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MEDICAL MICROBIOLOGY Microbial contamination of central supply systems for medical air Contaminação microbiana dos sistemas centrais de abastecimento de ar medicinal Carolina Machado Andrade; Tamara Brown; White Martins Gases Industriais Ltda, Desenvolvimento de Aplicações Medicinais, Rio de Janeiro, RJ, Brasil; Praxair Inc., Pesquisa e Desenvolvimento na Área Medicinal, Tonawanda, NY, USA Correspondence ABSTRACT There are many standards and recommendations for breathing air quality associated with respiratory protection equipment, but little has been done regarding the possible microbial contamination of medical air. The present study demonstrates quantitatively and qualitatively that pipelines might be incriminated as source of microbial contamination of compressed and synthetic air for medical use. Air samples were drawn into an especially pressure-resistant device and the bacterial and fungi contents were identified after growth on agar plates. The bacterial flora isolated from peripheral air outlets was virtually the same as that found in the central air-generating installations, consisting of a mixture of pathogens and normal skin bacteria. Several factors contributing to microbial contamination of medical air are mentioned and preventive measures are discussed. Key words: microbial contamination, compressed air, synthetic air, medical air. RESUMO Existem vários padrões e recomendações para a qualidade do ar respirável relacionado aos equipamentos de proteção respiratória, mas pouco tem sido feito em relação a uma possível contaminação microbiana do ar medicinal. O presente trabalho demonstra quantitativa e qualitativamente que as linhas de ar estão relacionadas à contaminação microbiológica do ar comprimido e ar sintético para uso medicinal. Amostras de ar foram coletadas por um equipamento especialmente resistente a pressão, e o conteúdo bacteriano e fúngico foi identificado após crescimento em placa. A flora bacteriana isolada tanto dos sistemas periféricos de ar foi virtualmente a mesma encontrada nas instalações centralizadas, sendo uma mistura de patógenos e bactérias normais da pele. Vários fatores contribuintes para a contaminação microbiana do ar medicinal e medidas preventivas são discutidas. Palavras-chave: contaminação microbiana, ar comprimido, ar sintético, ar medicinal. INTRODUCTION Air is a mixture of elements and components that, at atmospheric pressure and temperature, is colorless, unscented and flavorless, according to CGA G-7 - 1990. Medical gases for use in health care facilities are typically regulated by national entities, like Brazilian Sanitary Surveillance Agency (Anvisa) in Brazil and the National Fire Protection Agency (NFPA) in the United States. According to Anvisa (3), three types of compressed air can be found in health institutions. These include (1) Industrial compressed air that is used for cleaning and operation of equipment, (2) Medicinal compressed air, which is used for therapeutic purposes, generated by a water seal-, membrane-, or dry lubricated piston compressor. The central supply system must contain filters and/or purifying apparatus, as necessary, to produce medicinal air with the maximum limits for pollutants determined by Anvisa; (3) Synthetic medicinal air is produced by mixing oxygen (21%) and nitrogen (79%). Synthetic medicinal air is used for therapeutic purposes as an alternative to medicinal compressed air. The central supply system for synthetic medicinal air must have oxygen and nitrogen sources meeting purity specifications compatible to those for medicinal use. It has been recently acknowledged that microorganisms can be a contamination hazard in gases (5,6), although no episode relating to contamination of medicinal gases by pathogens has been reported to this date (7). Therefore, the FDA considers Medicinal Air USP as pharmaceutical product. The European Pharmacopoeia also has suggested the classification of medicinal air as a category 2 product: a non-sterile product with contaminant concentration of < 100 CFU (colony forming units)/m<sup>3</sup> of gas, free of pathogenic microorganisms. The recommendations for preventive measures include alternatives that can lead to a reduction/control in contaminant level. Usually, anesthesia personnel assume that the medical gases delivered from the wall outlets in the operating room are clean, correct and safe (5), while ventilators and respiratory care systems are a known source of infection due to bacterial contamination (10,11). However, owners or administrators do not easily accept the idea that the contamination source could be beyond the walls in the pipe systems, possibly due to liability issues and the need to clean the systems once the contamination is identified. Previous preliminary sampling by Praxair Inc. and White Martins Industrial Gases investigated the presence of biological, chemical, and physical contamination in medical air. This work is a continuation of these previous investigations, focusing on the pipeline for medicinal air from compressed and synthetic sources in order to quantify microbial contamination and other contamination levels in the pipe systems. MATERIALS AND METHODS Sites Description Samples were obtained from medicinal air systems of each of four Rio de Janeiro, Brazil-area hospitals. Sites are identified as Hospital I (HPI), Hospital II (HPII), Hospital III (HPIII), and Hospital IV (HPIV). HPI is a 100-bed non-profit hospital, and HPII is a 50-bed private hospital. HPIII is a clinic with less than 50 beds, while HPIV is also a private hospital with approximately 100 beds. The samples for this study were obtained from the medicinal air system located outside and in an indoor, insulated location, respectively for HPI-HPIII and for HPIV. Gas Sampling A portable battery-operated compressed gas sampler (SMA CA-200, Veltex, PA) was used to collect samples. A range of 15 to 84 liters of gas was pumped through the 90 mm-plates placed inside the head of the sampler. In each sample, two types of growth media were selected, SDA (Sabouraud Dextrose Agar) and TSA (Trypticase Soya Agar). Air Quality Evaluation Samples from different points were collected, according to the criteria established in Resolution RE09 (4). The plates were incubated at 30-35°C (TSA) and 20-25°C (SDA), to allow the growth of bacteria and fungi, respectively. Burge et al.(8) have shown that, among the various fungal isolation media, SDA recovers the broadest range and the highest number of airborne fungal species. TSA agar was used to culture environmental bacteria; however, human commensal bacteria could also grow on it. The total microorganism (CFU/m<sup>3</sup>) for each air sample was calculated and the ratio for each group (fungi or bacteria) determined. The three most predominant types of fungi present were identified to the genus level. The isolated bacteria and fungi were identified by standard methods. Gas Analysis A sample for chemical and particulate analysis was also obtained at a use point via a cylinder for laboratory processing by standard methods. Statistical Analysis The main effects and interactions of the variables were calculated using the software Minitab for windows, release 13.32. Coefficients smaller than two times the standard error were presumed to be due to experimental error and were therefore neglected. A total of 65 runs were necessary to estimate the sampling time and the place of sample, considering 95% confidence limit. RESULTS AND DISCUSSION Medicinal Air Sampling (Synthetic Air Mixer) Fig. 1A shows the results found in the HPI. Similarly, for HPII, the average number of bacteria found in the room outlet was 32 cfu/m<sup>3</sup> and the average fungi count was 1.4x10<sup>2</sup> cfu/m<sup>3</sup> (not shown). The total average contaminant level in the emergency outlet of the pipeline and at the outlet of the mixer shown the same variances (95% confidence limit). Background Environmental Air Sampling The average number of microorganisms in the ambient air within the hospital ranged from 0.8 to 1.6x10<sup>2</sup> cfu/m<sup>3</sup> and from 0.9 to 2.0x10<sup>2</sup> cfu/m<sup>3</sup>, for bacteria and fungi, respectively. The highest bacteria count was found in the ICU (Intensive Care Unit), and the highest fungi count was found in the emergency room. HPI outdoors shows average count of 1.7x10<sup>2</sup> cfu/m<sup>3</sup> for both bacteria and fungi. For HPII, the average number of bacteria was 8.0x10<sup>2</sup> cfu/m<sup>3</sup> outdoors and 1.7x10<sup>2</sup> cfu/m<sup>3</sup> within the hospital room. The average fungi count was 1.4x10<sup>2</sup> cfu/m<sup>3</sup> outdoors and 2.6x10<sup>2</sup> cfu/m<sup>3</sup>, within the room. The exact location where the samples were collected may have an effect on the total amount of microorganisms, which could explain the good correlation observed between the number of microorganisms observed in the mixer compared with the microorganisms found in the environmental air. The results show no difference in the contaminant level next to the mixer or in the room (p



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