Research Progress on Air-liquid Interface Activity and Function in Respiratory Tract and Digestive Tract

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Abstract: Air-liquid interface activity is closely related to the health of the body. The change of interfacial activity affects the normal physiological functions, such as the internal environment, microecological balance and nutrient absorption and utilization of the body. In order to provide a new idea for the evaluation and pre-treatment of respiratory and digestive diseases, and provide theoretical support for the development of functional food. The formation mechanism of gas-liquid interface is introduced, the relationship between gas-liquid interface activity and respiratory tract and digestive tract is combed, and the factors affecting the interface activity are summarized.

1 INTRODUCTION

The air-liquid exchange interface in the body has a significant impact on the normal physiological function of organs. In the unhealthy state, the activity of the air-liquid interface will change accordingly. The occurrence of respiratory diseases such as asthma, bronchitis, chronic obstructive pulmonary disease, oral, gastric and inflammatory bowel diseases goes hand in hand with the change of air-liquid interface activity. Asthma and other airway inflammatory diseases are accompanied by a significant increase in airway mucus viscoelasticity (Johnson 2011). In the lungs of patients with respiratory distress syndrome, protein rich pulmonary edema can inhibit or destroy the habitual function of surfactants, resulting in increased surface tension of pulmonary fluid (Luo *et al.* 2017). Inflammatory bowel disease is accompanied by lipid peroxidation and decreased antioxidant capacity, resulting in an imbalance between oxidation and antioxidant, increasing surface pressure. The increase of surface pressure causes damage to small intestinal mucosa (Xie & Wan 2020).

Although more and more studies have found that the air-liquid interface plays a key role in the occurrence and prevention of diseases, the causal relationship between the activity of the air-liquid interface and the occurrence of diseases and the related molecular mechanism is not clear. This review expounds and summarizes the formation of air-liquid interface, the application of air-liquid interface activity in the health evaluation of respiratory system and digestive system, and the factors affecting interface activity. In order to provide a unique perspective for the prevention and treatment of related diseases, and provide a theoretical reference for the design and production of functional foods for the treatment and prevention of related diseases.

2 FORMATION OF GAS-LIQUID INTERFACE

The surface of the oral cavity, airway, stomach, intestine and other organs of the organism is covered with a thickness of 2-10 μM mucus layer (Puchelle *et al*. 1987), which is the first line of defense of human immune system (Bajka *et al*. 2015). It plays a role in

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resisting the invasion of bacteria, viruses and harmful compounds. Mucus is a gelatinous polymer composed of glycosylated mucin, containing 90%-95% of water, 1%-2% lipids and electrolytes (Paola 2020, Widdicombe & Wine 2015). Therefore, mucus has complex rheological properties, such as shear thinning, viscoelasticity, adhesion (Puchelle *et al.* 1987). The exertion of various physiological functions of the body largely depends on the appropriate rheological properties of mucus. In the respiratory system, the protective and scavenging function of airway mucus is dependent on the appropriate rheological properties of mucus, otherwise airway mucus may become a factor of airway obstruction. For example, the airway of asthmatic patients will over secrete mucus with high viscosity, resulting in poor fluidity of mucus and difficult to discharge from the airway. In severe cases, airway mucus embolism will be formed in the airway (Johnson 2011), resulting in atelectasis, asphyxia, respiratory failure and even sudden death (Luo *et al.* 2017). In the digestive system, human digestive tract mucus plays an important role in the digestion and transportation of food in the gastrointestinal tract and the lubrication and protection of gastrointestinal mucosal surface (Zhou *et al.* 2004). Moreover, intestinal fluid rheology is also closely related to the colonization, distribution and growth of intestinal flora, and then affects the absorption effect of nutrients and drugs (Boegh *et al.* 2014).

There is a surface layer of the contact between the mucus layer and the external atmospheric environment, referred to as the air-liquid interface. The surface response of air-liquid interface to dynamic conditions is closely linked to the stability and health of various tissues and organs. In the development of natural science and light industry, the application of air-liquid interface activity has been widespread. For example, the production principle of Yuba is to denature the protein in soybean milk by heating. The hydrophobic groups inside the protein are exposed, while the hydrophilic groups outside the protein are transferred to the inside of the molecule. The protein is concentrated, collided and condense, precipitate, and gradually grow into a film. Then, the sugar is adsorbed on the protein by van der Waals force or hydrogen bond to gradually form a certain toughness of Yuba (Chen 2010). However, there is little understanding of how the air-liquid interface in a human body affects the normal physiological function of various tissues and organs. The main reason is that the structure of the human body is much more complex than that of other organisms. And it belongs to soft matter from the perspective of materials, which are essentially different from inorganic materials (Jin 2014). Therefore, it's great significance for understanding the mechanism of physiological activities and disease occurrence and development to study the air-liquid interface with special functions in the human body at the micro scale.

3 ACTIVITY AND FUNCTION OF AIR-LIQUID INTERFACE IN RESPIRATORY TRACT

3.1 Nose

The surface of nasal mucosa is the subject with a mucus blanket with a thickness of 5-10 μm. It can adsorb about 80% of the particles in the gas, and the mass of these particles is greater than 12.5 μg (Zhang *et al.* 2006). Mucociliary blanket and ciliated epithelial layer below it constitute nasal mucociliary transmission system, which is an important mechanical defense system of human body. Guo Yongqing (Guo *et al.* 2004) tried to construct a Construction of ciliary differentiation model of cultured human nasal epithelial by using air-liquid interface culture technology, and quantitatively analyzed the degree of ciliary differentiation. The research provide an advanced research means for exploring the physiological and pathological characteristics of respiratory epithelial cells. Zhao Yanyan (Zhao *et al.* 2020) infected human nasal epithelial cells cultured at the air-liquid interface with Staphylococcus aureus standard strains and clinical isolates of chronic rhinosinusitis, and analyzed the effects of Staphylococcus aureus infection on the tight junction and cell viability of nasal epithelial cells.

3.2 Hung

The human airway is covered by mucus layer. The changes of rheological properties such as mucus viscosity and surface tension affect the physiological functions such as airway stability, lung defense barrier and mucociliary clearance. Long Yun (Long *et al.* 2006) and others think that the collapse of airway compliance largely depends on the pressure load of surface tension on airway wall. Due to the compression of the airway, the increase of the curvature of the air-liquid interface will increase the surface tension, resulting in the increase of the load on the airway wall, and finally affect the compliance of the airway wall. Pulmonary surfactant (PS) is a lipid complex protein mixture. It covers the alveolar surface and makes the respiratory function normal by reducing the surface tension at the air-liquid interface (Wang＆Hu 2020). At the same time, it also has the functions of stabilizing the airway and protecting against the pulmonary edema (Liu *et al*. 2020). Ann M. czyzewski (Czyzewski *et al.* 2018) et al. used biomimetic lipid mixture to simulate the physiological activity of pulmonary surfactant proteins B and C at the air-liquid interface. The study provides a strong theoretical basis for the new medical research that natural surfactants can effectively treat acute lung injury. Ji Xiaoli (Ji *et al*. 2021) constructed a primary culture model of mouse tracheal epithelial cells by air-liquid interface culture. It was applied to the evaluation of the inhalation toxicological *in vitro*.

4 ACTIVITY AND FUNCTION OF AIR-LIQUID INTERFACE IN DIGESTIVE TRACT

4.1 Oral Cavity

Human saliva is mainly produced in the oral cavity by the parotid gland, submersible gland and the sublingual gland. It is a major biological liquid to maintain oral health. Saliva not only serves as a barrier to pathogens, but also promotes food intake. It also provides a medium for dissolved and suspended food substances to stimulate the taste. As the core component of salivary membrane covering teeth, mucin plays a role in helping lubricate, protect and heal oral mucosa (Liu 2018). Rossetti (Rossetti *et al.* 2013, Rossetti *et al.* 2013) examined the effect of the interaction between proteins of human oral saliva and different polyphenols on the expansion characteristics of air-liquid interface during oral digestion. Firstly, the complexity of kinetics and interfacial expansion rheology of salivary membrane was investigated. The results showed that small molecular weight protein components adsorbed on the interface and produced solute like surfactant reaction, but the adsorption of higher molecular weight proteins makes the interface more and more insoluble.

4.2 Stomach

There are many kinds of internal and external secretory cells in gastric mucosa, such as principal cells, parietal cells and G cells. Its synthetic and released gastric juice and various gastrointestinal hormones are widely involved in digestion and absorption (Liu *et al*. 2020). Mucosa can protect the stomach from gastric acid erosion. Once damaged, the stomach will be invaded by gastric acid and bacteria for a long time. In severe cases, it will be expected to result in gastric ulcer, gastric perforation and other diseases (Jin 2014). Helicobacter pylori infection will destroy the structure and function of gastric mucosal epithelium. Jin Lin (Jin 2014) explored the mechanism of Helicobacter pylori infection of gastric mucosal epithelium by establishing H. pylori gastric mucosal epithelial air-liquid interface. The study provides a theoretical basis for the prevention and treatment of gastrointestinal diseases caused by infection.

4.3 Intestine

In the intestine, the small intestine is the main port for nutrient absorption. The lumen surface is composed of intestinal epithelial cells and covered by a mucus layer, which is a barrier against harsh digestive environment (Huang 2021, Ou *et al*. 2021). Many microbiota are colonized on the surface of intestinal mucosa, which coexist with the human body and maintains the normal physiological function of the host. Once the flora structure is changed, the proportion of bacteria is unbalanced, and the metabolism will be disorder. It may lead to the occurrence of various diseases such as diabetes mellitus (Yao 2020). Studies have shown that changes in duodenal fluid rheology usually affect the colonization, distribution and growth of intestinal flora, and then affect the absorption effect of nutrients and drugs (Morrison & Preston 2016). Johansson (Johansson *et al*. 2015) et al. further found that the number of goblet cells in the intestinal mucus of green fluorescent mice was depressed, and the intestinal mucus was anchored on the goblet cells and could not be sucked out through the experiment. Compared with conventional mice, green fluorescent mice have poor intestinal mucus rheology and reduced air-liquid interface activity. It makes intestinal flora colonize difficultly, and then affecting the distribution and growth of intestinal flora. TARGO (Tamargo *et al.* 2018) also show that the increase of colonic mucus viscosity is conducive to the reproduction of anaerobic bacteria, and the reproduction of the intestinal flora will also increase the viscosity of the culture system. The changes in the composition of intestinal flora also reduce the thickness of the intestinal mucus layer and increase mucus permeability (Wells *et al*. 2017). Some intestinal probiotics have been proved to regulate intestinal epithelial function by promoting the formation of mucosal layer, secreting antibacterial factors, promoting the secretion of secretory immunoglobulin A and competitive adhesion with intestinal epithelial cells (Liu *et al*. 2020), and reduce surface tension and increase expansion modulus to maintain intestinal barrier homeostasis and promote health.

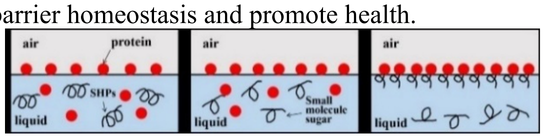

Figure 1: Model of soy hull polysaccharides-protein in simulated intestinal fluid (Huang 2020).

5 IMPACT FACTORS OF THE ACTIVITY OF AIR-LIQUID INTERFACE

5.1 Effect of Surfactant on Interfacial Activity

Surfactant plays a decisive role in regulating the viscosity of mucus and the rheological properties of air-liquid interface. As shown in Figure 2, natural PS provides surface activity of alveoli during respiration. During inhalation, PS forms a surface active membrane which adsorbs to the air-liquid interface to cover the alveolar area. When exhaling, PS forms a compressible multilayer structure. The surfactant film is filled with PS that greatly reduces the alveolar surface tension. When the alveolar dilation is caused by re inhalation, the surfactant membrane expands again. And the phospholipids at the air-liquid interface redistribute laterally to cover the alveolar area.

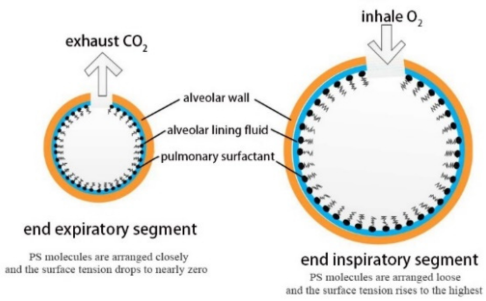

Figure 2: Schematic diagram of alveolar structure and function (Dong *et al*. 2016).

Studies have shown that there is a substance similar to PS in the eustachian tube, mainly composed of phosphatidylcholine and protein. It can reduce the surface tension of the ciliated mucus blanket on the surface of the eustachian tube, and prevent the adhesion of the tube wall and contribute to the discharge of secretions. The reduction of surfactant is not conducive to the opening of the eustachian tube and the discharge of secretions (Zhu *et al*. 2013). Fornadley (Dong *et al*. 2016) et al. found that by injecting exogenous surfactant, the open pressure of eustachian tube in gerbils with secretory otitis media model decreased significantly and closed to the normal value.

Due to the electrostatic attraction of opposite charges, intestinal mucin can combine with some lipids or polysaccharides to form surfactant. It can change the activity of air-liquid interface. Luo Xinyou (Luo *et al*. 2015) et al. treated sinusitis of children with a composite surfactant composed of soybean based grain extract and cationic surfactant (chitosan quaternary ammonium salt). Through the inhibitory effect of Chitosan on the biological interface of Streptococcus mutants and the reduction of toxicity and enhancement of bactericidal effect of grain extract, the nasal ventilation and sinus drainage are improved. Guri (Guri *et al*. 2015) et al. used epithelial mucus extracted from human colon adenocarcinoma cell HT29-MTX to explore the interaction between tea polyphenols and milk protein and intestinal mucus at the air-liquid interface. It is found that the main component of tea polyphenols (epigallocatechin-3-gallate) can bind to milk protein. The formed complex reduces the interfacial surface tension and increases the expansion modulus. It indicate that nutrient molecules are more likely to interact with intestinal mucus before they are effectively transported and absorbed by intestinal cells. The research of Huang Jinghang (Huang *et al.* 2020) shows that soybean hull polysaccharide may form a similar polysaccharide mucin complex in the intestinal digestion in the delivery system. Therefore, in the process of interfacial adsorption, the interfacial tension decreases and the interfacial expansion modulus increases are conducive to the release and absorption of nutrients. The study provides theoretical support for the development of beneficial functional foods of soy hull polysaccharide.

5.2 Effect of pH on Interfacial Activity

A large number of experiments have proved that the change of pH value will affect the activity of air-liquid interface. By studying the film-forming law of protein at the air-liquid interface, Hua Jian (Hua

2003) found that the surface pressure will gradually increase with the increase of pH value. And at isoelectric point nearly, the both of the interface activity and the ability to reduce surface tension are also inferior, and the surface pressure is small. This may be because the change of pH of the system will lead to the change of charge, which will affect its stability. Dong Lei (Dong *et al*. 2014) et al. found that the change of charge density can inhibit the formation of a tight adsorption layer at the air-liquid interface of protein molecules and the increase of surface pressure and expansion modulus. In addition, the change of pH value of mucus will also affect the viscosity of mucus, and then change the activity of air-liquid interface. For example, some people believe that Helicobacter pylori will not only increase the pH value of mucus, but also reduce the viscoelasticity of mucus and increase the mobility of mucus. It also can reduce the activity of the air-liquid interface.

5.3 Effect of Ions on Interfacial Activity

Ionic strength affects the adsorption kinetics of particles at the air-liquid interface, and then affects the interfacial activity. Wang Mei (Wang *et al*. 2020) et al. analyzed the change of air-liquid interface pressure under the strength of Na^+ and Ca^{2+} at pH 7. They found that the greater the ionic strength, the faster the surface pressure rises. This may be because ions make the protein molecular structure more loose and it is easier to adsorb to the air-liquid interface, leading to the interfacial tension and increasing the surface pressure. Moreover, the increase of ionic strength will also enhance the flexibility of protein. The hydrophobic groups of protein molecules are more likely to be exposed to the air-liquid interface, it can reduces the tension of the air-liquid interface and accelerates the rate of expansion and rearrangement. Zhang Xuan (Zhang *et al*. 2020) et al. found that the surface tension equilibrium value decreased with the increase of Ca^{2+} and Na^+ content. It is mainly caused by the increase of adsorption capacity of nanoparticles at the air-liquid interface. At the air-liquid interface, the electrostatic repulsion between particles is inversely proportional to the salt content. Meanwhile, the negative repulsion between particles and air-liquid interface will also decrease with the increase of salt content, resulting in the increase of particle adsorption density at the interface. In addition, studies have shown that the contact angle of particles will increase with the increase of ion content (Wang *et al*. 2021), it makes the surface tension decrease more obviously.

5.4 Effect of Antioxidants on Interfacial Activity

At present, it has been found that a variety of experimental intestinal mucosal injury and intestinal diseases are related to the participation of aerobic free radicals (Qian, et al. 2017). Lipid peroxidation and the decrease of antioxidant capacity lead to the imbalance between oxidation and antioxidant, the increase of surface pressure and the damage of small intestinal mucosa. Antioxidants can protect the function of intestinal mucosal barrier by increasing the activity of intestinal antioxidant enzymes, scavenging oxygen free radicals, improving the state of oxidative stress, improving tissue antioxidant capacity and changing the activity of air-liquid interface.

6 CONCLUSIONS

In recent years, with the development of science and technology, air-liquid interfacial activity has long been not only used to explain natural science and applied in food industry, industry, manufacturing and other production and processing. In terms of body health evaluation, the air-liquid interface also plays an important role. Although there are a few reports on the formation of air-liquid interface, the function of air-liquid interface activity in digestive tract and respiratory tract and the factors affecting interface activity, there are still many outstanding problems. For example, the mechanism of air-liquid interface activity regulating body health and the influence mechanism of surfactant on interface activity are not clear. However, based on the available information of air-liquid interface activity, it can be determined that air-liquid interface activity affects physiological functions such as environmental stability of tissues and organs, microecological balance and nutrient absorption and utilization. It is hoped that with the gradual deepening of people's research, the air-liquid interface activity will provide unlimited possibilities for the treatment of related diseases. And the idea that the activity of air-liquid interface in vivo can be adjusted by oral administration of exogenous surfactants can be realized as soon as possible. At the same time, the study of air-liquid interface also provides multi angle demonstration for the functional food to be developed.

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