37	2.92	3.22	2.21	3.92
598572	*Staphylococcus hominis*	0.01	2.38	1.96	1.30	0.62	0.54	0.99
607150	*Acinetobacter nosocomialis*	0.02	1.36	1.02	0.00	0.00	0.66	0.25

Most prevalent OTUs across samples

Out of the total 878 bacterial OTUs identified in the hospital community, 32 were present in more than 10% of the samples (Figure 6), and 70 were present in more than 5% of the samples. The three most abundant were *Escherichia coli* – OTU624449 (38%), *Staphylococcus epidermidis* – OTU623550 (35%), *and Acinetobacter baumannii* – OTU624096 (30%). Here as well, the presence of several nosocomial pathogens was notable. Indeed, 30% of the OTUs present in more than 5% of the samples were composed by nosocomial pathogens and 20% by rare nosocomial pathogens; only 14% were composed by non-pathogens. The pathogen status of the 70 OTUs is presented in Table S5.

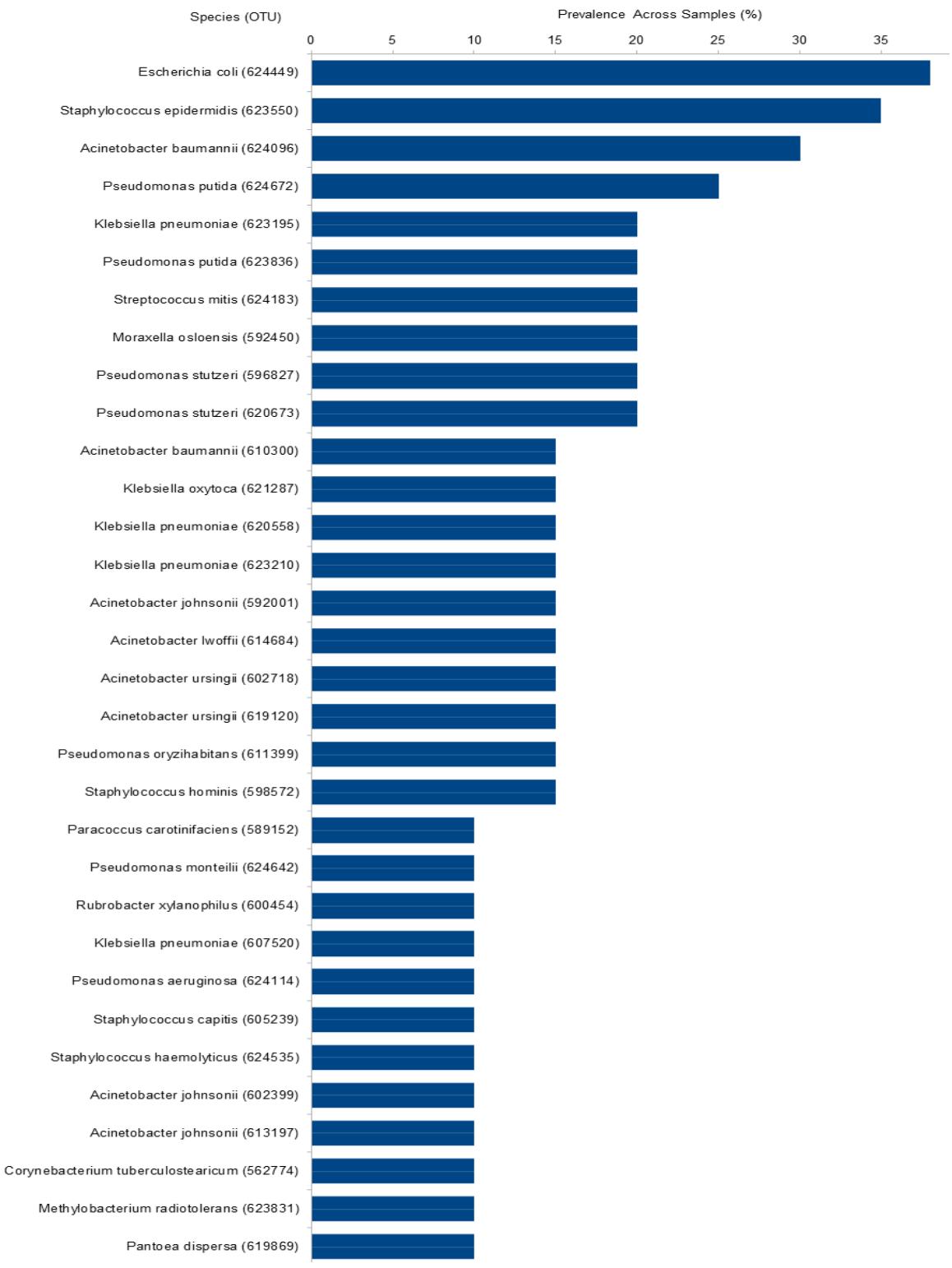

Figure 5. Prevalence of the 32 taxa present in more than 10% of the samples. Axis X represents species' name followed by OTU number between parentheses.

Co-occurrence network analysis

The hospital microbial network (Figure 6) consisted of 70 nodes or OTUs (representing the 70 taxa present in more than 5% of the samples) and 274 edges (with a mean of 3.9 edges per node). The clustering coefficient (that is, the extent to which nodes are embedded in their neighborhood) was 0.074 and the modularity index was 0.282 (values >0.4 suggest that the network has a modular structure). Those results indicated that the hospital microbial network was relatively low connected, forming a clustered topology, but not structured among groups of nodes (i.e., modules). From the 274 interactions (Table S6), 140 (51.46%) were negative and 134 (48.54%) were positive.

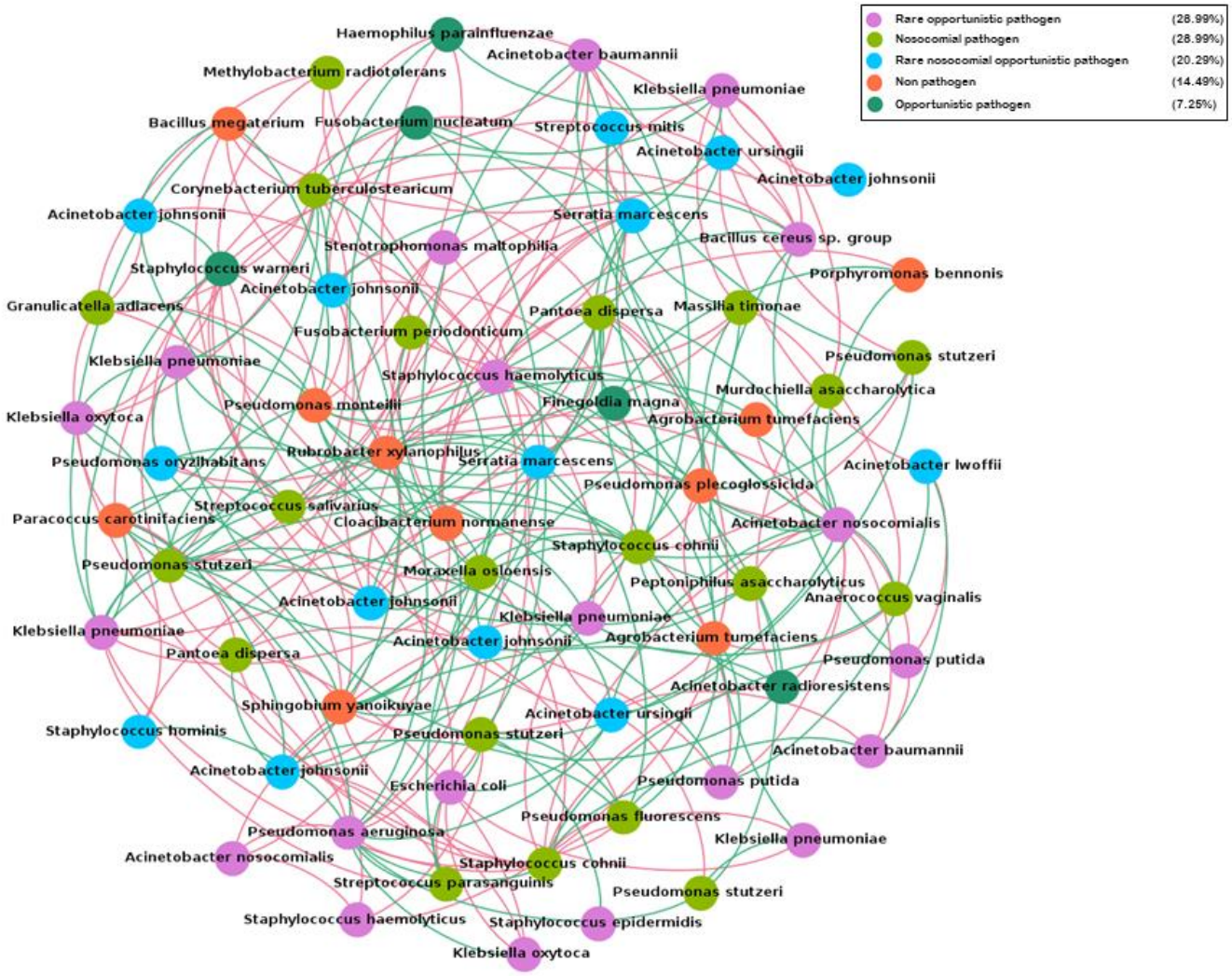

Figure 6. Network analysis of the hospital microbiome present in more than 5% of the samples. Nodes are colored according to the pathogen status of each species (Table S5). Red edges represent negative interactions and green edges represent positive interactions.

To further explore the clinical relevance of the spatial co-occurrence network from the hospital microbiota, we analyzed the interaction patterns of non-pathogenic species against nosocomial and rare nosocomial pathogens. The results indicated that 43.47% of the interactions were negative, including negative interactions with several nosocomial pathogens like *Acinetobacter nosocomialis*, *Klebsiella pneumoniae*, *Serratia marcescens*, and *Staphylococcus haemolyticus* (Table S6).

To study the influence of potential keystone species within the hospital microbial network, two measures were used: betweenness centrality (which indicates the relevance of a node as capable of holding together communicating nodes) and eigenvector centrality (used to measure the importance of a node by the number of important nodes the node links to). The ranking of OTUs was different for each parameter, which was expected considering that they were calculated in different ways (Table S5). However, they both had a particular feature, i.e. only the first three OTUs in each rank presented high scores to be considered as keystone OTUs. *Acinetobacter nosocomialis* OTU606882, for example, presented the highest number of interactions (Degree: 21) and score for Betweeness Centrality (148.87), though only prevalent in 5% of the samples.

Pattern of samples with no reads

Due to the high number of samples with no reads, we tried to find any pattern related to them. In this regard, samples were grouped in different categories. The results are summarized in Figure 7. In terms of Facilities, SC (33.3%) followed by both ICU-A (27.8%) and ICU-B (26.3%) presented the highest percentage of samples with no reads. Purge Room (50.0%), Surgery Room (45.8%), Recovery Room (44.4%), and Medication Room (33.3%) presented the highest percentage of samples with no reads, while Reception (0.0%), Nurse Chief Room (2.8%), Resting Room (2.8%), and Lunch Room (3.3%) presented the lowest; sample-types related to these rooms followed the same pattern.

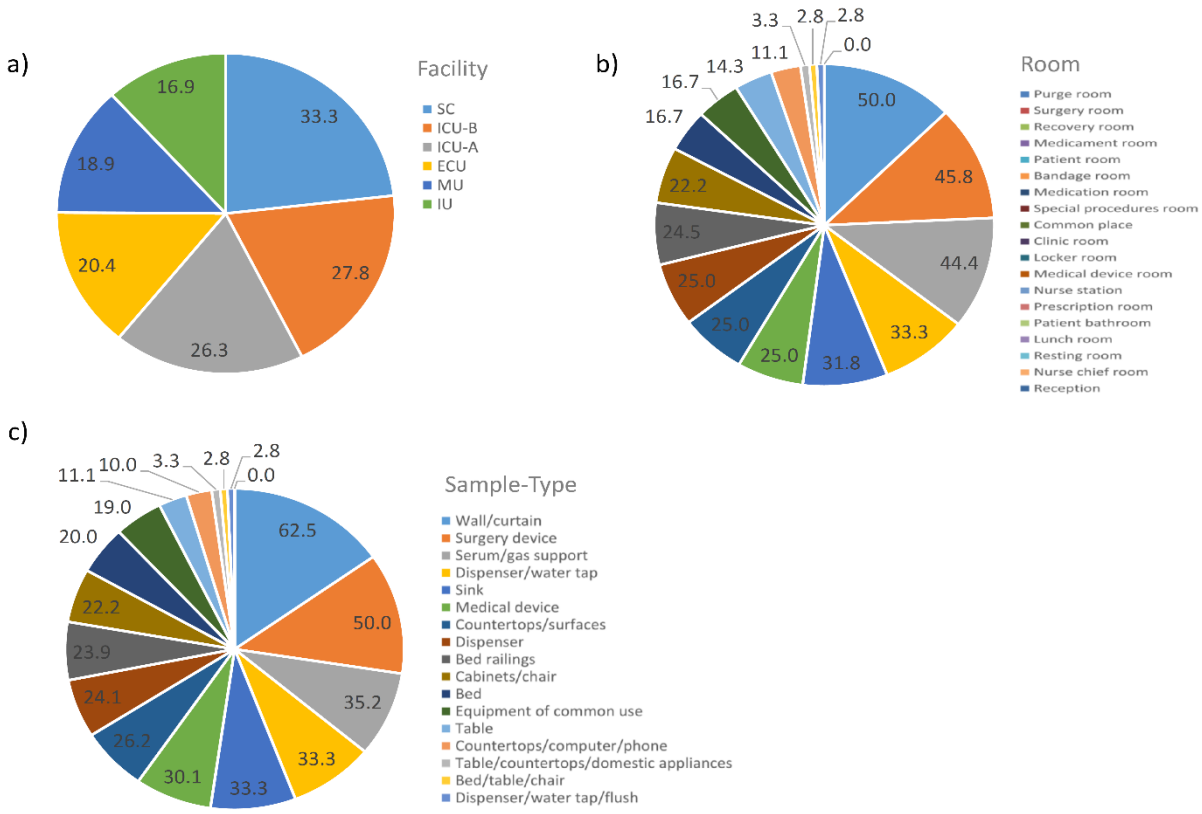

Figure 7. Percentage of the 161 samples with no reads in each one of the three grouping category. (a) Facility; (b) Room; (c) Sample Type. Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Discussion

In this study, we used high quality sequences generated from 16S rRNA gene amplicons to explore the bacterial population at inanimate surfaces of different hospital environments and ascertain the accuracy of routine microbiological identification of a broad range of clinically relevant bacterial taxa. To increase taxonomic profiling and specificity of 16S based identification, we applied a strict sequence quality filtering process including pre-clustering of our sequences into OTUs at the 100% identity level (with the removal of clusters with less than 5 reads), followed by alignment against a curated reference database at the 99% identity level.

Given our interest to identify medically relevant taxa, we focused our analysis on OTUs classified at the species level sharing 99% 16S rRNA gene sequence identity to

match bacterial names with standing in nomenclature. In our analysis, 17±9% of the sequences were not able to be classified at the species level.

Another important aspect of our approach was the primers chosen for the amplification of the 16s rRNA nege. To improve the taxonomic resolution in our analysis, we used the primers 341F and 806R, which generated an amplicon of 465 bp flanking the hypervariable V3-V4 region of the 16S rRNA. This primer set provides ample information for taxonomic classification of microbial communities from specimens associated with human microbiome studies [26]. Therefore the data generated using the V3-V4 primer pair from sequencing using the MiSeq platform can be compared with the existing data in the human microbiome literature, especially for bacterial skin microbiome [27], which are of primary relevance for high-touch surface samples. High-touch surfaces are recognized as a possible reservoir of infectious agents and their contamination can pose a risk also for the spread of pathogens [28, 29]. In fact, there is now compelling evidence from modeling of transmission routes, microbiologic studies, observational epidemiology studies, intervention studies, and outbreak reports that contaminated surfaces contribute to the transmission of hospital pathogens [30]. In a retrospective study, for example, there was more than a two-fold risk of acquiring *Clostridium difficile* if the prior room occupant had this infection [31]. A similar risk was noted for *Acinetobacter* spp. and *Pseudomonas aeruginosa* [32].

But the actual proportion of HAIs attributed to environmental surfaces is largely unknown. This uncertainty is due, in part, to the complexity of microbial transmission in the healthcare setting and the difficulty in directly linking an environmental source to a specific transmission event or infection. Many attempts have been made, but they are individual [30]. At the hospital level, it is important to have a comprehensive infection prevention program that tracks not only HAIs and major nosocomial pathogens individually, but also all other microorganisms composing that community. Such an approach is essential to assess for temporal or geographic patterns that might suggest how microbial taxa spread and interact with each other in the hospital environment, which is of critical importance in determining the role of hospital surfaces and equipment in vectoring pathogen and non-pathogen microbes.

Given the sparse and fragmented knowledge related to the microbial communities present in the hospital environment, our comprehensive study was designed to provide a general picture of the hospital microbiome with its diversities and dynamics while being able to focus as well on those taxa of primary relevance for HAIs. Our findings complement

the major work of Lax et al. (2017) focused on the bacterial dynamics among hospital surfaces, patients, and staff over the course of 1 year as a new hospital became operational [33]. Overall, the results of beta diversity revealed an overlap among the bacterial communities present in the six facilities, which suggests that the hospital microbiome presents a homogeneous structure. Differences in alpha diversity were dependent on individual sample grouping at Room and Sample-Type categories.

Contrasting with previous studies suggesting for higher diversity of microbial communities in hospital environments [16, 19], our study indicates a massive dominance of a few taxa in all taxonomic levels down to the genera, where the ten most abundant genera in each facility represented 64.4% of all observed taxa. These differences may be partially explained by the removal of 17% of the sequences not classified at the species level for analysis; and, most probably, due to the different and roughly strict pipeline we employed to analyze the 16S rRNA gene high-throughput sequencing data.

While previous pipelines generate high numbers of OTUs (many of them considered spurious as the result of low quality filtering processes) usually with low taxonomic resolution and ambiguous taxon level identities [17, 18, 24, 35], our analysis resulted in 878 OTUs, where many of them belong to the same species. This means the hospital environment is predominantly inhabited by closely related taxa, a pattern quite different from other environments usually composed by high diversity even at high taxonomy levels [9, 11]. Such pattern also explain the results in the PCoA analysis, which indicated that the structure of the bacterial communities in hospital environments were similar.

In this sense, closely related taxa may play similar roles within the hospital, while their distributions may vary significantly in each environment. The closely related species *A. pitti* and *A. nosocomialis*, for example, may play a role similar to *A. baumannii*, but the relative distribution of these three Acb complex species seem to vary geographically [36]. Notably, we observed a high presence of several potential nosocomial pathogens, including *Acinetobacter baumannii, Acinetobacter nosocomialis, Bacillus cereus, Klebsiella oxytoca, Pseudomonas aeruginosa, Pseudomonas putida, Staphylococcus aureus, Staphylococcus epidermidis, Serratia marcescens*, among others. Some of these nosocomial species were also differentially abundant in the hospital facilities analyzed, which may give better clues on the preferential habitats of these particular species as well as their potential reservoirs in hospital environments. The presence of potential pathogens differentially abundant in ICU-B and ICU-A may be explained by the fact that these

facilities usually receive patients with severe illnesses, some of them associated with severe infections caused by these pathogens. Nevertheless, their presence in inanimate surfaces should be of primary concern and demonstrate that the cleaning routine of these environments should be reviewed.

By employing network analyses, we described the complex pattern of inter-relationships between bacterial taxa co-occurring in the hospital environment. Only correlations with $r \geq \pm 0.9$ ($p \leq 0.05$) were used to generate the hospital network. Such strict cutoff increases the confidence of our analysis for detecting only strong interactions, which ensures that strongly non-random distribution patterns are mostly due to ecological reasons. Positive correlations suggest the occurrence of a mutualistic interaction while negative correlations suggest the presence of direct or indirect competition between the bacterial taxa co-occurring in the hospital environment.

The results indicated that the hospital microbial network presents a unique co-occurrence pattern by forming a low connected and clustered topology, but not structured among groups of nodes (i.e., modules) as usually found in most natural environments [37, 38]. These structural properties offer the potential for comparisons among different health care ecosystems in order to explore how the general traits of a given hospital may influence the assembly of microbial communities. It also helps us understand which organisms are most important in maintaining the structure and interactions of microbial communities in hospital facilities. In this sense, the identification of potential keystone species is of primary relevance. A keystone species is a species that plays a critical role in maintaining the structure of an ecological community and whose impact on the community is greater than would be expected based on its relative abundance or total biomass [39]. In this study, the two most suitable parameters (defined by Rampelotto et al., 2014 [40]) identified only a few potential keystone taxa, which suggests that the hospital microbial network is evenly distributed.

A novel and important strength of our co-occurrence analysis is the ability to detect ecologically relevant relationships between specific microbial taxa, in especial, potential competition between pathogens and non-pathogens. To date, a limited number of studies addressing the events associated with microbial competition in a spatial and multiplexed fashion have been performed, due in part to the lack of available tools. In this sense, we hypothesize that this strategy could be used as a theoretical framework to identify potentially strong negative correlation between pathogens and non-pathogens, which in turn could guide more focused and experimental studies to screen for bioactive compounds

against pathogenic bacteria derived from any non-pathogenic microbe that possesses a competitive advantage. As a proof of concept, by using *in vitro* co-cultivation, Gonzalez et al. 2011 demonstrated that the non-pathogenic *Bacillus subtilis*, a bacterium that is nearly ubiquitous in nature, was able to inhibit the growth of an epidemic *Staphylococcus aureus* isolate and possessed the ability to directionally release a molecule with antimicrobial and metabolism-altering properties [41]. In another interesting case, a recent *in vitro* study provided the first evidence that the harmless bacteria *Corynebacterium accolens*, which commonly colonizes the nose, can help inhibit *Streptococcus pneumoniae* through a direct antagonistic interaction between these species [42].

In our study, we have identified different taxa presenting strong negative co-occurrence with a variety of nosocomial and rare nosocomial pathogens. These taxa could be the focus of future *in vitro* co-culture experiments exploring the underlying mechanisms of antagonistic interactions between commensal and pathogenic bacteria to address how one species prevent the growing of another and to identify which components are involved in such interactions.

Another promising application would be the direct use of the non-pathogenic species or genetically engineered harmless variants of rare opportunistic pathogens as microbial-based sanitizing agents to reduce and control the colonization of nosocomial pathogens. This new concept, originally suggested by Falagas and Makris [43], has already successfully been applied in recent years as an alternative method to chemical disinfectants [44, 46]. The rational design and use of probiotic bacteria and biosurfactants for nosocomial infection control may overcome the problems associated with the chemical germicides, which present risks towards the environment and the patient's safety [47]. Several studies have demonstrated that more than 50% of hospital room surfaces are inadequately cleaned and disinfected when conventional chemical disinfectants are used. In addition, disinfectants can select resistant bacterial strains against the own disinfectant and also against antibiotics [48], which has been recently reported for chlorhexidine induction of resistance against Colistin [49], an antibiotic considered, until 2016, as a last-resort drug for treatment of infections sustained by multidrug-resistant (MDR) Gram-negative bacteria.

This emerging concept of microbial remediation for the prevention and control of hospital-acquired infections is a paradigm shift in the field, in which, instead of eradicating all pathogens, replacing pathogens by beneficial microbes might be more effective in decreasing infections [50-52].

Although 16S rRNA gene sequencing as a clinical screening tool has many advantages over traditional culture-based techniques, it is important to ponderate about its limitations. As any amplification based on rRNA gene, it only analyzes a short, specific genomic region, and taxonomic resolution or functional inference may be limited, especially for closely related species (i.e. sharing > 99% similarity in their 16S rRNA gene sequence) [53-55]. As an example, *Clostridium botulinum* and *Clostridium sporogenes* exhibit a 99.7% similarity [56, 57]. Another example may be found in the genus *Rickettsia* in which 16S rRNA gene sequence similarity values > 99% are found among all 26 species that have names with standing in nomenclature [58]. In general, 2.4% of complete sequenced genomes have 16S rRNA sequences with < 99% mean similarity [59]. Another major limitation of the 16S rRNA sequence is its inability to discriminate among virulent strains, which means it is not possible to distinguish pathogenic *Clostridium difficile* or *Escherichia coli* strains from nonpathogenic strains. Thus, for accurate identification of certain bacterial species and virulent strains, further methods such as multiplex PCR assay, mass spectrometry or whole genome sequencing must be applied.

Despite these limitations, the framework provided in this study for detection of multiple clinically relevant microbial targets is a promising addition to current diagnostic techniques and can play an important role in routine healthcare-associated infections surveillance.

Conclusion

The results of our investigation provide new insights into different aspects of the hospital microbiome and indicate that the high-throughput sequencing of the 16S rRNA gene can be used as a robust first-step tool for microbiological identification and characterization of a wide range of common bacterial pathogens in hospital settings with high resolution. Through the use of a well-annotated database of 16S rRNA sequences and the use of a rigorous filtering process, we demonstrate that high-resolution profiling of bacterial communities can be achieved and conclude that the framework developed in this study may be an integral part of routine diagnostic testing for hospital surveillance and infection control. Further improvements on the framework can make this technology an even more user-friendly tool in the routine of hospitals to control, and more important, prevent hospital outbreaks. This approach also shows potential to clinical application in

infection diseases diagnostics, an area in which 16S rRNA gene sequence identification might have an immediate and direct impact on patient care.

Conflicts of Interest: The authors declare no conflict of interest.

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6. DISCUSSÃO GERAL

Considerando que o espectro de microrganismos que causam infecções nosocomiais é uma ameaça constante e que o surgimento de novos patógenos relacionados a surtos graves está aumentando a cada dia, um dos grandes desafios na vigilância da saúde pública é a identificação rápida e precisa de agentes infecciosos. A rápida caracterização dos inúmeros táxons microbianos é essencial para a identificação de patógenos, fontes de infecções e potenciais vias de transmissão, bem como desempenha um importante papel na descontaminação de ambientes infectados e na prevenção de surtos. No caso de IRAS, a maioria dos países não possui sistemas de vigilância, e aqueles que têm sistemas, muitas vezes lutam com a complexidade e falta de critérios padronizados para diagnosticar as infecções (WHO, 2011).

Apesar dos avanços tecnológicos, os atuais métodos clínicos de diagnóstico microbiológico e monitoramento ambiental em um hospital baseiam-se, principalmente, em técnicas convencionais baseadas em cultura (Manaka et al., 2017). Estes métodos tradicionais requerem meios de cultura apropriados e condições de cultura adequadas às bactérias alvo. Além disso, algumas bactérias requerem um tempo prolongado de isolamento e cultura, seguido de testes de suscetibilidade e caracterização adicional caso a caso (Fenollar e Raoult, 2004). Dependendo do patógeno, esse procedimento pode levar dias para a cultura, dias adicionais para a identificação de espécies e testes de

suscetibilidade e semanas para uma tipagem molecular (Köser et al., 2012). Esses desafios clínicos resultaram em práticas de limpeza e prevenção ineficientes e na administração inadequada de antibióticos, o que pode explicar, em parte, as altas taxas de IRAS e o desenvolvimento relativamente recente de bactérias multirresistentes em hospitais e outros estabelecimentos de saúde em todo o mundo.

Vários métodos para testes rápidos de diagnóstico diretamente com amostras clínicas foram desenvolvidos e avaliados, incluindo PCR e MALDI-TOF MS (Burillo et al., 2014; Maurer et al., 2017; Patel et al., 2017). No entanto, estes métodos ainda são limitados, no sentido de que são dependentes da cultura, apenas fornecem informações sobre um número limitado de táxons e não podem ser facilmente comparados entre diferentes laboratórios.

Por outro lado, as técnicas de sequenciamento de nova geração (NGS) são independentes da cultura e podem ser extremamente úteis para a identificação de táxons raros ou mesmo desconhecidos, com grande aplicação na microbiologia clínica e na prevenção de infecções (Deurenberg et al., 2017). Além disso, NGS pode simultaneamente detectar e quantificar um número teoricamente ilimitado de bactérias em múltiplas amostras ao mesmo tempo, o que elimina a necessidade de seleção de alvos clínicos específicos para cultura e, também, cria uma oportunidade para observar o estado geral do microbioma. Isso é de interesse clínico particular, já que, concomitante ao advento das técnicas de NGS, o microbioma emergiu como um dos principais fatores relacionados à saúde e a doenças.

Apesar das vantagens do sequenciamento de todo o genoma (Hasman et al., 2014; Kwong et al., 2015; Quainoo et al., 2017), a análise de grandes conjuntos de dados requer uma combinação de habilidades de bioinformática e recursos computacionais que, atualmente, é praticamente inexistente em laboratórios microbiológicos de diagnóstico e instituições de saúde. Além disso, as abordagens de sequenciamento do genoma inteiro

consomem tempo, pois o tempo necessário para analisar um conjunto de dados metagenômicos por um profissional é de aproximadamente quatro a cinco dias.

Para preencher a lacuna entre os métodos convencionais e o sequenciamento de todo o genoma, uma abordagem envolvendo sequenciamento direcionado, o qual foca em genes ou regiões do genoma específicas, parece ser uma excelente abordagem para detectar e identificar táxons bacterianos. Em comparação com o sequenciamento completo do genoma, o sequenciamento direcionado é mais rápido e mais barato, menos complicado e, portanto, mais provável de ser implementado em laboratórios de diagnóstico dentro de um curto período de tempo (Clarridge, 2004). A sequência do gene 16S rRNA provou ser um marcador genético confiável para sequenciamento direcionado, pois está presente em todas as bactérias e sua função não mudou ao longo do tempo. Além disso, tem regiões de sequência conservada e variável evoluindo em taxas muito diferentes, críticas para a amplificação universal simultânea e medição de relações filogenéticas (Woo et al., 2008). Essas características permitem o uso do gene 16S rRNA para caracterização microbiana diretamente de amostras biológicas, fornecendo informações aprimoradas sobre a presença de DNA microbiano dentro de um período clinicamente relevante (Schlaberg et al., 2012; Srinivasan et al., 2015). Isso é necessário para o tratamento rápido e preciso de infecções e a identificação imediata de surtos, o que é fundamental para programas eficazes de vigilância de controle de infecções hospitalares.

Embora o sequenciamento do gene 16S rRNA possa ter grande impacto na microbiologia clínica e no diagnóstico de doenças infecciosas, a correta identificação das espécies bacterianas continua sendo um dos problemas mais difíceis enfrentados pelos microbiologistas clínicos. Existe a falta de um valor de limiar universal para a atribuição de espécies, uma vez que são observados diferentes níveis de diversidade de sequências entre diferentes táxons bacterianos, que evoluem a taxas diferentes. Embora um nível de

similaridade de 97% tenha sido proposto e amplamente utilizado para especiação bacteriana, uma diferença menor que 0,5% pode ser indicativo de uma nova espécie. Foi relatado que para aproximadamente 42% dos gêneros bacterianos, haverá pares de sequências dentro do gênero que não podem ser facilmente distinguidas porque suas sequências do gene 16S rRNA apresentam mais de 97% de similaridade (Vetrovsky e Baldrian, 2013). Além disso, os bancos de dados disponíveis publicamente para análise de sequências 16S rRNA estão associadas a problemas que os tornam não ideais para estudos clínicos, principalmente devido à falta de acurácia das sequências depositadas (Beiko, 2016; Balvočiūtė e Huson, 2017). Por isso, neste trabalho foi usado um limiar de 99% de similaridade contra um banco de dados curado, o que garantiu alta acurácia taxonômica dos táxons identificados.

Outro aspecto importante do nosso método foi a escolha dos *primers*. Esta escolha é um dos fatores limitantes mais críticos que afetam a análise de 16S rRNA, pois a seleção de *primers* inapropriados pode comprometer a interpretação dos resultados. De fato, vários estudos recentes demonstram que a maioria dos *primers* comuns usados em trabalhos anteriores apresentam eficiências de amplificação diferencial dos fragmentos de 16S rRNA (Morales e Holben, 2009). Mesmo quando as diversidades microbianas são medidas na mesma amostra, os resultados podem diferir de forma significativa dependendo da escolha dos *primers* (Claesson et al., 2010). Esse problema ocorre porque as diferentes regiões hipervariáveis do rRNA 16S evoluem em taxas diferentes e espécies diferentes do mesmo gênero podem ser semelhantes em algumas dessas regiões e mais divergentes em outras. Por esta razão, qualquer combinação de *primers* provavelmente perderá alguns grupos bacterianos ou, pelo menos, subestimará a abundância de alguns desses táxons.

Originalmente descritos em 2011, os *primers* 515F e 806R (amplicon de 291 bp), que amplificam a região V4 do 16S rRNA, foram selecionados pelo Earth Microbiome

Project como padrão para genomas de procariontes (Caporaso et al., 2011). Desde então, este par de *primers* tem sido amplamente utilizado para caracterizar a composição de comunidades microbianas em diferentes ambientes (Thompson et al., 2017). No entanto, devido ao pequeno fragmento gerado, a maioria das sequências não pode ser classificada ao nível da espécie. Para melhorar a resolução taxonômica, em nossa análise foram utilizados os *primers* 341F e 806R, que geraram um amplicon de 465 pb flanqueando a região V3-V4 do gene RNAr 16S. Este conjunto de *primers* fornece informação molecular para a classificação taxonômica de comunidades microbianas de táxons associados ao microbioma humano (Fadrosh et al., 2014). Portanto, os dados gerados usando o par de *primers* 341F e 806R, a partir do sequenciamento usando a plataforma MiSeq, podem ser comparados com os dados existentes na literatura do microbioma humano, especialmente para estudos envolvendo o microbioma de pele (Castelino et al, 2017), que são de grande relevância para amostras de superfícies de alto contato.

Em relação aos outros trabalhos envolvendo microbioma hospitalar, este projeto teve algumas diferenças significativas em termos de metodologia, principalmente em relação a quatro aspectos relevantes: número de amostras, região do gene 16S amplificada, banco de dados de referência, e *pipelines* robustos que removam OTUs espúrias.

Poza et al. (2012) analisaram 11 amostras hospitalares divididas em dois grupos (Hall de entrada e UTI), usando o conjunto de *primers* da região V7–V9 do gene rRNA 16S e o banco de dados Greengenes como referência taxonômica; o número total de OTUs em todas as amostras foi de 2499. Oberauner et al. (2013) analisaram 24 amostras de uma UTI neonatal, usando o conjunto de *primers* da região V3–V5 e o banco de dados RDP como referência taxonômica; o número total de OTUs em todas as amostras foi de 3925. Hewitt et al. (2013) analisaram 17 amostras de duas UTIs neonatal, usando o conjunto de *primers* da região V2 e o banco de dados Greengenes como referência taxonômica; o

número total de OTUs em todas as amostras foi de 1621. Tang et al. 2015 analisaram 16 amostras de um centro de cuidados respiratórios, usando o conjunto de *primers* da região V1–V2 e o banco de dados SILVA como referência taxonômica; o número total de OTUs em todas as amostras foi de 3503. Chen et al. 2017b analisaram 203 amostras de 4 hospitais, usando o conjunto de *primers* da região V1–V2 e o banco de dados SILVA como referência taxonômica; o número total de OTUs em todas as amostras foi de 8504. Em todos esses estudos, não foram aplicadas etapas de remoção de OTUs espúrias.

Assim, a maioria desses estudos se baseou em poucas amostras, geralmente relacionadas a uma unidade do hospital, e coletadas em um determinado momento. Apenas um desses estudos foi mais abrangente, baseado em 203 amostras coletadas em 4 hospitais de Janeiro a Dezembro (Chen et al. 2017b). Devido a esta variabilidade no número de amostras, *primers*, e bancos de referência, os resultados desses trabalhos muitas vezes são contrastantes. Mas, de modo geral, todos concluem que o microbioma hospitalar apresenta alta diversidade, uma consequência do número elevado de OTUs espúrias que não foram filtradas.

Considerando os altos níveis de robustez e resolução dos métodos aplicados neste estudo, acreditamos que os vieses destacados no parágrafo anterior foram minimizados e que nossos resultados foram consistentes. Com base nos resultados obtidos, foi possível responder às duas perguntas principais do projeto. A resposta à primeira pergunta, relacionada às diferenças entre as comunidades microbianas dos diferentes ambientes hospitalares, foi contrária à nossa hipótese inicial que previa uma heterogeneidade entre as unidades do hospital. Esta heterogeneidade era esperada com base nos trabalhos anteriores que indicavam diferenças significativas quando duas ou mais unidades do hospital eram comparadas; e também pelas próprias características únicas de operação de cada unidade. A ICU-A recebe pacientes em estado de saúde menos grave e a ICU-B recebe pacientes em

estado de saúde mais grave, com maior tempo de internação. A MU é utilizada pelo hospital como uma unidade de isolamento, que recebe quase que exclusivamente pacientes colonizados ou com infecção por bactérias multirresistentes. A IU recebe pacientes que estão aguardando por uma cirurgia eletiva ou pacientes pós-cirúrgicos. O SC recebe pacientes para cirurgia. A ECU é uma unidade apenas para atendimentos de urgência e emergência, com necessidade de encaminhamento para outras unidades caso o paciente necessite de internação. Além dessas características únicas de operação, cada unidade possui sua própria rotina de limpeza e esterilização, o que pode gerar pressões seletivas diferentes em cada ambiente.

Por outro lado, os resultados obtidos indicam que o microbioma hospitalar de modo geral é homogêneo (como observados nas análises de alfa e beta diversidade). O fato de estarmos analisando superfícies de alto contato, onde a transferência de microrganismos de um local para outro é mais intensa, é uma das explicações para a homogeneidade observada. Além disso, outro fator importante é a alta taxa de rotatividade dos pacientes entre as diversas unidades hospitalares (54,5% dos pacientes são internados em mais de uma unidade do hospital).

Essa característica homogênea das comunidades microbianas deu base para a resposta à segunda pergunta do projeto, isto é, se existe um padrão que caracteriza o microbioma hospitalar. Os resultados de alfa e beta diversidade, bem como os de composição e redes de co-ocorrência, corroboram com a hipótese de que o ambiente hospitalar apresenta um padrão característico, com uma estrutura homogênea, composta por uma dominância massiva de alguns táxons e formada por uma rede de co-ocorrência não-clusterizada. Semelhante a outras comunidades microbianas de ambientes internos (Gilbert e Stephens, 2018), estas comunidades foram parcialmente formadas por bactérias associadas ao ser humano. Embora seja impossível predizer a patogenicidade de uma cepa

com base nas sequências 16S rRNA, a proporção de táxons intimamente relacionados com patógenos humanos foi bastante elevada, como *Pseudomonas*, *Acinetobacter*, *Klebsiella*, *Staphylococcus*, *Enterobacter*, entre outras.

Essa homogeneidade oriunda de uma intensa transferência de microrganismos de um local para outro, associada à falta de um sistema de vigilância eficiente, reforça o problema da ampla disseminação de bactérias relacionadas às IRAS na instituição hospitalar. Algumas das áreas com maiores contaminações são classicamente reportadas pela literatura como críticas, como o entorno do paciente – o seu leito, principalmente. Também por meio do sequenciamento do 16S rRNA, Lax et al. (2017) demonstraram uma forte interação entre a pele dos pacientes e seus leitos e Hewitt et al. (2013) demonstraram a interação entre a microbiota humana e as contaminações encontradas em incubadoras de pacientes neonatos. Gaudart et al. (2013) também demonstraram uma alta contaminação de diversos itens dos leitos de pacientes como as grades da cama, a mesa de cabeceira, o carro de armazenamento de materiais e o dispensador de álcool em gel.

No entanto, o presente estudo incluiu, além das áreas adjacentes aos pacientes, áreas de trabalho e descanso dos profissionais da saúde, como a copa e as salas de repouso. Estes locais também apresentaram maior concentração de bactérias, muitas delas com alto potencial de patogenicidade. Esses achados podem ser explicados por possível negligência, em relação ao uso inadequado e falhas de higienização dessas áreas, especialmente aquelas destinadas aos intervalos de descanso dos profissionais da saúde. É importante ressaltar que todas as ações que permitam diminuir o risco de disseminação de bactérias potencialmente patogênicas são ações no sentido de garantir a segurança do paciente e dos próprios profissionais de saúde. Ou seja, as boas práticas de uso de todas as áreas que compõe as instituições de saúde, as boas práticas de higienização das mãos aplicadas a todos os locais, o uso racional de vestimentas exclusivas às unidades hospitalares e as boas práticas de

cuidado ao paciente, somente quando realizadas de forma linear e conjunta, poderão contar positivamente para a garantia da segurança de todos na instituição.

Embora foi demonstrado nesse trabalho que o sequenciamento do gene 16S rRNA pode ser usado como uma ferramenta de caracterização molecular do microbioma hospitalar, é importante ponderar sobre suas limitações. Há mais de uma década, pesquisadores encontram problemas de resolução ao nível de espécie para identificação bacteriana, incluindo principalmente a família Enterobacteriaceae (em particular Enterobacter e Pantoea), micobactérias de crescimento rápido, *Achromobacter*, *Stenotrophomonas* e *Actinomyces* (Janda e Abbott, 2007). Srinivasan et al. (2015) utilizaram um repositório de 617 isolados clínicos, com representantes de 30 espécies clinicamente importantes para avaliar a capacidade do sequenciamento do 16S rRNA na identificação bacteriana ao nível de espécie, utilizando para isso técnicas de *machine learning*, um algoritmo de classificação (Naive Bayes) e um conjunto diversificado de sequências de alta qualidade de bactérias de importância médica. Os resultados mostraram uma taxa de concordância ao nível do gênero de 96,0% e de 87,5% ao nível de espécie. Foram apontados diversos casos de identificação equivocada por métodos tradicionais baseados em cultivo, mas também limitações da identificação por 16S rRNA, em particular para espécies da família Enterobacteriaceae, provavelmente devido a uma combinação de imprecisões na base de dados e a própria variabilidade biológica, limitando o poder de resolução taxonômica do 16S rRNA. Os resultados demonstraram uma alta confiabilidade do 16S rRNA para a identificação ao nível de gênero e um bom nível de confiabilidade para o nível de espécie, com exceções de certos gêneros, como *Stenotrophomonas*, *Enterobacter*, *Citrobacter* e *Escherichia*. Segundo os autores, as dificuldades de identificação ao nível de espécie são devidas, provavelmente, a ambiguidades taxonômicas, sugerindo falta de poder discriminatório desse marcador. Os autores ainda ressaltam que

os debates taxonômicos das ambiguidades continuarão e provavelmente evoluirão à medida que surjam mais dados de identificação bacteriana baseados em sequenciamento de nova geração, porém, é certo que o sequenciamento de marcadores baseados em genes bacterianos continuará sendo uma medida válida e não subjetiva de identificação bacteriana (em comparação com as técnicas de cultivo e os ensaios bioquímicos), embora alguns desses marcadores possam proporcionar um poder discriminatório não-otimizado para grupos específicos de microrganismos.

Embora este método não possa determinar quais microrganismos são viáveis e metabolicamente ativos, demonstrou-se que ele pode ajudar a rastrear a disseminação de um táxon microbiano no ambiente hospitalar, o que é de grande importância na determinação do papel das superfícies e equipamentos hospitalares na transmissão de microrganismos. Além disso, a presença de DNA de células microbianas não-viáveis ou mortas pode ser relevante porque quase todas as bactérias são capazes de transferência horizontal de genes e transformação (Soucy et al., 2015). Ao integrar o DNA "morto" dos ambientes circundantes, incorporando em suas células e multiplicando, estas bactérias podem espalhar genes de virulência ou antimicrobianos em hospitais e outros ambientes, bem como se tornar cepas resistentes ou virulentas (Chen and Dubnau 2004; Ellison et al., 2018).

7. CONCLUSÃO

Os resultados da nossa investigação fornecem uma nova visão sobre diferentes aspectos do microbioma hospitalar e indicam que o sequenciamento do gene 16S rRNA pode ser usado como uma ferramenta robusta para a identificação e caracterização microbiológica de uma ampla gama de táxons bacterianos comuns ao ambiente hospitalar. Consequentemente, acreditamos que este trabalho possa desempenhar um papel importante na vigilância rotineira de infecções associadas aos cuidados de saúde.

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ANEXO A

CURRICULUM VITAE - Resumido

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Bolsista do(a): Coordenação de Aperfeiçoamento de Pessoal de Nível Superior, CAPES, Brasil.

2012 - 2014

Mestrado em Ciências Biológicas

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Bolsista do(a): Fundação de Amparo à Pesquisa do Estado do Rio Grande do Sul, FAPERGS, Brasil.

2009 - 2010

Graduação em Ciências Biológicas - Licenciatura

Universidade Federal de Santa Maria, UFSM, Brasil.

2005 - 2008

Graduação em Ciências Biológicas - Bacharelado

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PUBLICAÇÕES RELEVANTES NOS ÚLTIMOS ANOS:

Rampelotto, P.H. Molecular Mechanisms of Microbial Evolution. Basel: Springer, 2018.

Rampelotto, P.H. The Relevance and Challenges of Studying Microbial Evolution. In: Pabulo H Rampelotto. (Org.). Molecular Mechanisms of Microbial Evolution. Basel: Springer, 2018, p.1-11.

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ANEXO B

Material suplementar do Capítulo I.

Table S1. Library characterization. Month, Facility, Room, and Sample-Type represent the four categories which samples were grouped. (%). Abbreviations: ECU, Emergency Care Unit; MU, Medical Unit; SC, Surgery Center; IU, Inpatient Unit; ICU-A, Intensive Care Unit A; ICU-B, Intensive Care Unit B.

Sample Code	Sample Description	Sample-Type	Room	Facility	Month
150410886613.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	IU	04 - April
150410886614.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	04 - April
150410886615.1.1	Bed railings from Patient Room A	bed railings	Patient room	IU	04 - April
150410886616.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	04 - April
150410886617.1.1	Bed railings from Patient Room B	bed railings	Patient room	IU	04 - April
150410886618.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	04 - April
150410886619.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	04 - April
150410886620.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	04 - April
150410886621.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	04 - April
150410886622.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	04 - April
150410886623.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	04 - April
150410886624.1.1	Bed, table and chair	bed/table/chair	Resting room	IU	04 - April

150410886625.1.1	Dispenser from the Hall	dispenser	Common place	IU	04 - April
150410886626.1.1	Glicosimeter	medical device	Common place	IU	04 - April
150410886641.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	04 - April
150410886642.1.1	Bed railings from Patient Room A	bed railings	Patient room	MU	04 - April
150410886643.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	04 - April
150410886644.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	04 - April
150410886645.1.1	Bed railings from Patient Room B	bed railings	Patient room	MU	04 - April
150410886646.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	04 - April
150410886647.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	04 - April
150410886648.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	04 - April
150410886649.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	04 - April
150410886650.1.1	Cabinets and chairs	cabinets/chair	Locker room	MU	04 - April
150410886651.1.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	04 - April
150410886652.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	04 - April
150410886653.1.1	Bed, table and chair	bed/table/chair	Resting room	MU	04 - April
150410886654.1.1	Dispenser from the Hall	dispenser	Common place	MU	04 - April
150410886655.1.1	Glicosimeter	medical device	Common place	MU	04 - April
150410886664.1.1	Bed railings	bed railings	Recovery room	SC	04 - April
150410886665.1.1	Serum and gas support	serum/gas support	Recovery room	SC	04 - April
150410886666.1.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	04 - April
150410886667.1.1	Surgical table from Room 1	bed	Surgery room	SC	04 - April
150410886668.1.1	Oxigen buttons	surgery device	Surgery room	SC	04 - April
150410886669.1.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	04 - April
150410886670.1.1	Anesthesia syringes	surgery device	Surgery room	SC	04 - April
150410886671.1.1	Surgical table from Room 2	bed	Surgery room	SC	04 - April
150410886672.1.1	Surgical light	surgery device	Surgery room	SC	04 - April
150410886673.1.1	Syringes	surgery device	Surgery room	SC	04 - April

150410886674.1.1	Electric scalpel	surgery device	Surgery room	SC	04 - April
150410886675.1.1	Cabinets and chairs	cabinets/chair	Locker room	SC	04 - April
150410886676.1.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	04 - April
150410886677.1.1	Bed, table and chair	bed/table/chair	Resting room	SC	04 - April
150410886678.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	04 - April
150410886679.1.1	Computer	equipment of common use	Nurse chief room	SC	04 - April
150410886680.1.1	Stretcher from the Hall	bed	Common place	SC	04 - April
150410886681.1.1	Dispenser from the Hall	dispenser	Common place	SC	04 - April
150410886696.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	04 - April
150410886697.1.1	Infusion pumps	medical device	Patient room	ICU-A	04 - April
150410886698.1.1	Screen monitors	medical device	Patient room	ICU-A	04 - April
150410886699.1.1	Curtain	wall/curtain	Patient room	ICU-A	04 - April
150410886700.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	04 - April
150410886701.1.1	Utensils table	table	Patient room	ICU-A	04 - April
150410886702.1.1	Wall	wall/curtain	Patient room	ICU-A	04 - April
150410886703.1.1	Termometer	medical device	Patient room	ICU-A	04 - April
150410886704.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	04 - April
150410886705.1.1	Clipboards	equipment of common use	Nurse station	ICU-A	04 - April
150410886706.1.1	Computers	equipment of common use	Prescription room	ICU-A	04 - April
150410886707.1.1	SEquipMateriais - EquipLimpos	medical device	Medical device room	ICU-A	04 - April
150410886708.1.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	04 - April
150410886709.1.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	04 - April
150410886710.1.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	04 - April
150410886711.1.1	Dryer	equipment of common use	Purge room	ICU-A	04 - April
150410886712.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	04 - April
150410886713.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	04 - April
150410886714.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	04 - April
150410886727.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	04 - April

150410886728.1.1	Infusion pumps	medical device	Patient room	ICU-B	04 - April
150410886729.1.1	Screen monitors	medical device	Patient room	ICU-B	04 - April
150410886730.1.1	Curtain	wall/curtain	Patient room	ICU-B	04 - April
150410886731.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	04 - April
150410886732.1.1	Utensils table	table	Patient room	ICU-B	04 - April
150410886733.1.1	Wall	wall/curtain	Patient room	ICU-B	04 - April
150410886734.1.1	Termometer	medical device	Patient room	ICU-B	04 - April
150410886735.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	04 - April
150410886736.1.1	Refrigerator	equipment of common use	Nurse station	ICU-B	04 - April
150410886737.1.1	Computers	equipment of common use	Prescription room	ICU-B	04 - April
150410886738.1.1	Dispenser from the Hall	dispenser	Common place	ICU-B	04 - April
150410886739.1.1	Utensils from the Bath cart	medical device	Common place	ICU-B	04 - April
150410886740.1.1	Glicosimeter	medical device	Common place	ICU-B	04 - April
150410886741.1.1	Cufometer	medical device	Common place	ICU-B	04 - April
150410886742.1.1	Bath cart	medical device	Common place	ICU-B	04 - April
150410886743.1.1	Emergency cart	medical device	Common place	ICU-B	04 - April
150410886744.1.1	X-Raio cart	medical device	Common place	ICU-B	04 - April
150410886745.1.1	Ultrasound cart	medical device	Common place	ICU-B	04 - April
150410886746.1.1	Passant	medical device	Common place	ICU-B	04 - April
150410886747.1.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	04 - April
150410886765.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	04 - April
150410886766.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	04 - April
150410886767.1.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	04 - April
150410886768.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	04 - April
150410886769.1.1	Bed railings from Patient Room C	bed railings	Patient room	ECU	04 - April
150410886770.1.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	04 - April
150410886771.1.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	04 - April

150410886772.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	04 - April
150410886773.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	04 - April
150410886774.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	04 - April
150410886775.1.1	Chairs	chair	Medication room	ECU	04 - April
150410886776.1.1	Serum and gas support	serum/gas support	Medication room	ECU	04 - April
150410886777.1.1	Computers	equipment of common use	Prescription room	ECU	04 - April
150410886778.1.1	Sink	sink	Special procedures room	ECU	04 - April
150410886779.1.1	SProcedEspeciais - Leito	bed	Special procedures room	ECU	04 - April
150410886780.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	04 - April
150410886781.1.1	Bed	bed	Clinic room	ECU	04 - April
150410886782.1.1	Computer table	table	Clinic room	ECU	04 - April
150410886783.1.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	04 - April
150410886784.1.1	Chairs	chair	Reception	ECU	04 - April
150410886785.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	04 - April
150410886786.1.1	Bed, table and chair	bed/table/chair	Resting room	ECU	04 - April
150410886787.1.1	Dispenser from the Hall	dispenser	Common place	ECU	04 - April
150410886788.1.1	Medication cart	medical device	Common place	ECU	04 - April
150501772314.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	IU	05 - May
150501772315.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	05 - May
150501772316.1.1	Bed railings from Patient Room A	bed railings	Patient room	IU	05 - May
150501772317.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	05 - May
150501772318.1.1	Bed railings from Patient Room B	bed railings	Patient room	IU	05 - May
150501772319.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	05 - May
150501772320.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	05 - May
150501772321.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	05 - May
150501772322.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	05 - May
150501772323.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	05 - May

150501772324.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	05 - May
150501772325.1.1	Bed, table and chair	bed/table/chair	Resting room	IU	05 - May
150501772326.1.1	Dispenser from the Hall	dispenser	Common place	IU	05 - May
150501772327.1.1	Glicosimeter	medical device	Common place	IU	05 - May
150501772342.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	05 - May
150501772343.1.1	Bed railings from Patient Room A	bed railings	Patient room	MU	05 - May
150501772344.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	05 - May
150501772345.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	05 - May
150501772346.1.1	Bed railings from Patient Room B	bed railings	Patient room	MU	05 - May
150501772347.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	05 - May
150501772348.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	05 - May
150501772349.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	05 - May
150501772350.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	05 - May
150501772351.1.1	Cabinets and chairs	cabinets/chair	Locker room	MU	05 - May
150501772352.1.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	05 - May
150501772353.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	05 - May
150501772354.1.1	Bed, table and chair	bed/table/chair	Resting room	MU	05 - May
150501772355.1.1	Dispenser from the Hall	dispenser	Common place	MU	05 - May
150501772356.1.1	Glicosimeter	medical device	Common place	MU	05 - May
150501772365.1.1	Bed railings	bed railings	Recovery room	SC	05 - May
150501772366.1.1	Serum and gas support	serum/gas support	Recovery room	SC	05 - May
150501772367.1.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	05 - May
150501772368.1.1	Surgical table from Room 1	bed	Surgery room	SC	05 - May
150501772369.1.1	Oxigen buttons	surgery device	Surgery room	SC	05 - May
150501772370.1.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	05 - May
150501772371.1.1	Anesthesia syringes	surgery device	Surgery room	SC	05 - May

150501772372.1.1	Surgical table from Room 2	bed	Surgery room	SC	05 - May
150501772373.1.1	Surgical light	surgery device	Surgery room	SC	05 - May
150501772374.1.1	Syringes	surgery device	Surgery room	SC	05 - May
150501772375.1.1	Electric scalpel	surgery device	Surgery room	SC	05 - May
150501772376.1.1	Cabinets and chairs	cabinets/chair	Locker room	SC	05 - May
150501772377.1.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	05 - May
150501772378.1.1	Bed, table and chair	bed/table/chair	Resting room	SC	05 - May
150501772379.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	05 - May
150501772380.1.1	Computer	equipment of common use	Nurse chief room	SC	05 - May
150501772381.1.1	Stretcher from the Hall	bed	Common place	SC	05 - May
150501772382.1.1	Dispenser from the Hall	dispenser	Common place	SC	05 - May
150501772397.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	05 - May
150501772398.1.1	Infusion pumps	medical device	Patient room	ICU-A	05 - May
150501772399.1.1	Screen monitors	medical device	Patient room	ICU-A	05 - May
150501772400.1.1	Curtain	wall/curtain	Patient room	ICU-A	05 - May
150501772401.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	05 - May
150501772402.1.1	Utensils table	table	Patient room	ICU-A	05 - May
150501772403.1.1	Wall	wall/curtain	Patient room	ICU-A	05 - May
150501772404.1.1	Termometer	medical device	Patient room	ICU-A	05 - May
150501772405.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	05 - May
150501772406.1.1	Clipboards	equipment of common use	Nurse station	ICU-A	05 - May
150501772407.1.1	Computers	equipment of common use	Prescription room	ICU-A	05 - May
150501772408.1.1	SEquipMateriais - EquipLimpos	medical device	Medical device room	ICU-A	05 - May
150501772409.1.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	05 - May
150501772410.1.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	05 - May
150501772411.1.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	05 - May
150501772412.1.1	Dryer	equipment of common use	Purge room	ICU-A	05 - May
150501772413.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	05 - May

150501772414.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	05 - May
150501772415.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	05 - May
150501772428.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	05 - May
150501772429.1.1	Infusion pumps	medical device	Patient room	ICU-B	05 - May
150501772430.1.1	Screen monitors	medical device	Patient room	ICU-B	05 - May
150501772431.1.1	Curtain	wall/curtain	Patient room	ICU-B	05 - May
150501772432.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	05 - May
150501772433.1.1	Utensils table	table	Patient room	ICU-B	05 - May
150501772434.1.1	Wall	wall/curtain	Patient room	ICU-B	05 - May
150501772435.1.1	Termometer	medical device	Patient room	ICU-B	05 - May
150501772436.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	05 - May
150501772437.1.1	Refrigerator	equipment of common use	Nurse station	ICU-B	05 - May
150501772438.1.1	Computers	equipment of common use	Prescription room	ICU-B	05 - May
150501772439.1.1	Dispenser from the Hall	dispenser	Common place	ICU-B	05 - May
150501772440.1.1	Utensils from the Bath cart	medical device	Common place	ICU-B	05 - May
150501772441.1.1	Glicosimeter	medical device	Common place	ICU-B	05 - May
150501772442.1.1	Cufometer	medical device	Common place	ICU-B	05 - May
150501772443.1.1	Bath cart	medical device	Common place	ICU-B	05 - May
150501772444.1.1	Emergency cart	medical device	Common place	ICU-B	05 - May
150501772445.1.1	X-Raio cart	medical device	Common place	ICU-B	05 - May
150501772446.1.1	Ultrasound cart	medical device	Common place	ICU-B	05 - May
150501772447.1.1	Passant	medical device	Common place	ICU-B	05 - May
150501772448.1.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	05 - May
150501772466.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	05 - May
150501772467.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	05 - May
150501772468.1.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	05 - May
150501772469.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	05 - May
150501772470.1.1	Bed railings from Patient Room C	bed railings	Patient room	ECU	05 - May

150501772471.1.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	05 - May
150501772472.1.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	05 - May
150501772473.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	05 - May
150501772474.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	05 - May
150501772475.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	05 - May
150501772476.1.1	Chairs	chair	Medication room	ECU	05 - May
150501772477.1.1	Serum and gas support	serum/gas support	Medication room	ECU	05 - May
150501772478.1.1	Computers	equipment of common use	Prescription room	ECU	05 - May
150501772479.1.1	Sink	sink	Special procedures room	ECU	05 - May
150501772480.1.1	Bed	bed	Special procedures room	ECU	05 - May
150501772481.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	05 - May
150501772482.1.1	Bed	bed	Clinic room	ECU	05 - May
150501772483.1.1	Computer table	table	Clinic room	ECU	05 - May
150501772484.1.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	05 - May
150501772485.1.1	Chairs	chair	Reception	ECU	05 - May
150501772486.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	05 - May
150501772487.1.1	Bed, table and chair	bed/table/chair	Resting room	ECU	05 - May
150501772488.1.1	Dispenser from the Hall	dispenser	Common place	ECU	05 - May
150501772489.1.1	Medication cart	medical device	Common place	ECU	05 - May
150603610414.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	IU	06 - June
150603610415.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	06 - June
150603610416.1.1	Bed railings from Patient Room A	bed railings	Patient room	IU	06 - June
150603610417.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	06 - June
150603610418.1.1	Bed railings from Patient Room B	bed railings	Patient room	IU	06 - June
150603610419.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	06 - June

150603610420.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	06 - June
150603610421.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	06 - June
150603610422.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	06 - June
150603610423.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	06 - June
150603610424.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	06 - June
150603610425.1.1	Bed, table and chair	bed/table/chair	Resting room	IU	06 - June
150603610426.1.1	Dispenser from the Hall	dispenser	Common place	IU	06 - June
150603610427.1.1	Glicosimeter	medical device	Common place	IU	06 - June
150603610442.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	06 - June
150603610443.1.1	Bed railings from Patient Room B	bed railings	Patient Room	MU	06 - June
150603610444.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	06 - June
150603610445.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	06 - June
150603610446.1.1	Bed railings from Patient Room A	bed railings	Patient room	MU	06 - June
150603610447.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	06 - June
150603610448.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	06 - June
150603610449.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	06 - June
150603610450.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	06 - June
150603610451.1.1	Cabinets and chairs	cabinets/chair	Locker room	MU	06 - June
150603610452.1.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	06 - June
150603610453.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	06 - June
150603610454.1.1	Bed, table and chair	bed/table/chair	Resting room	MU	06 - June
150603610455.1.1	Dispenser from the Hall	dispenser	Common place	MU	06 - June
150603610456.1.1	Glicosimeter	medical device	Common place	MU	06 - June
150603610465.1.1	Bed railings	bed railings	Recovery room	SC	06 - June
150603610466.1.1	Serum and gas support	serum/gas support	Recovery room	SC	06 - June
150603610467.1.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	06 - June

150603610468.1.1	Surgical table from Room 1	bed	Surgery room	SC	06 - June
150603610469.1.1	Oxigen buttons	surgery device	Surgery room	SC	06 - June
150603610470.1.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	06 - June
150603610471.1.1	Anesthesia syringes	surgery device	Surgery room	SC	06 - June
150603610472.1.1	Surgical table from Room 2	bed	Surgery room	SC	06 - June
150603610473.1.1	Surgical light	surgery device	Surgery room	SC	06 - June
150603610474.1.1	Syringes	surgery device	Surgery room	SC	06 - June
150603610475.1.1	Electric scalpel	surgery device	Surgery room	SC	06 - June
150603610476.1.1	Cabinets and chairs	cabinets/chair	Locker room	SC	06 - June
150603610477.1.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	06 - June
150603610478.1.1	Bed, table and chair	bed/table/chair	Resting room	SC	06 - June
150603610479.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	06 - June
150603610480.1.1	Computer	equipment of common use	Nurse chief room	SC	06 - June
150603610481.1.1	Stretcher from the Hall	bed	Common place	SC	06 - June
150603610482.1.1	Dispenser from the Hall	dispenser	Common place	SC	06 - June
150603610497.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	06 - June
150603610498.1.1	Infusion pumps	medical device	Patient room	ICU-A	06 - June
150603610499.1.1	Screen monitors	medical device	Patient room	ICU-A	06 - June
150603610500.1.1	Curtain	wall/curtain	Patient room	ICU-A	06 - June
150603610501.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	06 - June
150603610502.1.1	Utensils table	table	Patient room	ICU-A	06 - June
150603610503.1.1	Wall	wall/curtain	Patient room	ICU-A	06 - June
150603610504.1.1	Termometer	medical device	Patient room	ICU-A	06 - June
150603610505.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	06 - June
150603610506.1.1	Clipboards	equipment of common use	Nurse station	ICU-A	06 - June
150603610507.1.1	Computers	equipment of common use	Prescription room	ICU-A	06 - June
150603610508.1.1	SEquipMateriais - EquipLimpos	medical device	Medical device room	ICU-A	06 - June
150603610509.1.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	06 - June

150603610510.1.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	06 - June
150603610511.1.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	06 - June
150603610512.1.1	Dryer	equipment of common use	Purge room	ICU-A	06 - June
150603610513.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	06 - June
150603610514.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	06 - June
150603610515.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	06 - June
150603610528.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	06 - June
150603610529.1.1	Infusion pumps	medical device	Patient room	ICU-B	06 - June
150603610530.1.1	Screen monitors	medical device	Patient room	ICU-B	06 - June
150603610531.1.1	Curtain	wall/curtain	Patient room	ICU-B	06 - June
150603610532.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	06 - June
150603610533.1.1	Utensils table	table	Patient room	ICU-B	06 - June
150603610534.1.1	Wall	wall/curtain	Patient room	ICU-B	06 - June
150603610535.1.1	Termometer	medical device	Patient room	ICU-B	06 - June
150603610536.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	06 - June
150603610537.1.1	Refrigerator	equipment of common use	Nurse station	ICU-B	06 - June
150603610538.1.1	Computers	equipment of common use	Prescription room	ICU-B	06 - June
150603610539.1.1	Dispenser from the Hall	dispenser	Common place	ICU-B	06 - June
150603610540.1.1	Utensils from the Bath cart	medical device	Common place	ICU-B	06 - June
150603610541.1.1	Glicosimeter	medical device	Common place	ICU-B	06 - June
150603610542.1.1	Cufometer	medical device	Common place	ICU-B	06 - June
150603610543.1.1	Bath cart	medical device	Common place	ICU-B	06 - June
150603610544.1.1	Emergency cart	medical device	Common place	ICU-B	06 - June
150603610545.1.1	X-Raio cart	medical device	Common place	ICU-B	06 - June
150603610546.1.1	Ultrasound cart	medical device	Common place	ICU-B	06 - June
150603610547.1.1	Passant	medical device	Common place	ICU-B	06 - June
150603610548.1.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	06 - June
150603610566.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	06 - June

150603610567.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	06 - June
150603610568.1.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	06 - June
150603610569.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	06 - June
150603610570.1.1	Bed railings from Patient Room C	bed railings	Patient room	ECU	06 - June
150603610571.1.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	06 - June
150603610572.1.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	06 - June
150603610573.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	06 - June
150603610574.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	06 - June
150603610575.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	06 - June
150603610576.1.1	Chairs	chair	Medication room	ECU	06 - June
150603610577.1.1	Serum and gas support	serum/gas support	Medication room	ECU	06 - June
150603610578.1.1	Computers	equipment of common use	Prescription room	ECU	06 - June
150603610579.1.1	Sink	sink	Special procedures room	ECU	06 - June
150603610580.1.1	Bed	bed	Special procedures room	ECU	06 - June
150603610581.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	06 - June
150603610582.1.1	Bed	bed	Clinic room	ECU	06 - June
150603610583.1.1	Computer table	table	Clinic room	ECU	06 - June
150603610584.1.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	06 - June
150603610585.1.1	Chairs	chair	Reception	ECU	06 - June
150603610586.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	06 - June
150603610587.1.1	Bed, table and chair	bed/table/chair	Resting room	ECU	06 - June
150701108514.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	IU	07 - July
150701108515.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	07 - July
150701108516.1.1	Bed railings from Patient Room A	bed railings	Patient room	IU	07 - July
150701108517.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	07 - July

150701108518.1.1	Bed railings from Patient Room B	bed railings	Patient room	IU	07 - July
150701108519.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	07 - July
150701108520.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	07 - July
150701108521.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	07 - July
150701108522.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	07 - July
150701108523.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	07 - July
150701108524.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	07 - July
150701108525.1.1	Bed, table and chair	bed/table/chair	Resting room	IU	07 - July
150701108526.1.1	Dispenser from the Hall	dispenser	Common place	IU	07 - July
150701108527.1.1	Glicosimeter	medical device	Common place	IU	07 - July
150701108542.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	07 - July
150701108543.1.1	Bed railings from Patient Room A	bed railings	Patient room	MU	07 - July
150701108544.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	07 - July
150701108545.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	07 - July
150701108546.1.1	Bed railings from Patient Room B	bed railings	Patient room	MU	07 - July
150701108547.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	07 - July
150701108548.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	07 - July
150701108549.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	07 - July
150701108550.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	07 - July
150701108551.1.1	Cabinets and chairs	cabinets/chair	Locker room	MU	07 - July
150701108552.1.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	07 - July
150701108553.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	07 - July
150701108554.1.1	Bed, table and chair	bed/table/chair	Resting room	MU	07 - July
150701108555.1.1	Dispenser from the Hall	dispenser	Common place	MU	07 - July
150701108556.1.1	Glicosimeter	medical device	Common place	MU	07 - July

150701108565.1.1	Bed railings	bed railings	Recovery room	SC	07 - July
150701108566.1.1	Serum and gas support	serum/gas support	Recovery room	SC	07 - July
150701108567.1.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	07 - July
150701108568.1.1	Surgical table from Room 1	bed	Surgery room	SC	07 - July
150701108569.1.1	Oxigen buttons	surgery device	Surgery room	SC	07 - July
150701108570.1.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	07 - July
150701108571.1.1	Anesthesia syringes	surgery device	Surgery room	SC	07 - July
150701108572.1.1	Surgical table from Room 2	bed	Surgery room	SC	07 - July
150701108573.1.1	Surgical light	surgery device	Surgery room	SC	07 - July
150701108574.1.1	Syringes	surgery device	Surgery room	SC	07 - July
150701108575.1.1	Electric scalpel	surgery device	Surgery room	SC	07 - July
150701108576.1.1	Cabinets and chairs	cabinets/chair	Locker room	SC	07 - July
150701108577.1.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	07 - July
150701108578.1.1	Bed, table and chair	bed/table/chair	Resting room	SC	07 - July
150701108579.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	07 - July
150701108580.1.1	Computer	equipment of common use	Nurse chief room	SC	07 - July
150701108581.1.1	Stretcher from the Hall	bed	Common place	SC	07 - July
150701108582.1.1	Dispenser from the Hall	dispenser	Common place	SC	07 - July
150701108597.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	07 - July
150701108598.1.1	Infusion pumps	medical device	Patient room	ICU-A	07 - July
150701108599.1.1	Screen monitors	medical device	Patient room	ICU-A	07 - July
150701108600.1.1	Curtain	wall/curtain	Patient room	ICU-A	07 - July
150701108601.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	07 - July
150701108602.1.1	Utensils table	table	Patient room	ICU-A	07 - July
150701108603.1.1	Wall	wall/curtain	Patient room	ICU-A	07 - July
150701108604.1.1	Termometer	medical device	Patient room	ICU-A	07 - July
150701108605.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	07 - July
150701108606.1.1	Clipboards	equipment of common use	Nurse station	ICU-A	07 - July
150701108607.1.1	Computers	equipment of common use	Prescription room	ICU-A	07 - July

150701108608.1.1	SEquipMateriais-EquipLimpos	medical device	Medical device room	ICU-A	07 - July
150701108609.1.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	07 - July
150701108610.1.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	07 - July
150701108611.1.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	07 - July
150701108612.1.1	Dryer	equipment of common use	Purge room	ICU-A	07 - July
150701108613.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	07 - July
150701108614.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	07 - July
150701108615.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	07 - July
150701108628.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	07 - July
150701108629.1.1	Infusion pumps	medical device	Patient room	ICU-B	07 - July
150701108630.1.1	Screen monitors	medical device	Patient room	ICU-B	07 - July
150701108631.1.1	Curtain	wall/curtain	Patient room	ICU-B	07 - July
150701108632.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	07 - July
150701108633.1.1	Utensils table	table	Patient room	ICU-B	07 - July
150701108634.1.1	Wall	wall/curtain	Patient room	ICU-B	07 - July
150701108635.1.1	Termometer	medical device	Patient room	ICU-B	07 - July
150701108636.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	07 - July
150701108637.1.1	Refrigerator	equipment of common use	Nurse station	ICU-B	07 - July
150701108638.1.1	Computers	equipment of common use	Prescription room	ICU-B	07 - July
150701108639.1.1	Dispenser from the Hall	dispenser	Common place	ICU-B	07 - July
150701108640.1.1	Utensils from the Bath cart	medical device	Common place	ICU-B	07 - July
150701108641.1.1	Glicosimeter	medical device	Common place	ICU-B	07 - July
150701108642.1.1	Cufometer	medical device	Common place	ICU-B	07 - July
150701108643.1.1	Bath cart	medical device	Common place	ICU-B	07 - July
150701108644.1.1	Emergency cart	medical device	Common place	ICU-B	07 - July
150701108645.1.1	X-Raio cart	medical device	Common place	ICU-B	07 - July
150701108646.1.1	Ultrasound cart	medical device	Common place	ICU-B	07 - July
150701108647.1.1	Passant	medical device	Common place	ICU-B	07 - July

150701108648.1.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	07 - July
150701108666.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	07 - July
150701108667.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	07 - July
150701108668.1.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	07 - July
150701108669.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	07 - July
150701108670.1.1	Bed railings from Patient Room C	bed railings	Patient room	ECU	07 - July
150701108671.1.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	07 - July
150701108672.1.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	07 - July
150701108673.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	07 - July
150701108674.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	07 - July
150701108675.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	07 - July
150701108676.1.1	Chairs	chair	Medication room	ECU	07 - July
150701108677.1.1	Serum and gas support	serum/gas support	Medication room	ECU	07 - July
150701108678.1.1	Computers	equipment of common use	Prescription room	ECU	07 - July
150701108679.1.1	Sink	sink	Special procedures room	ECU	07 - July
150701108680.1.1	Bed	bed	Special procedures room	ECU	07 - July
150701108681.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	07 - July
150701108682.1.1	Bed	bed	Clinic room	ECU	07 - July
150701108683.1.1	Computer table	table	Clinic room	ECU	07 - July
150701108684.1.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	07 - July
150701108685.1.1	Chairs	chair	Reception	ECU	07 - July
150701108686.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	07 - July
150701108687.1.1	Bed, table and chair	bed/table/chair	Resting room	ECU	07 - July
150701108688.1.1	Dispenser from the Hall	dispenser	Common place	ECU	07 - July
150701108689.1.1	Medication cart	medical device	Common place	ECU	07 - July
150804524314.2.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	IU	08 - August

150804524315.2.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	08 - August
150804524316.2.1	Bed railings from Patient Room A	bed railings	Patient room	IU	08 - August
150804524317.2.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	08 - August
150804524318.2.1	Bed railings from Patient Room B	bed railings	Patient room	IU	08 - August
150804524319.2.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	08 - August
150804524320.2.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	08 - August
150804524321.2.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	08 - August
150804524322.2.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	08 - August
150804524323.2.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	08 - August
150804524324.2.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	08 - August
150804524325.2.1	Bed, table and chair	bed/table/chair	Resting room	IU	08 - August
150804524326.2.1	Dispenser from the Hall	dispenser	Common place	IU	08 - August
150804524327.2.1	Glicosimeter	medical device	Common place	IU	08 - August
150804524342.2.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	08 - August
150804524343.2.1	Bed railings from Patient Room A	bed railings	Patient room	MU	08 - August
150804524344.2.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	08 - August
150804524345.2.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	08 - August
150804524346.2.1	Bed railings from Patient Room B	bed railings	Patient room	MU	08 - August
150804524347.2.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	08 - August
150804524348.2.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	08 - August
150804524349.2.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	08 - August
150804524350.2.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	08 - August
150804524351.2.1	Cabinets and chairs	cabinets/chair	Locker room	MU	08 - August
150804524352.2.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	08 - August

150804524353.2.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	08 - August
150804524354.2.1	Bed, table and chair	bed/table/chair	Resting room	MU	08 - August
150804524355.2.1	Dispenser from the Hall	dispenser	Common place	MU	08 - August
150804524356.2.1	Glicosimeter	medical device	Common place	MU	08 - August
150804524365.2.1	Bed railings	bed railings	Recovery room	SC	08 - August
150804524366.2.1	Serum and gas support	serum/gas support	Recovery room	SC	08 - August
150804524367.2.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	08 - August
150804524368.2.1	Surgical table from Room 1	bed	Surgery room	SC	08 - August
150804524369.2.1	Oxigen buttons	surgery device	Surgery room	SC	08 - August
150804524370.2.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	08 - August
150804524371.2.1	Anesthesia syringes	surgery device	Surgery room	SC	08 - August
150804524372.2.1	Surgical table from Room 2	bed	Surgery room	SC	08 - August
150804524373.2.1	Surgical light	surgery device	Surgery room	SC	08 - August
150804524374.2.1	Syringes	surgery device	Surgery room	SC	08 - August
150804524375.2.1	Electric scalpel	surgery device	Surgery room	SC	08 - August
150804524376.2.1	Cabinets and chairs	cabinets/chair	Locker room	SC	08 - August
150804524377.2.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	08 - August
150804524378.2.1	Bed, table and chair	bed/table/chair	Resting room	SC	08 - August
150804524379.2.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	08 - August
150804524380.2.1	Computer	equipment of common use	Nurse chief room	SC	08 - August
150804524381.2.1	Stretcher from the Hall	bed	Common place	SC	08 - August
150804524382.2.1	Dispenser from the Hall	dispenser	Common place	SC	08 - August
150804524397.2.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	08 - August
150804524398.2.1	Infusion pumps	medical device	Patient room	ICU-A	08 - August
150804524399.2.1	Screen monitors	medical device	Patient room	ICU-A	08 - August
150804524400.2.1	Curtain	wall/curtain	Patient room	ICU-A	08 - August
150804524401.2.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	08 - August
150804524402.2.1	Utensils table	table	Patient room	ICU-A	08 - August

150804524403.2.1	Wall	wall/curtain	Patient room	ICU-A	08 - August
150804524404.2.1	Termometer	medical device	Patient room	ICU-A	08 - August
150804524405.2.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	08 - August
150804524406.2.1	Clipboards	equipment of common use	Nurse station	ICU-A	08 - August
150804524407.2.1	Computers	equipment of common use	Prescription room	ICU-A	08 - August
150804524408.2.1	SEquipMateriais-EquipLimpos	medical device	Medical device room	ICU-A	08 - August
150804524409.2.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	08 - August
150804524410.2.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	08 - August
150804524411.2.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	08 - August
150804524412.2.1	Dryer	equipment of common use	Purge room	ICU-A	08 - August
150804524413.2.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	08 - August
150804524414.2.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	08 - August
150804524415.2.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	08 - August
150804524428.2.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	08 - August
150804524429.2.1	Infusion pumps	medical device	Patient room	ICU-B	08 - August
150804524430.2.1	Screen monitors	medical device	Patient room	ICU-B	08 - August
150804524431.2.1	Curtain	wall/curtain	Patient room	ICU-B	08 - August
150804524432.2.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	08 - August
150804524433.2.1	Utensils table	table	Patient room	ICU-B	08 - August
150804524434.2.1	Wall	wall/curtain	Patient room	ICU-B	08 - August
150804524435.2.1	Termometer	medical device	Patient room	ICU-B	08 - August
150804524436.2.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	08 - August
150804524437.2.1	Refrigerator	equipment of common use	Nurse station	ICU-B	08 - August
150804524438.2.1	Computers	equipment of common use	Prescription room	ICU-B	08 - August
150804524439.2.1	Dispenser from the Hall	dispenser	Common place	ICU-B	08 - August
150804524440.2.1	Utensils from the Bath cart	medical device	Common place	ICU-B	08 - August
150804524441.2.1	Glicosimeter	medical device	Common place	ICU-B	08 - August
150804524442.2.1	Cufometer	medical device	Common place	ICU-B	08 - August

150804524443.2.1	Bath cart	medical device	Common place	ICU-B	08 - August
150804524444.2.1	Emergency cart	medical device	Common place	ICU-B	08 - August
150804524445.2.1	X-Raio cart	medical device	Common place	ICU-B	08 - August
150804524446.2.1	Ultrasound cart	medical device	Common place	ICU-B	08 - August
150804524447.2.1	Passant	medical device	Common place	ICU-B	08 - August
150804524448.2.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	08 - August
150804524466.2.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	08 - August
150804524467.2.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	08 - August
150804524468.2.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	08 - August
150804524469.2.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	08 - August
150804524470.2.1	Bed railings from Patient Room C	bed railings	Patient room	ECU	08 - August
150804524471.2.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	08 - August
150804524472.2.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	08 - August
150804524473.2.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	08 - August
150804524474.2.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	08 - August
150804524475.2.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	08 - August
150804524476.2.1	Chairs	chair	Medication room	ECU	08 - August
150804524477.2.1	Serum and gas support	serum/gas support	Medication room	ECU	08 - August
150804524478.2.1	Computers	equipment of common use	Prescription room	ECU	08 - August
150804524479.2.1	Sink	sink	Special procedures room	ECU	08 - August
150804524480.2.1	Bed	bed	Special procedures room	ECU	08 - August
150804524481.2.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	08 - August
150804524482.2.1	Bed	bed	Clinic room	ECU	08 - August
150804524483.2.1	Computer table	table	Clinic room	ECU	08 - August
150804524484.2.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	08 - August
150804524485.2.1	Chairs	chair	Reception	ECU	08 - August
150804524486.2.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	08 - August

150804524487.2.1	Bed, table and chair	bed/table/chair	Resting room	ECU	08 - August
150804524488.2.1	Dispenser from the Hall	dispenser	Common place	ECU	08 - August
150804524489.2.1	Medication cart	medical device	Common place	ECU	08 - August
150826658414.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	IU	09 - September
150826658415.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	IU	09 - September
150826658416.1.1	Bed railings from Patient Room A	bed railings	Patient room	IU	09 - September
150826658418.1.1	Bed railings from Patient Room B	bed railings	Patient room	IU	09 - September
150826658419.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	IU	09 - September
150826658420.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	IU	09 - September
150826658421.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	IU	09 - September
150826658422.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	IU	09 - September
150826658423.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	IU	09 - September
150826658424.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	IU	09 - September
150826658425.1.1	Bed, table and chair	bed/table/chair	Resting room	IU	09 - September
150826658426.1.1	Dispenser from the Hall	dispenser	Common place	IU	09 - September
150826658427.1.1	Glicosimeter	medical device	Common place	IU	09 - September
150826658442.1.1	Dispenser, water tap and flush from Patient Bathroom A	dispenser/water tap/flush	Patient bathroom	MU	09 - September
150826658443.1.1	Bed railings from Patient Room A	bed railings	Patient room	MU	09 - September
150826658444.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	MU	09 - September
150826658445.1.1	Dispenser, water tap and flush from Patient Bathroom B	dispenser/water tap/flush	Patient bathroom	MU	09 - September
150826658446.1.1	Bed railings from Patient Room B	bed railings	Patient room	MU	09 - September
150826658447.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	MU	09 - September
150826658448.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	MU	09 - September
150826658449.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	MU	09 - September
150826658450.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	MU	09 - September

150826658451.1.1	Cabinets and chairs	cabinets/chair	Locker room	MU	09 - September
150826658452.1.1	Dispenser and water tap	dispenser/water tap	Locker room	MU	09 - September
150826658453.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	MU	09 - September
150826658454.1.1	Bed, table and chair	bed/table/chair	Resting room	MU	09 - September
150826658455.1.1	Dispenser from the Hall	dispenser	Common place	MU	09 - September
150826658456.1.1	Glicosimeter	medical device	Common place	MU	09 - September
150826658465.1.1	Bed railings	bed railings	Recovery room	SC	09 - September
150826658466.1.1	Serum and gas support	serum/gas support	Recovery room	SC	09 - September
150826658467.1.1	Countertops and surfaces	countertops/surfaces	Recovery room	SC	09 - September
150826658468.1.1	Surgical table from Room 1	bed	Surgery room	SC	09 - September
150826658469.1.1	Oxigen buttons	surgery device	Surgery room	SC	09 - September
150826658470.1.1	Laryngoscope instrument tray	surgery device	Surgery room	SC	09 - September
150826658471.1.1	Anesthesia syringes	surgery device	Surgery room	SC	09 - September
150826658472.1.1	Surgical table from Room 2	bed	Surgery room	SC	09 - September
150826658473.1.1	Surgical light	surgery device	Surgery room	SC	09 - September
150826658474.1.1	Syringes	surgery device	Surgery room	SC	09 - September
150826658475.1.1	Electric scalpel	surgery device	Surgery room	SC	09 - September
150826658476.1.1	Cabinets and chairs	cabinets/chair	Locker room	SC	09 - September
150826658477.1.1	Dispenser and water tap	dispenser/water tap	Locker room	SC	09 - September
150826658478.1.1	Bed, table and chair	bed/table/chair	Resting room	SC	09 - September
150826658479.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	SC	09 - September
150826658480.1.1	Computer	equipment of common use	Nurse chief room	SC	09 - September
150826658481.1.1	Stretcher from the Hall	bed	Common place	SC	09 - September
150826658482.1.1	Dispenser from the Hall	dispenser	Common place	SC	09 - September
150826658497.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-A	09 - September
150826658498.1.1	Infusion pumps	medical device	Patient room	ICU-A	09 - September
150826658499.1.1	Screen monitors	medical device	Patient room	ICU-A	09 - September
150826658500.1.1	Curtain	wall/curtain	Patient room	ICU-A	09 - September

150826658501.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-A	09 - September
150826658502.1.1	Utensils table	table	Patient room	ICU-A	09 - September
150826658503.1.1	Wall	wall/curtain	Patient room	ICU-A	09 - September
150826658504.1.1	Termometer	medical device	Patient room	ICU-A	09 - September
150826658505.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-A	09 - September
150826658506.1.1	Clipboards	equipment of common use	Nurse station	ICU-A	09 - September
150826658507.1.1	Computers	equipment of common use	Prescription room	ICU-A	09 - September
150826658508.1.1	SEquipMateriais-EquipLimpos	medical device	Medical device room	ICU-A	09 - September
150826658509.1.1	Cabinets and chairs	cabinets/chair	Locker room	ICU-A	09 - September
150826658510.1.1	Dispenser, water tap and flush from Bathroom	dispenser/water tap/flush	Locker room	ICU-A	09 - September
150826658511.1.1	Countertops and surfaces	countertops/surfaces	Purge room	ICU-A	09 - September
150826658512.1.1	Dryer	equipment of common use	Purge room	ICU-A	09 - September
150826658513.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	09 - September
150826658514.1.1	Bed, table and chair	bed/table/chair	Resting room	ICU-A	09 - September
150826658515.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ICU-A	09 - September
150826658528.1.1	Bed railings from Patient Room A	bed railings	Patient room	ICU-B	09 - September
150826658529.1.1	Infusion pumps	medical device	Patient room	ICU-B	09 - September
150826658530.1.1	Screen monitors	medical device	Patient room	ICU-B	09 - September
150826658531.1.1	Curtain	wall/curtain	Patient room	ICU-B	09 - September
150826658532.1.1	Bed railings from Patient Room B	bed railings	Patient room	ICU-B	09 - September
150826658533.1.1	Utensils table	table	Patient room	ICU-B	09 - September
150826658534.1.1	Wall	wall/curtain	Patient room	ICU-B	09 - September
150826658535.1.1	Termometer	medical device	Patient room	ICU-B	09 - September
150826658536.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ICU-B	09 - September
150826658537.1.1	Refrigerator	equipment of common use	Nurse station	ICU-B	09 - September
150826658538.1.1	Computers	equipment of common use	Prescription room	ICU-B	09 - September
150826658539.1.1	Dispenser from the Hall	dispenser	Common place	ICU-B	09 - September
150826658540.1.1	Utensils from the Bath cart	medical device	Common place	ICU-B	09 - September

150826658541.1.1	Glicosimeter	medical device	Common place	ICU-B	09 - September
150826658542.1.1	Cufometer	medical device	Common place	ICU-B	09 - September
150826658543.1.1	Bath cart	medical device	Common place	ICU-B	09 - September
150826658544.1.1	Emergency cart	medical device	Common place	ICU-B	09 - September
150826658545.1.1	X-Raio cart	medical device	Common place	ICU-B	09 - September
150826658546.1.1	Ultrasound cart	medical device	Common place	ICU-B	09 - September
150826658547.1.1	Passant	medical device	Common place	ICU-B	09 - September
150826658548.1.1	Physiotherapeutic armchair	medical device	Common place	ICU-B	09 - September
150826658566.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	09 - September
150826658567.1.1	Serum and gas support from Patient Room 1	serum/gas support	Patient room	ECU	09 - September
150826658568.1.1	Bed railings from Patient Room B	bed railings	Patient room	ECU	09 - September
150826658569.1.1	Serum and gas support from Patient Room 2	serum/gas support	Patient room	ECU	09 - September
150826658570.1.1	Bed railings from Patient Room A	bed railings	Patient room	ECU	09 - September
150826658571.1.1	Serum and gas support from Patient Room 3	serum/gas support	Patient room	ECU	09 - September
150826658572.1.1	Dispenser, water tap and flush from Patient Bathroom	dispenser/water tap/flush	Patient bathroom	ECU	09 - September
150826658573.1.1	Countertops, computer and phone	countertops/computer/phone	Nurse station	ECU	09 - September
150826658574.1.1	Dispenser and water tap	dispenser/water tap	Medicament room	ECU	09 - September
150826658575.1.1	Countertops and surfaces	countertops/surfaces	Medicament room	ECU	09 - September
150826658576.1.1	Chairs	chair	Medication room	ECU	09 - September
150826658577.1.1	Serum and gas support	serum/gas support	Medication room	ECU	09 - September
150826658578.1.1	Computers	equipment of common use	Prescription room	ECU	09 - September
150826658579.1.1	Sink	sink	Special procedures room	ECU	09 - September
150826658580.1.1	Bed	bed	Special procedures room	ECU	09 - September
150826658581.1.1	Countertops and surfaces	countertops/surfaces	Bandage room	ECU	09 - September
150826658582.1.1	Bed	bed	Clinic room	ECU	09 - September
150826658583.1.1	Computer table	table	Clinic room	ECU	09 - September
150826658584.1.1	Dispenser and water tap	dispenser/water tap	Clinic room	ECU	09 - September

150826658585.1.1	Chairs	chair	Reception	ECU	09 - September
150826658586.1.1	Table, countertops and domestic appliances	table/countertops/domestic appliances	Lunch room	ECU	09 - September
150826658587.1.1	Bed, table and chair	bed/table/chair	Resting room	ECU	09 - September
150826658588.1.1	Dispenser from the Hall	dispenser	Common place	ECU	09 - September
150826658589.1.1	Medication cart	medical device	Common place	ECU	09 - September

Table S2. Significance of sample grouping on the overall bacterial community structure based on the ANOSIM statistical method. Month, Facility, Room, and Sample-Type represent the four categories tested. Abbreviations: ANOSIM, analysis of similarity. R, correlation coefficient.

	Bray-Curtis		**Sorensen-Dice**	
	R	**p-value**	**R**	**p-value**
Month	0.08	0.01	0.08	0.01
Facility	0.04	0.01	0.04	0.01
Room	0.1	0.01	0.1	0.01
Sample-Type	0.11	0.01	0.1	0.01

Table S3. Mantel test for the correlation between environmental parameters and samples.

	Bray-Curtis		**Sorensen-Dice**	
Environmental parameters	**Mantel r**	**p-value**	**Mantel r**	**p-value**
Ambient Temperature	0.01	0.24	0.01	0.38
Surface Temperature	0.01	0.53	0.01	0.55
Relative Humidity	0.01	0.71	0.01	0.95

Table S4. List of the 347 unique OTUs with their respective number of reads.

n. of reads	OTU	Species
50902	541137	*Pseudomonas cremoricolorata*
26321	569900	*Planomicrobium okeanokoites*
17773	590838	*Azotobacter chroococcum*
15505	539446	*Psychrobacter sanguinis*
7748	610016	*Cronobacter turicensis*
5135	592170	*Acinetobacter bereziniae*
4103	560590	*Rhizobium aggregatum*
3159	590735	*Lelliottia amnigena*
3145	545589	*Naxibacter indica*
3104	599439	*Lysinibacillus fusiformis*
3040	577367	*Paenarthrobacter ureafaciens*
2960	615166	*Elizabethkingia meningoseptica*
2729	567508	*Acinetobacter calcoaceticus*
2587	573755	*Pantoea wallisii*
2505	573645	*Paenisporosarcina quisquiliarum*
2442	619733	*Lactobacillus helveticus*
2316	594700	*Massilia varians*
2089	582154	*Brevundimonas terrae*
2056	611186	*Flavobacterium phragmitis*
2039	547383	*Arthrobacter globiformis*
1983	571581	*Acetobacter orientalis*
1943	622445	*Pseudomonas poae*
1806	586536	*Pantoea ananatis*
1777	591385	*Terribacillus saccharophilus*
1743	605087	*Leuconostoc pseudomesenteroides*

1600	595595	*Enterobacter ludwigii*
1549	610754	*Acinetobacter beijerinckii*
1493	622852	*Lactobacillus rhamnosus*
1415	569560	*Bacillus vietnamensis*
1312	574226	*Bacillus aquimaris*
1250	586694	*Asaia bogorensis*
1186	579745	*Psychrobacter celer*
1114	564189	*Pseudomonas jessenii*
1108	593218	*Atlantibacter hermannii*
1084	556296	*Stenotrophomonas maltophilia*
1001	570801	*Erwinia aphidicola*
1001	613007	*Pantoea vagans*
991	619885	*Corynebacterium nuruki*
969	613203	*Pantoea vagans*
909	561855	*Eubacterium limosum*
902	618246	*Corynebacterium simulans*
897	612418	*[Clostridium] innocuum*
813	562635	*Paenibacillus borealis*
796	594654	*Chryseobacterium indoltheticum*
766	593061	*Acinetobacter junii*
750	618890	*Citrobacter freundii*
743	579043	*Acinetobacter marinus*
721	570992	*Erwinia aphidicola*
711	624356	*Achromobacter xylosoxidans*
701	592363	*Bacillus simplex*
694	544670	*Serratia ureilytica*
648	580691	*Bacillus niabensis*
643	555547	*Acinetobacter towneri*
642	624358	*Bacillus simplex*
598	618369	*Serratia proteamaculans*

586	587860	*Bacillus niacini*
577	583646	*Chryseobacterium hispanicum*
571	572207	*Klugiella xanthotipulae*
569	592225	*Sphingobium limneticum*
537	577364	*Pseudarthrobacter equi*
521	571496	*Pluralibacter gergoviae*
521	545503	*Glutamicibacter arilaitensis*
521	617582	*Stenotrophomonas maltophilia*
518	593048	*Stenotrophomonas chelatiphaga*
499	539149	*Pantoea ananatis*
494	552190	*Pseudomonas migulae*
472	618466	*Fusobacterium mortiferum*
455	623021	*Serratia liquefaciens*
428	555209	*Paenibacillus graminis*
426	539750	*Enterobacter hormaechei*
424	614318	*Pseudarthrobacter chlorophenolicus*
424	560085	*Erwinia billingiae*
423	624727	*Campylobacter hominis*
421	554616	*Chryseobacterium pallidum*
418	585797	*Psychrobacter pulmonis*
403	543861	*Elizabethkingia meningoseptica*
394	593491	*Pseudomonas stutzeri*
388	585232	*Stenotrophomonas rhizophila*
385	554463	*Massilia brevitalea*
367	595014	*Psychrobacter meningitidis*
335	609922	*Bacteroides eggerthii*
317	599498	*Paenibacillus lautus*
313	608292	*Aeromonas hydrophila*
300	557030	*Weissella fabaria*
299	618760	*Acinetobacter sp.*

292	592408	*Bacillus flexus*
292	594253	*Ochrobactrum tritici*
275	576428	*Pseudomonas composti*
273	600621	*Fusobacterium mortiferum*
272	614559	*Klebsiella oxytoca*
270	587762	*Deinococcus grandis*
249	598146	*Rahnella aquatilis*
247	551122	*Aeromonas caviae*
243	615022	*Capnocytophaga sputigena*
243	582462	*Bacillus massiliosenegalensis*
229	618394	*Actinomyces sp.*
211	550840	*Acinetobacter towneri*
207	624463	*Sphingobium yanoikuyae*
185	543013	*Acinetobacter johnsonii*
184	624263	*Tetragenococcus halophilus*
183	622649	*Raoultella ornithinolytica*
182	582714	*Gluconobacter frateurii*
179	591785	*Glutamicibacter arilaitensis*
176	613062	*Weissella fabalis*
165	622269	*Streptococcus agalactiae*
161	572002	*Pseudomonas oryzihabitans*
157	569657	*Olivibacter jilunii*
144	618621	*Corynebacterium coyleae*
136	569925	*Lactobacillus casei*
135	616434	*Pseudarthrobacter scleromae*
135	618630	*Pseudomonas veronii*
135	544532	*Pseudomonas migulae*
127	618036	*Capnocytophaga gingivalis*
125	595270	*Klebsiella oxytoca*
124	571177	*Massilia aerilata*

111	583551	*Corynebacterium mastitidis*
111	598035	*Aeromonas caviae*
109	596164	*Bacillus benzoevorans*
106	619691	*Raoultella ornithinolytica*
105	615184	*Citrobacter rodentium*
105	552144	*Brevibacterium oceani*
105	622848	*Salmonella enterica*
102	607471	*Scardovia wiggsiae*
101	624241	*Pseudomonas putida*
101	586375	*Alistipes onderdonkii*
98	621566	*Rahnella aquatilis*
88	622294	*Bacillus cereus sp. group*
85	594146	*Arthrobacter pascens*
85	624019	*Pseudomonas putida*
79	617897	*Roseburia inulinivorans*
79	573545	*Veillonella rogosae*
79	586010	*Prevotella nanceiensis*
77	600070	*Advenella kashmirensis*
75	619563	*Oceanobacillus picturae*
69	585625	*Acinetobacter johnsonii*
67	595723	*Facklamia languida*
66	588889	*Morococcus cerebrosus*
66	558387	*Hyalangium minutum*
62	576022	*Sphingomonas dokdonensis*
59	613341	*Pseudomonas marginalis*
58	546076	*Lysinibacillus sphaericus*
57	591996	*Pseudomonas oleovorans*
55	571707	*Auritidibacter ignavus*
55	544133	*Stenotrophomonas maltophilia*
54	611697	*Actinomyces oris*

52	548650	*Sphingobacterium cladoniae*
51	552521	*Methylobacterium jeotgali*
50	580960	*Bacillus megaterium*
49	555159	*Empedobacter falsenii*
49	613570	*Acinetobacter junii*
47	594781	*Ralstonia mannitolilytica*
47	592569	*Pantoea agglomerans*
46	562552	*Pseudomonas rhodesiae*
44	600850	*Burkholderia ambifaria*
44	618392	*Actinomyces odontolyticus*
43	624543	*Pseudomonas nitroreducens*
41	587949	*Acinetobacter guillouiae*
40	596212	*Bacteroides massiliensis*
40	551860	*Prevotella oulorum*
40	616381	*Moraxella lincolnii*
39	566275	*Staphylococcus saccharolyticus*
39	611270	*Fusicatenibacter saccharivorans*
39	599232	*Dyadobacter soli*
38	611388	*Acinetobacter junii*
37	572770	*Rhizobium daejeonense*
36	588048	*Lactobacillus casei*
35	607852	*Brochothrix thermosphacta*
35	552585	*Cobetia crustatorum*
35	538465	*Aeromonas caviae*
34	619224	*Lactobacillus delbrueckii*
32	580143	*Pseudomonas panipatensis*
31	608486	*Lachnoanaerobaculum saburreum*
30	590871	*Lactococcus piscium*
30	623044	*Arcobacter butzleri*
30	562346	*Corynebacterium minutissimum*

30	600999	*Leptotrichia wadei*
30	614399	*Citrobacter werkmanii*
30	574263	*Solibacillus isronensis*
29	576273	*Luteibacter rhizovicinus*
29	602482	*Acinetobacter parvus*
29	548596	*Corynebacterium coyleae*
29	578939	*Raoultella ornithinolytica*
29	561660	*Sphingobium estrogenivorans*
28	608836	*Lactobacillus iners*
27	586962	*Anoxybacillus flavithermus*
27	600958	*Prevotella disiens*
27	614912	*Raoultella ornithinolytica*
27	558128	*Kaistia geumhonensis*
26	624244	*Bifidobacterium longum*
26	594207	*Paenibacillus amylolyticus*
26	559815	*Stenotrophomonas maltophilia*
25	603885	*Burkholderia multivorans*
25	562191	*Corynebacterium segmentosum*
24	609905	*[Eubacterium] siraeum*
23	624460	*Mesorhizobium huakuii*
23	562556	*Pseudomonas rhodesiae*
23	565437	*Facklamia ignava*
23	581613	*Sphingomonas dokdonensis*
23	592259	*Kurthia zopfii*
22	574496	*Rhodococcus artemisiae*
22	557235	*Staphylococcus carnosus*
22	616774	*Brevibacterium epidermidis*
21	571700	*Micrococcus terreus*
21	591451	*Paeniclostridium ghonii*
21	579661	*Sediminibacterium salmoneum*

21	583573	*Sphingomonas yunnanensis*
20	605579	*[Eubacterium] eligens*
20	578689	*Comamonas aquatica*
20	589558	*Acinetobacter haemolyticus*
20	552602	*Skermanella aerolata*
20	607799	*Brachybacterium faecium*
20	544919	*Prevotella melaninogenica*
20	587975	*Tatumella punctata*
20	598566	*Bacteroides vulgatus*
20	611300	*Methylobacterium komagatae*
19	622892	*Lactobacillus acidophilus*
19	611232	*Methylobacterium aquaticum*
19	615892	*Leuconostoc lactis*
19	564973	*Corynebacterium riegelii*
18	616880	*Dermacoccus profundi*
18	617396	*Shewanella putrefaciens*
18	617681	*Melittangium boletus*
18	619346	*Pantoea agglomerans*
18	599810	*Neisseria sicca*
18	623513	*Paracoccus kocurii*
17	584581	*Selenomonas noxia*
17	602393	*Citrobacter freundii*
17	612823	*Peptoniphilus coxii*
17	618874	*Rhodococcus fascians*
17	615628	*Neisseria perflava*
17	615748	*Bifidobacterium adolescentis*
17	599714	*Neisseria cinerea*
16	556253	*Rhizobium gallicum*
16	561365	*Rahnella aquatilis*
16	572041	*Parasegetibacter luojiensis*

16	577010	*Veillonella denticariosi*
16	581684	*Rhizobium sp.*
16	616914	*Streptococcus sinensis*
15	617730	*Campylobacter concisus*
15	622191	*Streptococcus intermedius*
15	617098	*Megasphaera micronuciformis*
15	567953	*Pseudoglutamicibacter cumminsii*
15	539822	*Bacteroides uniformis*
15	572437	*Stenotrophomonas maltophilia*
15	587173	*Rothia aeria*
14	569538	*Weissella hellenica*
14	619012	*Phenylobacterium haematophilum*
14	622476	*Cronobacter sakazakii*
14	574833	*Paenibacillus hunanensis*
14	617118	*Streptococcus equinus*
13	567613	*Leclercia adecarboxylata*
13	585487	*Sporosarcina thermotolerans*
13	624357	*Bosea thiooxidans*
13	608881	*Veillonella parvula*
13	541118	*Sphingomonas yabuuchiae*
13	572147	*Sphingomonas hunanensis*
13	551585	*Kingella denitrificans*
13	591711	*Bacteroides fragilis*
13	603663	*Neisseria bacilliformis*
12	550955	*Brevundimonas aurantiaca*
12	559028	*Cupriavidus pauculus*
12	576137	*Mycoplasma amphoriforme*
12	586262	*Chryseobacterium solincola*
12	622157	*Leuconostoc gelidum*
12	622198	*Streptococcus anginosus*

12	624978	*uncultured Clostridium sp.*
11	538401	*Pseudoclavibacter bifida*
11	589760	*Rickettsia endosymbiont of Bemisia tabaci*
11	597034	*Pedobacter antarcticus*
11	598118	*Parabacteroides goldsteinii*
11	568757	*Alloiococcus otitis*
11	623182	*Enterococcus faecium*
11	541958	*Sphingomonas oryziterrae*
11	620978	*Bacillus coagulans*
11	563179	*Fusobacterium simiae*
11	577941	*Haemophilus parahaemolyticus*
10	571036	*Cobetia marina*
10	591462	*Helcococcus seattlensis*
10	596253	*Faecalibacterium prausnitzii*
10	578222	*Kocuria rhizophila*
10	586187	*Brevundimonas kwangchunensis*
10	617277	*Cellulosimicrobium cellulans*
10	598433	*Blastococcus aggregatus*
10	537940	*Haemophilus parahaemolyticus*
10	568380	*Corynebacterium durum*
10	577023	*Sediminibacterium salmoneum*
10	583027	*Streptomyces radiopugnans*
9	552654	*Geodermatophilus obscurus*
9	571333	*Staphylococcus saprophyticus*
9	580777	*Actinomyces naeslundii*
9	618864	*Caulobacter sp.*
9	599805	*Selenomonas noxia*
9	620919	*Streptococcus lutetiensis*
9	575747	*Rhizorhapis suberifaciens*
9	604819	*Rheinheimera perlucida*

9	615366	*Bosea minatitlanensis*
9	550894	*Achromobacter xylosoxidans*
9	566808	*Acinetobacter johnsonii*
9	604189	*Parvimonas micra*
9	608511	*Eremococcus coleocola*
9	618875	*Bacillus licheniformis*
8	608416	*Streptococcus anginosus*
8	617624	*Caulobacter mirabilis*
8	559422	*Prevotella timonensis*
8	616544	*Lactobacillus jensenii*
8	539049	*Moraxella atlantae*
8	582745	*Macrococcus brunensis*
8	593664	*Xanthobacter flavus*
8	576222	*Corynebacterium aurimucosum*
8	600865	*Acinetobacter ursingii*
7	565772	*Desemzia incerta*
7	573780	*Cellulomonas marina*
7	579592	*Sphingopyxis ummariensis*
7	600174	*Oribacterium sinus*
7	615023	*Oligella urethralis*
7	618486	*Ralstonia pickettii*
7	623755	*Campylobacter gracilis*
7	559005	*Afipia genosp. 13*
7	554268	*Enterobacter cloacae*
7	595571	*Enterobacter cloacae*
7	540944	*Zoogloea resiniphila*
7	549389	*Tatumella punctata*
7	618758	*Filifactor alocis*
6	548444	*Sphingomonas rosea*
6	555709	*Enterococcus avium*

6	558269	*Streptococcus sobrinus*
6	615731	*Sphingomonas canadensis*
6	621950	*Fusobacterium nucleatum*
6	539305	*[Clostridium] hiranonis*
6	561203	*Sphingopyxis macrogoltabida*
6	569972	*Novosphingobium subterraneum*
6	571079	*Pelomonas saccharophila*
6	594851	*Pseudomonas putida*
6	600210	*Lactobacillus jensenii*
6	609392	*Staphylococcus lugdunensis*
6	615333	*Enterococcus malodoratus*
6	617762	*Streptococcus sobrinus*
6	580579	*Bacillus firmus*
6	582871	*Brevundimonas kwangchunensis*
6	614295	*Moraxella atlantae*
6	617611	*Kocuria rhizophila*
6	583487	*Achromobacter xylosoxidans*
6	601676	*Prevotella salivae*
5	554704	*Staphylococcus epidermidis*
5	590164	*Bacillus cibi*
5	561895	*Corynebacterium afermentans*
5	570018	*Sphingomonas jaspsi*

Table S5. Pathogen status of the 70 taxa present in more than 5% of the samples.

Species	OTU	Pathogen Status	Prevalence (%)	Degree	Betweenness Centrality	Eigenvector Centrality
Acinetobacter baumannii	610300	nosocomial pathogen	15	9	52.18	0.07
Acinetobacter baumannii	624096	nosocomial pathogen	30	5	0	0.28
Acinetobacter nosocomialis	606882	nosocomial pathogen	5	21	148.87	0.09
Acinetobacter nosocomialis	607150	nosocomial pathogen	5	4	1	0
Bacillus cereus sp. group	624484	nosocomial pathogen	5	8	0	0.37
Escherichia coli	624449	nosocomial pathogen	38	7	0	0.26
Klebsiella oxytoca	621287	nosocomial pathogen	15	4	3.45	0.08
Klebsiella oxytoca	621896	nosocomial pathogen	5	6	7.42	0.13
Klebsiella pneumoniae	607520	nosocomial pathogen	10	9	22.9	0.02
Klebsiella pneumoniae	620558	nosocomial pathogen	15	12	54.86	0.1
Klebsiella pneumoniae	620675	nosocomial pathogen	5	4	3.83	0.03
Klebsiella pneumoniae	623195	nosocomial pathogen	20	9	14.6	0.14
Klebsiella pneumoniae	623210	nosocomial pathogen	15	8	21.07	0.35
Pseudomonas aeruginosa	624114	nosocomial pathogen	10	14	0	0.56
Pseudomonas putida	623836	nosocomial pathogen	20	5	2.98	0.07
Pseudomonas putida	624672	nosocomial pathogen	25	7	0	0.61
Staphylococcus capitis	605239	nosocomial pathogen	10	0	0	0
Staphylococcus epidermidis	623550	nosocomial pathogen	35	1	0	0.09
Staphylococcus haemolyticus	591357	nosocomial pathogen	5	16	49.1	0.02
Staphylococcus haemolyticus	624535	nosocomial pathogen	10	4	0	0.15
Stenotrophomonas maltophilia	618830	nosocomial pathogen	5	6	18.23	0.09
Acinetobacter johnsonii	561250	rare nosocomial opportunistic pathogen	5	4	0	0
Acinetobacter johnsonii	570673	rare nosocomial opportunistic pathogen	5	5	3.23	0
Acinetobacter johnsonii	583122	rare nosocomial opportunistic pathogen	5	9	0	0
Acinetobacter johnsonii	592001	rare nosocomial opportunistic pathogen	15	2	0	0
Acinetobacter johnsonii	602399	rare nosocomial opportunistic pathogen	10	9	45.49	0.06
Acinetobacter johnsonii	613197	rare nosocomial opportunistic pathogen	10	8	11.83	0.03

Acinetobacter lwoffii	614684	rare nosocomial opportunistic pathogen	15	4	0	0
Acinetobacter ursingii	602718	rare nosocomial opportunistic pathogen	15	9	32.99	0.02
Acinetobacter ursingii	619120	rare nosocomial opportunistic pathogen	15	3	2	0
Pseudomonas oryzihabitans	611399	rare nosocomial opportunistic pathogen	15	8	11.7	0.02
Serratia marcescens	622311	rare nosocomial opportunistic pathogen	5	10	21.71	0.27
Serratia marcescens	623929	rare nosocomial opportunistic pathogen	5	14	35.52	0.4
Staphylococcus hominis	598572	rare nosocomial opportunistic pathogen	15	3	0.91	0.02
Streptococcus mitis	624183	rare nosocomial opportunistic pathogen	20	4	16.95	0.37
Acinetobacter radioresistens	613572	opportunistic pathogen	5	7	15.35	0.12
Finegoldia magna	618642	opportunistic pathogen	5	7	20.45	0.1
Fusobacterium nucleatum	611105	opportunistic pathogen	5	9	43.03	0.11
Haemophilus parainfluenzae	608415	opportunistic pathogen	5	6	4.48	0.04
Staphylococcus warneri	621868	opportunistic pathogen	5	11	66.5	0.21
Anaerococcus vaginalis	617614	rare opportunistic pathogen	5	4	15.82	0.06
Corynebacterium tuberculostearicum	562774	rare opportunistic pathogen	10	12	0	0
Fusobacterium periodonticum	600153	rare opportunistic pathogen	5	7	30.71	0.02
Granulicatella adiacens	600022	rare opportunistic pathogen	5	7	26	0.03
Massilia timonae	595823	rare opportunistic pathogen	5	7	6.37	0
Methylobacterium radiotolerans	623831	rare opportunistic pathogen	10	4	0	0.2
Moraxella osloensis	592450	rare opportunistic pathogen	20	12	24.73	0.01
Murdochiella asaccharolytica	587128	rare opportunistic pathogen	5	3	0	0.14
Pantoea dispersa	599261	rare opportunistic pathogen	5	10	20.14	0.02
Pantoea dispersa	619869	rare opportunistic pathogen	10	8	17.87	0.07
Peptoniphilus asaccharolyticus	565732	rare opportunistic pathogen	5	7	0	0
Pseudomonas fluorescens	624028	rare opportunistic pathogen	5	8	35.2	0.45
Pseudomonas stutzeri	596827	rare opportunistic pathogen	20	5	0	0
Pseudomonas stutzeri	620673	rare opportunistic pathogen	20	4	3.83	0.02
Pseudomonas stutzeri	621004	rare opportunistic pathogen	5	9	70.62	0.2
Pseudomonas stutzeri	621954	rare opportunistic pathogen	5	16	90.68	0.36
Staphylococcus cohnii	598217	rare opportunistic pathogen	5	13	16.3	0.01
Staphylococcus cohnii	623865	rare opportunistic pathogen	5	12	17.2	0.8
Streptococcus parasanguinis	608270	rare opportunistic pathogen	5	6	23.72	0.04

Streptococcus salivarius	620690	rare opportunistic pathogen	5	11	45.74	0.17
Agrobacterium tumefaciens	617866	non pathogen	5	9	19.36	0.07
Agrobacterium tumefaciens	624660	non pathogen	5	4	0	0.1
Bacillus megaterium	624482	non pathogen	5	7	1.75	0.17
Cloacibacterium normanense	542269	non pathogen	5	11	0	0
Paracoccus carotinifaciens	589152	non pathogen	10	11	23.19	0.01
Porphyromonas bennonis	584857	non pathogen	5	3	0	0.13
Pseudomonas monteilii	624642	non pathogen	10	15	0	1
Pseudomonas plecoglossicida	623690	non pathogen	5	10	20.76	0.29
Rubrobacter xylanophilus	600454	non pathogen	10	11	59.53	0.03
Sphingobium yanoikuyae	623802	non pathogen	5	11	6.87	0.56

Table S6. Interaction patterns of the 70 taxa present in more than 5% of the samples. n=negative correlation; p= positive correlation

Source		Target		Corr
Species	**OTU**	**Species**	**OTU**	
Acinetobacter baumannii	610300	*Fusobacterium nucleatum*	611105	n
Acinetobacter baumannii	610300	*Serratia marcescens*	622311	n
Acinetobacter baumannii	610300	*Stenotrophomonas maltophilia*	618830	n
Acinetobacter baumannii	610300	*Streptococcus salivarius*	620690	n
Acinetobacter baumannii	610300	*Staphylococcus cohnii*	623865	p
Acinetobacter johnsonii	592001	*Acinetobacter baumannii*	610300	n
Acinetobacter johnsonii	613197	*Agrobacterium tumefaciens*	624660	n
Acinetobacter johnsonii	583122	*Bacillus megaterium*	624482	n
Acinetobacter johnsonii	583122	*Finegoldia magna*	618642	n
Acinetobacter johnsonii	583122	*Fusobacterium nucleatum*	611105	n
Acinetobacter johnsonii	561250	*Fusobacterium periodonticum*	600153	n
Acinetobacter johnsonii	570673	*Granulicatella adiacens*	600022	n
Acinetobacter johnsonii	602399	*Klebsiella oxytoca*	621287	n
Acinetobacter johnsonii	592001	*Klebsiella pneumoniae*	607520	n
Acinetobacter johnsonii	583122	*Pantoea dispersa*	619869	n
Acinetobacter johnsonii	561250	*Pseudomonas aeruginosa*	624114	n
Acinetobacter johnsonii	613197	*Serratia marcescens*	623929	n
Acinetobacter johnsonii	613197	*Staphylococcus cohnii*	623865	n
Acinetobacter johnsonii	570673	*Staphylococcus haemolyticus*	591357	n
Acinetobacter johnsonii	613197	*Staphylococcus warneri*	621868	n
Acinetobacter johnsonii	583122	*Stenotrophomonas maltophilia*	618830	n
Acinetobacter johnsonii	613197	*Stenotrophomonas maltophilia*	618830	n
Acinetobacter johnsonii	602399	*Streptococcus parasanguinis*	608270	n
Acinetobacter johnsonii	583122	*Streptococcus salivarius*	620690	n
Acinetobacter johnsonii	602399	*Acinetobacter radioresistens*	613572	p
Acinetobacter johnsonii	561250	*Agrobacterium tumefaciens*	624660	p
Acinetobacter johnsonii	583122	*Bacillus cereus sp. group*	624484	p

Acinetobacter johnsonii	570673	*Bacillus megaterium*	624482	p
Acinetobacter johnsonii	613197	*Finegoldia magna*	618642	p
Acinetobacter johnsonii	583122	*Haemophilus parainfluenzae*	608415	p
Acinetobacter johnsonii	602399	*Pantoea dispersa*	619869	p
Acinetobacter johnsonii	602399	*Pseudomonas fluorescens*	624028	p
Acinetobacter johnsonii	561250	*Serratia marcescens*	622311	p
Acinetobacter johnsonii	583122	*Sphingobium yanoikuyae*	623802	p
Acinetobacter johnsonii	570673	*Staphylococcus warneri*	621868	p
Acinetobacter lwoffii	614684	*Acinetobacter ursingii*	619120	n
Acinetobacter lwoffii	614684	*Pseudomonas putida*	623836	n
Acinetobacter lwoffii	614684	*Acinetobacter baumannii*	624096	p
Acinetobacter lwoffii	614684	*Pseudomonas putida*	624672	p
Acinetobacter nosocomialis	606882	*Acinetobacter baumannii*	624096	n
Acinetobacter nosocomialis	606882	*Acinetobacter radioresistens*	613572	n
Acinetobacter nosocomialis	606882	*Agrobacterium tumefaciens*	624660	n
Acinetobacter nosocomialis	607150	*Klebsiella pneumoniae*	623195	n
Acinetobacter nosocomialis	606882	*Methylobacterium radiotolerans*	623831	n
Acinetobacter nosocomialis	607150	*Pseudomonas aeruginosa*	624114	n
Acinetobacter nosocomialis	606882	*Pseudomonas putida*	623836	n
Acinetobacter nosocomialis	606882	*Pseudomonas putida*	624672	n
Acinetobacter nosocomialis	607150	*Staphylococcus haemolyticus*	624535	n
Acinetobacter nosocomialis	606882	*Acinetobacter baumannii*	610300	p
Acinetobacter nosocomialis	606882	*Finegoldia magna*	618642	p
Acinetobacter nosocomialis	606882	*Murdochiella asaccharolytica*	587128	p
Acinetobacter nosocomialis	606882	*Porphyromonas bennonis*	584857	p
Acinetobacter nosocomialis	606882	*Pseudomonas monteilii*	624642	p
Acinetobacter nosocomialis	606882	*Pseudomonas plecoglossicida*	623690	p
Acinetobacter nosocomialis	606882	*Pseudomonas stutzeri*	621004	p
Acinetobacter nosocomialis	606882	*Serratia marcescens*	617614	p
Acinetobacter nosocomialis	606882	*Streptococcus mitis*	624183	p
Acinetobacter radioresistens	613572	*Murdochiella asaccharolytica*	587128	n
Acinetobacter radioresistens	613572	*Pseudomonas stutzeri*	621004	p

Acinetobacter radioresistens	613572	*Staphylococcus cohnii*	623865	p
Acinetobacter ursingii	602718	*Acinetobacter baumannii*	624096	n
Acinetobacter ursingii	619120	*Bacillus megaterium*	624482	n
Acinetobacter ursingii	602718	*Pseudomonas stutzeri*	620673	n
Acinetobacter ursingii	602718	*Serratia marcescens*	622311	n
Acinetobacter ursingii	602718	*Staphylococcus warneri*	621868	n
Acinetobacter ursingii	602718	*Acinetobacter nosocomialis*	606882	p
Acinetobacter ursingii	602718	*Escherichia coli*	624449	p
Acinetobacter ursingii	602718	*Pseudomonas aeruginosa*	624114	p
Acinetobacter ursingii	619120	*Pseudomonas monteilii*	624642	p
Agrobacterium tumefaciens	617866	*Pseudomonas plecoglossicida*	623690	n
Agrobacterium tumefaciens	617866	*Sphingobium yanoikuyae*	623802	n
Agrobacterium tumefaciens	617866	*Staphylococcus haemolyticus*	624535	n
Agrobacterium tumefaciens	617866	*Pseudomonas fluorescens*	624028	p
Agrobacterium tumefaciens	617866	*Pseudomonas putida*	624672	p
Agrobacterium tumefaciens	617866	*Serratia marcescens*	623929	p
Bacillus megaterium	624482	*Pseudomonas monteilii*	624642	p
Cloacibacterium normanense	542269	*Acinetobacter nosocomialis*	607150	n
Cloacibacterium normanense	542269	*Klebsiella pneumoniae*	607520	n
Cloacibacterium normanense	542269	*Serratia marcescens*	623929	n
Cloacibacterium normanense	542269	*Staphylococcus haemolyticus*	591357	n
Cloacibacterium normanense	542269	*Acinetobacter baumannii*	610300	p
Cloacibacterium normanense	542269	*Acinetobacter johnsonii*	570673	p
Cloacibacterium normanense	542269	*Moraxella osloensis*	592450	p
Cloacibacterium normanense	542269	*Paracoccus carotinifaciens*	589152	p
Cloacibacterium normanense	542269	*Pseudomonas aeruginosa*	624114	p
Cloacibacterium normanense	542269	*Pseudomonas monteilii*	624642	p
Cloacibacterium normanense	542269	*Rubrobacter xylanophilus*	600454	p
Corynebacterium tuberculostearicum	562774	*Bacillus megaterium*	624482	n
Corynebacterium tuberculostearicum	562774	*Granulicatella adiacens*	600022	n

Corynebacterium tuberculostearicum	562774	*Haemophilus parainfluenzae*	608415	n
Corynebacterium tuberculostearicum	562774	*Klebsiella oxytoca*	621896	n
Corynebacterium tuberculostearicum	562774	*Pantoea dispersa*	599261	n
Corynebacterium tuberculostearicum	562774	*Pseudomonas monteilii*	624642	n
Corynebacterium tuberculostearicum	562774	*Fusobacterium nucleatum*	611105	p
Corynebacterium tuberculostearicum	562774	*Klebsiella pneumoniae*	623210	p
Corynebacterium tuberculostearicum	562774	*Moraxella osloensis*	592450	p
Corynebacterium tuberculostearicum	562774	*Paracoccus carotinifaciens*	589152	p
Corynebacterium tuberculostearicum	562774	*Pseudomonas stutzeri*	621954	p
Corynebacterium tuberculostearicum	562774	*Serratia marcescens*	623929	p
Finegoldia magna	618642	*Klebsiella pneumoniae*	623195	n
Finegoldia magna	618642	*Porphyromonas bennonis*	584857	p
Finegoldia magna	618642	*Serratia marcescens*	623929	p
Fusobacterium nucleatum	611105	*Serratia marcescens*	623929	n
Fusobacterium nucleatum	611105	*Pseudomonas plecoglossicida*	623690	p
Fusobacterium nucleatum	611105	*Pseudomonas stutzeri*	621004	p
Fusobacterium periodonticum	600153	*Klebsiella pneumoniae*	620558	n
Fusobacterium periodonticum	600153	*Pseudomonas stutzeri*	621954	n
Fusobacterium periodonticum	600153	*Serratia marcescens*	623929	n
Fusobacterium periodonticum	600153	*Stenotrophomonas maltophilia*	618830	n
Granulicatella adiacens	600022	*Escherichia coli*	624449	n
Granulicatella adiacens	600022	*Methylobacterium radiotolerans*	623831	n
Granulicatella adiacens	600022	*Rubrobacter xylanophilus*	600454	n
Granulicatella adiacens	600022	*Pseudomonas monteilii*	624642	p
Haemophilus parainfluenzae	608415	*Bacillus cereus sp. group*	624484	n

Haemophilus parainfluenzae	608415	*Pseudomonas monteilii*	624642	n
Klebsiella oxytoca	621287	*Escherichia coli*	624449	n
Klebsiella oxytoca	621896	*Klebsiella pneumoniae*	623210	p
Klebsiella oxytoca	621287	*Pseudomonas aeruginosa*	624114	p
Klebsiella oxytoca	621896	*Pseudomonas stutzeri*	621954	p
Klebsiella pneumoniae	607520	*Bacillus cereus sp. group*	624484	n
Klebsiella pneumoniae	620675	*Escherichia coli*	624449	n
Klebsiella pneumoniae	607520	*Murdochiella asaccharolytica*	587128	n
Klebsiella pneumoniae	620558	*Pseudomonas aeruginosa*	624114	n
Klebsiella pneumoniae	623195	*Staphylococcus cohnii*	623865	n
Klebsiella pneumoniae	623210	*Streptococcus mitis*	624183	n
Klebsiella pneumoniae	620675	*Bacillus cereus sp. group*	624484	p
Klebsiella pneumoniae	620558	*Bacillus megaterium*	624482	p
Klebsiella pneumoniae	607520	*Fusobacterium nucleatum*	611105	p
Klebsiella pneumoniae	607520	*Haemophilus parainfluenzae*	608415	p
Klebsiella pneumoniae	623195	*Pseudomonas monteilii*	624642	p
Klebsiella pneumoniae	623210	*Pseudomonas monteilii*	624642	p
Klebsiella pneumoniae	620558	*Pseudomonas stutzeri*	621954	p
Klebsiella pneumoniae	607520	*Serratia marcescens*	623929	p
Klebsiella pneumoniae	623195	*Sphingobium yanoikuyae*	623802	p
Klebsiella pneumoniae	620558	*Staphylococcus cohnii*	623865	p
Klebsiella pneumoniae	620558	*Staphylococcus warneri*	621868	p
Klebsiella pneumoniae	620558	*Streptococcus salivarius*	620690	p
Massilia timonae	595823	*Fusobacterium periodonticum*	600153	n
Massilia timonae	595823	*Klebsiella pneumoniae*	607520	n
Massilia timonae	595823	*Pseudomonas monteilii*	624642	n
Massilia timonae	595823	*Serratia marcescens*	617614	n
Massilia timonae	595823	*Acinetobacter johnsonii*	602399	p
Massilia timonae	595823	*Serratia marcescens*	623929	p
Moraxella osloensis	592450	*Acinetobacter johnsonii*	602399	n
Moraxella osloensis	592450	*Haemophilus parainfluenzae*	608415	n
Moraxella osloensis	592450	*Klebsiella pneumoniae*	620558	n

Moraxella osloensis	592450	*Staphylococcus cohnii*	598217	n
Moraxella osloensis	592450	*Streptococcus salivarius*	620690	n
Moraxella osloensis	592450	*Acinetobacter johnsonii*	613197	p
Moraxella osloensis	592450	*Acinetobacter ursingii*	602718	p
Moraxella osloensis	592450	*Fusobacterium periodonticum*	600153	p
Moraxella osloensis	592450	*Sphingobium yanoikuyae*	623802	p
Moraxella osloensis	592450	*Staphylococcus cohnii*	623865	p
Pantoea dispersa	599261	*Agrobacterium tumefaciens*	617866	n
Pantoea dispersa	619869	*Klebsiella oxytoca*	621287	n
Pantoea dispersa	599261	*Klebsiella pneumoniae*	620558	n
Pantoea dispersa	599261	*Pseudomonas plecoglossicida*	623690	n
Pantoea dispersa	619869	*Pseudomonas putida*	624672	n
Pantoea dispersa	619869	*Pseudomonas stutzeri*	621954	n
Pantoea dispersa	619869	*Sphingobium yanoikuyae*	623802	n
Pantoea dispersa	599261	*Acinetobacter johnsonii*	613197	p
Pantoea dispersa	599261	*Acinetobacter nosocomialis*	606882	p
Pantoea dispersa	599261	*Pseudomonas stutzeri*	621954	p
Pantoea dispersa	599261	*Serratia marcescens*	622311	p
Pantoea dispersa	599261	*Staphylococcus cohnii*	623865	p
Paracoccus carotinifaciens	589152	*Fusobacterium nucleatum*	611105	n
Paracoccus carotinifaciens	589152	*Granulicatella adiacens*	600022	n
Paracoccus carotinifaciens	589152	*Pantoea dispersa*	619869	n
Paracoccus carotinifaciens	589152	*Staphylococcus haemolyticus*	624535	n
Paracoccus carotinifaciens	589152	*Staphylococcus hominis*	598572	n
Paracoccus carotinifaciens	589152	*Acinetobacter johnsonii*	602399	p
Paracoccus carotinifaciens	589152	*Klebsiella pneumoniae*	620558	p
Paracoccus carotinifaciens	589152	*Streptococcus salivarius*	620690	p
Peptoniphilus asaccharolyticus	565732	*Acinetobacter nosocomialis*	606882	n
Peptoniphilus asaccharolyticus	565732	*Massilia timonae*	595823	p
Peptoniphilus asaccharolyticus	565732	*Paracoccus carotinifaciens*	589152	p
Peptoniphilus asaccharolyticus	565732	*Pseudomonas oryzihabitans*	611399	p
Peptoniphilus asaccharolyticus	565732	*Pseudomonas plecoglossicida*	623690	p

Peptoniphilus asaccharolyticus	565732	*Pseudomonas stutzeri*	620673	p
Peptoniphilus asaccharolyticus	565732	*Staphylococcus haemolyticus*	591357	p
Pseudomonas fluorescens	624028	*Pseudomonas aeruginosa*	624114	n
Pseudomonas fluorescens	624028	*Acinetobacter baumannii*	624096	p
Pseudomonas oryzihabitans	611399	*Sphingobium yanoikuyae*	623802	n
Pseudomonas oryzihabitans	611399	*Staphylococcus warneri*	621868	n
Pseudomonas oryzihabitans	611399	*Klebsiella oxytoca*	621896	p
Pseudomonas oryzihabitans	611399	*Klebsiella pneumoniae*	623195	p
Pseudomonas oryzihabitans	611399	*Klebsiella pneumoniae*	623210	p
Pseudomonas oryzihabitans	611399	*Streptococcus salivarius*	620690	p
Pseudomonas plecoglossicida	623690	*Staphylococcus cohnii*	623865	n
Pseudomonas plecoglossicida	623690	*Serratia marcescens*	623929	p
Pseudomonas putida	623836	*Acinetobacter baumannii*	624096	n
Pseudomonas putida	623836	*Bacillus cereus sp. group*	624484	p
Pseudomonas stutzeri	596827	*Klebsiella pneumoniae*	607520	n
Pseudomonas stutzeri	621954	*Klebsiella pneumoniae*	623210	n
Pseudomonas stutzeri	621954	*Pseudomonas fluorescens*	624028	n
Pseudomonas stutzeri	596827	*Staphylococcus cohnii*	623865	n
Pseudomonas stutzeri	621004	*Staphylococcus haemolyticus*	624535	n
Pseudomonas stutzeri	596827	*Acinetobacter baumannii*	610300	p
Pseudomonas stutzeri	596827	*Acinetobacter ursingii*	602718	p
Pseudomonas stutzeri	621004	*Escherichia coli*	624449	p
Pseudomonas stutzeri	620673	*Klebsiella pneumoniae*	620675	p
Pseudomonas stutzeri	621004	*Klebsiella pneumoniae*	623210	p
Pseudomonas stutzeri	620673	*Pseudomonas aeruginosa*	624114	p
Pseudomonas stutzeri	621954	*Pseudomonas aeruginosa*	624114	p
Pseudomonas stutzeri	621004	*Pseudomonas fluorescens*	624028	p
Pseudomonas stutzeri	621954	*Pseudomonas monteilii*	624642	p
Pseudomonas stutzeri	596827	*Pseudomonas plecoglossicida*	623690	p
Pseudomonas stutzeri	621954	*Pseudomonas putida*	624672	p
Pseudomonas stutzeri	621954	*Serratia marcescens*	622311	p
Pseudomonas stutzeri	621954	*Sphingobium yanoikuyae*	623802	p

Pseudomonas stutzeri	621954	*Staphylococcus cohnii*	623865	p
Pseudomonas stutzeri	621004	*Staphylococcus epidermidis*	623550	p
Rubrobacter xylanophilus	600454	*Acinetobacter johnsonii*	602399	n
Rubrobacter xylanophilus	600454	*Acinetobacter nosocomialis*	606882	n
Rubrobacter xylanophilus	600454	*Serratia marcescens*	623929	n
Rubrobacter xylanophilus	600454	*Agrobacterium tumefaciens*	624660	p
Rubrobacter xylanophilus	600454	*Fusobacterium nucleatum*	611105	p
Rubrobacter xylanophilus	600454	*Klebsiella pneumoniae*	623210	p
Rubrobacter xylanophilus	600454	*Pseudomonas aeruginosa*	624114	p
Rubrobacter xylanophilus	600454	*Serratia marcescens*	622311	p
Rubrobacter xylanophilus	600454	*Streptococcus salivarius*	620690	p
Serratia marcescens	617614	*Agrobacterium tumefaciens*	617866	n
Serratia marcescens	623929	*Pseudomonas monteilii*	624642	n
Serratia marcescens	623929	*Streptococcus mitis*	624183	n
Serratia marcescens	617614	*Klebsiella pneumoniae*	623195	p
Serratia marcescens	622311	*Pseudomonas fluorescens*	624028	p
Serratia marcescens	622311	*Pseudomonas monteilii*	624642	p
Serratia marcescens	622311	*Sphingobium yanoikuyae*	623802	p
Sphingobium yanoikuyae	623802	*Staphylococcus cohnii*	623865	p
Staphylococcus cohnii	598217	*Acinetobacter nosocomialis*	606882	n
Staphylococcus cohnii	598217	*Acinetobacter radioresistens*	613572	n
Staphylococcus cohnii	598217	*Klebsiella oxytoca*	621896	n
Staphylococcus cohnii	598217	*Klebsiella pneumoniae*	620558	n
Staphylococcus cohnii	598217	*Klebsiella pneumoniae*	620675	n
Staphylococcus cohnii	598217	*Klebsiella pneumoniae*	623195	n
Staphylococcus cohnii	598217	*Pantoea dispersa*	619869	n
Staphylococcus cohnii	598217	*Pseudomonas fluorescens*	624028	n
Staphylococcus cohnii	623865	*Pseudomonas putida*	624672	n
Staphylococcus cohnii	598217	*Sphingobium yanoikuyae*	623802	n
Staphylococcus cohnii	598217	*Pseudomonas aeruginosa*	624114	p
Staphylococcus cohnii	598217	*Pseudomonas putida*	623836	p
Staphylococcus cohnii	598217	*Pseudomonas putida*	624672	p

Staphylococcus haemolyticus	591357	*Klebsiella pneumoniae*	623195	n
Staphylococcus haemolyticus	591357	*Methylobacterium radiotolerans*	623831	n
Staphylococcus haemolyticus	591357	*Pantoea dispersa*	599261	n
Staphylococcus haemolyticus	591357	*Porphyromonas bennonis*	584857	n
Staphylococcus haemolyticus	591357	*Pseudomonas aeruginosa*	624114	n
Staphylococcus haemolyticus	591357	*Pseudomonas oryzihabitans*	611399	n
Staphylococcus haemolyticus	591357	*Serratia marcescens*	622311	n
Staphylococcus haemolyticus	591357	*Serratia marcescens*	623929	n
Staphylococcus haemolyticus	591357	*Acinetobacter nosocomialis*	606882	p
Staphylococcus haemolyticus	591357	*Acinetobacter radioresistens*	613572	p
Staphylococcus haemolyticus	591357	*Agrobacterium tumefaciens*	617866	p
Staphylococcus haemolyticus	591357	*Bacillus cereus sp. group*	624484	p
Staphylococcus haemolyticus	591357	*Pseudomonas monteilii*	624642	p
Staphylococcus hominis	598572	*Pseudomonas plecoglossicida*	623690	n
Staphylococcus hominis	598572	*Pseudomonas aeruginosa*	624114	p
Staphylococcus warneri	621868	*Bacillus cereus sp. group*	624484	n
Staphylococcus warneri	621868	*Klebsiella oxytoca*	621896	n
Staphylococcus warneri	621868	*Methylobacterium radiotolerans*	623831	n
Staphylococcus warneri	621868	*Pseudomonas monteilii*	624642	n
Staphylococcus warneri	621868	*Pseudomonas stutzeri*	621954	n
Stenotrophomonas maltophilia	618830	*Escherichia coli*	624449	n
Stenotrophomonas maltophilia	618830	*Pseudomonas stutzeri*	621954	n
Streptococcus mitis	624183	*Bacillus cereus sp. group*	624484	n
Streptococcus parasanguinis	608270	*Klebsiella pneumoniae*	620558	n
Streptococcus parasanguinis	608270	*Pseudomonas stutzeri*	621004	n
Streptococcus parasanguinis	608270	*Escherichia coli*	624449	p
Streptococcus parasanguinis	608270	*Finegoldia magna*	618642	p
Streptococcus parasanguinis	608270	*Pseudomonas aeruginosa*	624114	p
Streptococcus salivarius	620690	*Bacillus megaterium*	624482	n
Streptococcus salivarius	620690	*Sphingobium yanoikuyae*	623802	n
Streptococcus salivarius	620690	*Staphylococcus warneri*	621868	n
Streptococcus salivarius	620690	*Pseudomonas plecoglossicida*	623690	p