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REVIEW ARTICLE

Pneumonia prevention in the elderly patients: the other sides

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Abstract

Pneumonia is one of the leading causes of morbidity and mortality from infection in elderly patients. The increased frequency of pneumonia among elderly subjects can be explained by the physiological changes linked to the progressive aging of the respiratory tree and the diminished immunological response. A spiral of event leads to frailty, infection and possible death; preventing pneumonia consists of controlling the risk factors. Dysphagia, which is associated with malnutrition and dehydration, is recognized as one of the major pathophysiological mechanism leading to pneumonia and its screening is crucial for the pneumonia risk assessment. The impairment in the oropharyngeal reflexes results in stagnation of foreign material in the lateral cavities of the pharynx which may then get aspirated repeatedly in the lungs and cause pneumonia. Pneumonia prevention starts with lifestyle modifications such as alcohol and tobacco cessation. A careful review of the risk–benefit of the prescribed medication is critical and adaptation may be required in elders with multiple morbidities. Respiratory physiotherapy and mobilization improve the functional status and hence may help reduce the risk of pneumonia. Maintaining teeth and masticatory efficiency is important if malnutrition and its consequences are to be avoided. Daily oral hygiene and regular professional removal of oral biofilm can prevent the onset of periodontitis and can avoid an oral environment favoring the colonization of respiratory pathogens than can then be aspirated into the lungs.

Keywords Pneumonia · Prevention · Aged over 80 · Oral hygiene · Nutrition · Dysphagia

Introduction

Pneumonia is one of the leading causes of morbidity and mortality from infection in elderly patients [1]. The incidence of community-acquired pneumonia (CAP) is approximately 3 episodes/1000 persons in those aged between 65 and 69 years and increases to 22/1000 persons between the ages of 85 and 89 years [2]. Clinical pneumonia resulted in 6.8 million hospitalizations worldwide and about in 1.1 million in-hospital deaths occurred among them in 2015. The hospital admission rate was increased with age and higher in men [3].

The diagnosis of pneumonia is based on respiratory clinical symptoms such as cough, dyspnea and fever and the presence of a new infiltrate on the chest X-ray. In the elderly, the diagnosis is more complex due to age-related atypical symptoms, like for example, lowering of the temperature threshold. Patients often present to the emergency for falls or with a simultaneous decompensation of comorbidities [4, 5]. A good-quality chest X-ray is often difficult to perform and to interpret in ≥ 65 years old, and the low-dose computed tomography proved to be a more adequate tool for diagnosing pneumonia in the elderly population [6].

Prognosis largely depends on comorbidities such as chronic heart failure, chronic respiratory diseases, neurological diseases, level of dependence, nutritional and cognitive status of the patient [7, 8]. Functional status is an

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independent predictor for short- and long-term mortality in patients hospitalized for CAP and is associated with a risk of recurrent pneumonia in the elderly [9]. Comorbidities increase the risk of CAP by two-to-fourfold. The more frequent ones are: cardiovascular and chronic respiratory diseases, cerebrovascular and neurodegenerative diseases such as dementia and Parkinson's disease, chronic renal or liver disease and immunosuppression [10]. A summary of the effect of morbidities on the risk of CAP is presented in Table 1.

Mortality varies between 5 and 15% in hospitalized patients to 30–50% in intensive care unit patients. In addition, mortality is increased not only during the first months, but also in the years following pneumonia, partly related to an increase in cardiovascular events [11].

Given the high incidence of mortality, pneumonia prevention is particularly relevant in elderly patients. Vaccination is the most effective and known preventive measure and will be treated in another article of this issue. The objectives of this review are to summarize relevant information regarding the particularities of pneumonia in the elderly and its predisposing factors, and to provide an overview of the preventive measures other than vaccination for the elderly population.

Pneumonia: etiology and pathophysiology

Pneumonia is an inflammatory condition of the lung affecting primarily the alveoli. It is most commonly caused by bacteria, but the implication of viruses is now recognized [12]. The etiological agent is rarely identified, especially in elders [13]. In healthy individuals, bacterial density in the lower airways is defined by a constant exchange from the upper respiratory tract and oral microbiota by microaspiration, and retrograde clearance by respiratory epithelium cilia and cough [14]. An imbalance to this system occurs in case of host immunodeficiency, the presence of viral pathogens, or a significant bacterial inoculum which favors the development of pneumonia. Different mechanisms were proposed to how pathogens reach the lower respiratory tract. The inhalation of pathogens can give access to the lower respiratory tract, another major route is the aspiration of secretions from the oropharynx either directly or by reflux from the stomach [15]. A contiguous extension of a colonization/infection and the haematogenous carriage can also lead to pneumonia [16]. Pneumonia is often classified in relation to where it was acquired: community-acquired pneumonia (CAP), nursing home-acquired pneumonia (NHAP), hospital-acquired pneumonia (HAP) or ventilator-associated pneumonia (VAP) have been described. HAP is defined as pneumonia which manifests 48 h or more after admission to hospital, and VAP generally occurs more than 48–72 h after endotracheal intubation. Healthcare-acquired pneumonia (HCAP) occurs in patients with frequent contact with the health system, numerous antibiotic intake and/or a functional state of frailty [17, 18].

Pneumonia is also classified by its physio-pathological mechanism (i.e., aspiration pneumonia) or, if identified, by the etiological pathogen [15]. Jain et al. showed that the presence of influenza and *S. pneumoniae* was five times higher in pneumonia patients older than 65 years when compared to younger ones, and rhinovirus presence was ten times higher [12]. Obtaining high-quality samples is difficult with older patients and only 6% could provide high-quality sputum [13]. Comprehensive molecular testing performed in nasopharyngeal and oropharyngeal swabs is poorly predictive of the presence of pneumonia and proved less sensitive than routine microbiological methods for old patients in a cohort of 199 with a mean age of 83 years [19]. In a review of 33 studies published between January 2005 and July 2012 and focusing on the etiology and treatment of CAP among adults in Europe, Torres et al. concluded that *Streptococcus pneumoniae*, *Haemophilus influenzae* and respiratory viruses were the most frequently observed pathogens, and that *Mycoplasma pneumoniae* was less frequently found among patients ≥ 65 years old [20]. General hygiene measures as contact and droplet precaution and use of masks are necessary tools to tackle global respiratory infections, among which viral infections [21].

Table 1 Chronic diseases and their effect on pneumonia onset (according to Torres et al. 2013)

Morbidities	Risk of CAP
Chronic cardiovascular disease	3 × the risk
Chronic respiratory disease	2 to 4 × the risk
Neurological disease (cerebrovascular disease or stroke, and neurodegenerative disease)	2 × the risk
Chronic renal disease	2 × the risk
Chronic liver disease	2 × the risk
Diabetes mellitus	Moderate risk
Cancer	Moderate risk
Immunosuppression: asplenia, HIV	2 × the risk
Rheumatoid arthritis	Moderate risk
Previous pneumonia	Moderate risk

Dysphagia and malnutrition

The high frequency of pneumonia among elderly subjects can be explained by the physiological changes linked to the progressive decline of the respiratory tree. The mucociliary clearance and the coughing reflexes are reduced leading to a poor airway clearance. Furthermore, the mobility of the oropharynx is often impaired resulting in swallowing

problems. The immunosenescence involves alterations to the innate and adaptive immune systems favoring infections after broncho-aspirations [22, 23]. Oropharyngeal dysphagia (OD) is recognized as one of the major pathophysiological mechanisms leading to aspiration pneumonia in the elders [15, 24]. Swallowing disorders or dysphagia affects 30–40% of the population over 65 years with a prevalence of 75% in stroke patients, 82% in patients with neurodegenerative disease such as Parkinson's disease or 84% with dementia [25]. Dysphagia is also prevalent in 30% of independently living older adults and is independently associated with malnutrition in population of elders with a mean age 82 years [26]. Malnutrition (evaluated by mini nutritional assessment, body mass index (BMI) and serum albumin levels) is also highly prevalent and strongly associated with OD in older patients hospitalized for an acute disease [27]. Neurological diseases, incompetent lip closure, tongue protrusion and poor buccinators muscle strength induce poor masticatory ability, frequently associated with a decreased BMI and serum albumin concentrations [28]. The preparation of a food bolus can be impaired following the loss of teeth, which increases the risk of macro-aspiration into the airways [29]. The loss of masticatory efficiency changes food selection towards decreased consumption of vegetables, fruits, proteins, minerals and vitamins [30]. This poor nutritional intake worsens the decrease of lean muscular mass that occurs naturally with age and aggravates sarcopenia [31]. Moreover, dysphagia patients are often dehydrated because of a reduced sensation of thirst, or the fear of aspiration during drinking, potentially leading to oral dryness. Oral dryness favors oropharyngeal bacterial colonization [25]. The impaired swallowing efficacy often causes the stagnation of foreign material in the lateral cavities of the pharynx which may then get aspirated in the lungs when patients are placed in supine position [25]. Aspiration is often clinically silent because of impaired cough reflexes [32]. Having a low BMI is a predictor for a poor outcome for pneumonia in elder patients hospitalized or in intensive care [8]. For the very old patients aged over 85 years who are hospitalized for CAP, nutritional parameters at admission such as serum protein and albumin are associated with a decreased likelihood of death [7]. Malnutrition is independently associated with 30-day mortality in HAP in nursing home residents [33].

A seated position with the head-up by approximately 30°, during the day and at night, may reduce gastroesophageal regurgitation-associated aspiration [34]. Percutaneous endoscopic gastrostomy which is often performed in case of bronchoaspiration did not decrease its incidence when compared to patients fed with a nasogastric tube [35].

In acute stroke patient who are particularly at risk of pneumonia because of swallowing disorders and an altered mental status, antibiotic prophylaxis is sometimes prescribed for pneumonia prevention. Nevertheless, a recent cluster-randomized trial showed the absence of benefit of antibiotic prophylaxis in patients with dysphagia after stoke and hospitalized in stroke units [36].

Lifestyle-related risk factors

Along with the aging of the global population, the number of smokers aged 65 years or more is increasing worldwide. A recent study in Europe estimated their prevalence to be 11.5% as of 2010 [37]. Their mortality is doubled when compared to non-smokers [38]. Smokers, former smokers and passive smokers have an increased risk of CAP [10, 39]. Former and current smokers have an increased mortality due to infectious diseases in general, and current heavy smoking (> 20 cigarettes/days) is a strong risk factor for pneumonia (HR = 3.30) [40]. Current smoking increases the 30-day mortality in pneumococcal CAP versus non-smokers and ex-smokers [41]. Tobacco smoking plays an indirect role by causing chronic obstructive pulmonary disease (COPD), which is also recognized as a CAP risk factor [42]. Smoking cessation decreases the risk of CAP when compared to current smokers, but the time free from tobacco required to achieve a decreased CAP risk is long (> 10 year) [43]. Moreover, the risk is only decreased if the former smoker does not present with COPD [44]. For old passive smokers, the risk of CAP is also increased when compared to non-exposed persons (OR 1.59) when exposed to smoke for extended periods [39]. Smoking cessation can increase life expectancy, with those quitting at younger age benefiting the most. Quitting at age 65 years increases the life expectancy 1.4–2 years for men and 2.7–3.7 years for women [45, 46].

Another risk factor for CAP relates to alcohol consumption, which is one of the leading causes of mortality worldwide [47]. Excessive alcohol ingestion is commonly linked with liver disease, but it is also an independent risk factor for CAP [10, 48, 49]. Drinkers have an 83% increase in the risk of CAP when compared to non-drinkers. The effect is dose dependent and consuming 10–20 g of alcohol per day was linked to an 8% increase of acquiring CAP [50]. The severity of CAP and the frequency of pneumococcal pneumonia are higher in drinkers than in the general population [51]. For older drinkers, the CAP can result in a severe sepsis [52]. They tend to spend more time in the hospital for pneumonia and have a higher risk of in-hospital mortality when compared to non-drinkers [53]. Moreover, certain pneumococcal serotypes (serotypes 4, 11A, and 19F) were more frequently associated with invasive pneumococcal pneumonia in alcohol abusers [48]. Alcohol intake disturbs the local lung immunological defenses by altering the mucociliary clearance and by diminishing neutrophil chemotaxis and alveolar macrophage functions. With chronic alcohol consumption,

oxidative stress pathways interfere with the normal immune reaction, thus impairing pathogen clearance [54].

Environmental exposures are less-noticed risk factors for pneumonia but they are known to be involved in the pathogenesis of lung diseases. Even in older adults, the long-term exposure to nitrogen dioxide and fine particulate matter were associated with hospitalization for pneumonia [55].

Medication

Elderly patients have often several comorbidities requiring multiple drug prescription. Immunosuppressive therapy and oral steroids have been reported to be risk factors for pneumonia of which clinicians need to be aware when prescribing these drugs [56].

The risk of pneumonia with gastric-acid-suppressive drugs (proton-pump inhibitors (PPI) and Histamine 2 receptor antagonists) has also been reported in many studies as the acid suppression promotes an increase of gastric colonization by pathogens and consequently an alteration of the gut microbiome [57]. The risk seems to be even higher when the treatment has been only recently prescribed [58].

Inhaled corticosteroids (ICS) and antipsychotics (Aps) may increase also the risk of pneumonia and negatively impact on its clinical outcome [59, 60]. ICS, recommended to prevent and treat COPD, lower the pulmonary host defense, hence reducing extracellular release of nitric oxide by alveolar macrophages and decreasing cytokine production [61]. Concerning the use of Aps, a systematic review suggested an increase in the risk for CAP [62], being highest during the early phase of treatment and being dose dependent [63]. The extrapyramidal adverse effect could explain the increased risk for aspiration pneumonia. The anticholinergic side effects of Aps induce dryness of the mouth hence poor bolus preparation and transport. The sedative and anticholinergic side effects of Aps decrease the peristalsis resulting in an increased risk of aspirations [63]. The current use (< 90 days) of benzodiazepine receptor agonists is associated with hospitalization for pneumonia occurrence and is dose dependent, the risk of hospitalization from benzodiazepine hypnotic agents was higher than non-hypnotic and anxiolytic agents. Midazolam was found to increase almost five times the risk of hospitalization for pneumonia. Short- and intermediate-acting benzodiazepine receptor agonists increased the risk more than long-acting agents. [64]. The risk of pneumonia was also increased with non-hypnotic benzodiazepine in older adults [65]. The muscarinics-1 and histaminergic-1 effects cause esophageal dilation and hypomotility in addition to sedation thus explaining the increased risk for pneumonia infection [66].

The susceptibility to pneumonia is increased among patients with diabetes and could be associated to antidiabetic drugs as thiazolinediones, which have immunomodulatory and glucocorticoid-like properties [67]. Nevertheless, the evidence is conflicting and large epidemiological studies are needed to confirm the association of antidiabetic drugs with the incidence of pneumonia [68–70].

On the other side, some medications were reported to have a protective effect on the occurrence of pneumonia. A meta-analysis supported the hypothesis that statins may reduce the risk of pneumonia but the quality of the currently available scientific evidence is unfortunately very low [71].

There is conflicting information on the protective effect of angiotensin II receptor blockers (ARB), and angiotensin-converting enzyme inhibitors (ACE) on the incidence of pneumonia. ARB and ACE were shown to decrease the risk of pneumonia in old hypertensive patients with Parkinson's disease and in post-stroke patients with aspiration pneumonia [72, 73].The possible explanation for the action of statins, ARB and ACE would be a modulatory effect in the inflammatory state by altering the production of proinflammatory cytokines, improving endothelial and mitochondrial dysfunction and to decreasing the reactive oxidative species [74]. Another mechanism of ACE would be an increase of substance P levels in the airways and plasma, improving swallowing function and cough reflexes [75].

Amantadine was found to have a beneficial effect on the risk of pneumonia in patients with a history of stroke, the dopamine supplementation improved the swallowing reflexes thus reducing by 20% the pneumonia risk [76].

More evidence from well-designed trials is needed to assess the link between therapeutic drugs and the risk of pneumonia, but at any rate, clinicians need to be particularly cautious when treating elders with multiple morbidity who take already multiple prescribed medications. The adequate policy for elders is to minimize prescriptions and potentially stop inappropriate ones [77].

Physiotherapy

Physiotherapy is not commonly suggested as prevention of pneumonia [78], although poor functional status is considered a risk factor for recurrent CAP [9]. The loss of muscle mass is an independent predictive factor of 3-month mortality in patients hospitalized for aspiration pneumonia [79]. In functionally unimpaired older adults, the development of a mobility limitation increases the risk of developing pneumonia [80]. Hence, two physiotherapeutic novel approaches could modify the pneumonia risk. The first concerns respiratory physiotherapy, which consists in guiding the patient into performing deep breathing exercises, coordinated breathing. The assisted cough helps the patient clear the secretions. It was shown that non-invasive ventilation and continuous positive airway pressure used prior to abdominal high-risk

elective surgeries reduce the risk of post-operative pulmonary complication such as pneumonia and atelectasis [81]. Preoperative inspiratory muscle training in adults undergoing cardiac and major abdominal surgery decreases the post-operative pulmonary complications and reduces the length of hospital stay [82–84]. A pulmonary rehabilitation program initiated on the day following surgery showed a reduction in the incidence of pneumonia in elderly patients treated for hip fracture [85]. Moreover, early rehabilitation reduced the 30-day in-hospital mortality in geriatric patients with aspiration pneumonia [86].

The second approach is the mobilization physiotherapy; it targets mobility rather than chest function; it is well documented in post-operative care. The mobilization protocols are not structured and standardized and their effectiveness is still questionable, but mobilization reduces nevertheless the risk of complications associated with bed rest [87]. In a post-operative context, early mobilization and respiratory physiotherapy are associated with a decrease in pneumonia incidence and respiratory complications [88].

Among medical patients, early mobilization reduced the incidence of HAP with a hazard ratio of 0.39, it also reduced the length of stay in hospital [89]. Including an inpatient exercise-based rehabilitation program improves the functional status of the patient as well as quality of life but may not have a direct impact on pulmonary function or the length of hospitalization [90].

Oral health

A natural dentition can be a major advantage in term of retaining the masticatory capabilities and chewing muscle bulk. Nevertheless, keeping one's natural teeth can become more challenging when physical limitations occur, as poor oral hygiene fosters the accumulation of dental plaque, a bacterial biofilm adhering to the teeth [91]. Patients having more than ten natural teeth and periodontal pocket depths of more than 4 mm presented an increased mortality due to pneumonia than those with periodontal pockets smaller than 4 mm [92, 93]. The quantity of periodontal bacteria in 85-year-old persons increases with the number of teeth [94]. Broncho-alveolar lavage samples from patients admitted to the hospital with clinical symptoms of pneumonia have confirmed the involvement of oral microorganism [95]. Anaerobic germs (*Veillonella* sp. and *Porphyromonas gingivalis*) implicated in periodontal disease were found in dentate patients as well as a high concentration of *Staphylococcus aureus* in patients wearing prosthetic appliances [95].

Periodontal disease leads to an increase in the mass and the diversity of the microbiota causing the destruction of the tissues surrounding the teeth [96]. It prompts a true immune response of the host with triggering of the inflammatory cascade, various signaling molecules are implicated, i.e. cytokines, the cytokines secreted into mucosal saliva may promote adhesion and colonization by respiratory pathogens such as *Pseudomonas aeruginosa* and enteric bacilli [97]. Ortega et al. showed that oral colonization by respiratory pathogens was more common in frail patients with oral dysphagia (with or without pneumonia) than in healthy subjects without dysphagia (93% vs. 67%) [98]. Hence, the oral biofilm constitutes a likely reservoir for pulmonary pathogens [99–103]. It has also been suggested that an imbalanced oropharyngeal microbiota reduces colonization resistance allowing pathogens to spread [104]. Elderly pneumonia patients presented a higher proportion of some *Streptococcus* species and *Rothia*, whereas *Gemellales*, *Prevotella*, *Veillonella dispar*, *Parascardovia* and *Leptotrichia* had lower relative abundance compared to non-pneumonia controls. An age-related decrease in anaerobic colonization (*Prevotella, Veillonella, Leptotrichia*) increased the susceptibility to pneumonia. Bacterial diversity (expressed as Shannon diversity index) and bacterial load of oropharyngeal microbiota were increased in elderly pneumonia patients in comparison to elderly without pneumonia [104].

These cytokines produced at high levels in gingival fluid and serum are in contact with respiratory epithelial cells via the hematogenous route. This inflammatory event can lead to epithelial and endothelial injury that increases susceptibility to infections; they were shown to result in a shift from a diverse microbiota in the lungs to a dominance by single species such as *Streptococcus pneumonia* and *Pseudomonas aeruginosa* [105].

A systematic review conducted in 2008 analyzed the link between oral hygiene and the risk of pneumonia and concluded that one in ten deaths could be avoided in elders living in institutions through adequate and regular dental hygiene [106]. However, according to the authors, the mechanical cleaning of the teeth should be done by dental professionals, such as hygienists or dentists, because the risk of pneumonia was unchanged when hygiene was provided by the nursing or auxiliary staff [107].

Tongue coating is also associated with a higher bacteria count in the saliva and the development of aspiration pneumonia [108]. Furthermore, patients wearing removable dentures at night were more likely to develop pneumonia than those removing it before bedtime [109]. The dentists' current recommendation is to remove dentures at night to prevent oral candidiasis promoting bacterial superinfection and to keep them dry or in a disinfectant bath [110–112].

Chlorhexidine mouthwashes significantly reduce bacterial colonization in elders [113]. However, chlorhexidine is an adjuvant and cannot replace mechanical cleaning or brushing because its chemical efficacy against bacterial biofilm is limited. Thus, concentrations of 0.12–0.2% can be used, an initial debridement of dental plaque is still necessary [114].

Table 2 Overview of pneumonia preventive measures (adapted from Prina et al. 2015)

Risk factor	Recommendation
Patient status	
Old age	Vaccination against *H. influenza* and *S. pneumonia*
Swallowing disorder	Thicken liquids, small bolus, adjust head position
Hyposalivation	Chewing of gum, oral moisturizers
Malnutrition	Food supplements, adjustment of diet, assuring chewing efficiency
Physical frailty	Physiotherapy
	CPAP prior to surgery and preoperative training
Environment	
Environmental exposure	Limit exposure to nitrogen dioxide and fine particulate matter
Living in a nursing home or frequently exposed to healthcare environment	Favor home care when possible
Habits	
Smoking	Smoking cessation
Alcohol	Adjust alcohol consumption
Medications	
Immunosuppressive drugs and oral steroids	Monitor carefully when used and adjust if possible
PPI and H2 receptor antagonists	Monitor carefully when used and adjust if possible
Inhaled corticosteroids	Monitor carefully when used and adjust if possible
Antipsychotics	Monitor salivary flow and in case of hyposalivation treat xerostomia
Benzodiazepine	Check level of sedation and adjust where necessary
Statin	Prescribe when indicated, as it may reduce the risk of pneumonia
ACE inhibitors and ARB's	Prescribe when indicated, as it may reduce the risk of pneumonia in Parkinson patients
Amantadine	Prescribe when indicated, as it may reduce the risk of pneumonia in stroke patients
Oral health	
Oral hypofunction	Regular dental check-up visits and treatment where indicated
Poor oral hygiene	Daily oral hygiene in addition to regular oral hygiene provided by dental hygienist
Poor tongue hygiene	Use of tongue scrapping
Dental prosthesis	Assure denture hygiene and remove denture during sleep

CPAP continuous positive airway pressure, *PPI* proton-pump inhibitors, *H2* histamine 2 receptor antagonist, *ACE inhibitors* angiotensin-converting enzyme inhibitors, *ARB* angiotensin II receptor blockers

A recent systematic review evaluated the effect of oral care measures for residents in nursing homes and long-term facilities. The analysis was unable to confirm that professional oral care resulted in a lower incidence of NHAP or reduced the risk of pneumonia associated mortality. The authors concluded that professional oral care or oral care measures may reduce mortality due to pneumonia but the level of evidence is still insufficient [115].

An overview of suggested preventive measures than can be undertaken to control the risk factors for pneumonia in the elderly is presented in Table 2.

Conclusion

While the vaccine remains the best known and best studied pneumonia prevention strategy, it is important to recognize the modifiable risk factors that are related to the onset of pneumonia. Dysphagia is a major risk factor for pneumonia, its screening and management is also important to prevent malnutrition, dehydration and xerostomia. Controlling lifestyle related risk factors such as smoking, and alcohol consumption could decrease the risk of pneumonia, even in elderly patients. A careful review of the prescribed medications and a close monitoring of side effects can also reduce the risk of pneumonia. Maintaining teeth and thus chewing muscle mass can improve oral function, nutritional and functional status. Simple everyday practices such as oral biofilm removal or removing the denture before bedtime are easy to implement and efficient measures to reduce the risk of pneumonia.

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Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest. The funders had no role in the design of the study, in the collection, analyses, or interpretation of data, in the writing of the manuscript, or in the decision to publish the results.

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors.

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