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Immunization via the anal mucosa and adjacent skin to protect against respiratory virus infections and allergic rhinitis: A hypothesis

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SUMMARY

Exposure of the immune system to environmental antigens and infectious agents by way of the anal mucosa and perianal skin could play an important role in protecting the respiratory tract against allergic conditions and virus infections. Hygienic practices that have reduced exposure of the immune system to such agents include the use of modern toiletry, disposable diapers and clothes dryers. Historically, the anal region was cleansed following defecation with natural materials that would have brought antigens and infectious agents from the environment into frequent contact with the perianal skin and anal mucosa. This practice was a crude form of transcutaneous and mucosal vaccination, whereby antigenic agents that are topically applied to skin or mucosal surfaces, penetrate into the tissues and stimulate immune responses that can extend to the respiratory tract. Furthermore, until the 1960s, diapers and other cloth items were often dried outdoors where they would have collected environmental antigens that, when applied to the body, could have made contact with the immune system in the skin. Herein, it is hypothesized that prevention of allergic rhinitis and possibly other disorders involving the immune system could be achieved by the daily application of preparations composed of environmental antigens and infectious agents to the anal mucosa and adjacent skin. In support of the proposal, immunotherapy for allergic rhinitis currently involves administration of specific allergens to subcutaneous tissue or to the sublingual mucosa. It is considered that superior protection could be achieved by applying the allergens to the anal region where they would target the immune system in both mucosal tissue and adjacent skin. It is also hypothesized that respiratory viruses applied to the anal region would infect tissues at that site and induce immune responses that would protect the respiratory tract against the common cold and influenza. This approach is supported by evidence that orally administered adenovirus vaccine can induce an infection in the intestinal mucosa that stimulates immunity to protect the respiratory tract. Although other respiratory viruses are unlikely to survive passage through the intestinal tract, rhinovirus has on rare occasion been detected in stool specimens, suggesting the possibility of an infection at the terminal end of the digestive tract. Respiratory syncytial viruses and influenza viruses are amenable to modification by reverse genetics and other techniques and it is expected that natural or modified viruses applied to the anal region could serve to immunize the respiratory tract.

Introduction

A review by Chang and Pan [1] associates the emergence and rising prevalence of allergic diseases during the past two centuries with environmental and lifestyle changes that have occurred as wealth in societies has increased. The "hygiene hypothesis" relates this phenomenon to a decreasing incidence of childhood infections [2]. Chang and Pan [1] broadened the hypothesis by suggesting that to protect against allergic disease, the immune system would require frequent stimuli from environmental antigens, as well as from microbial infections, and that the exposure should continue beyond childhood. They defined environmental antigens as "those otherwise innocuous antigens that come in contact with the mucosal surfaces, particularly those of the airway, gastrointestinal tract and eyes of people and animals and that can potentially induce immunological responses". Environmental antigens would include allergens such as pollens, dust mites, moulds and animal dander.

Insufficient stimuli of the immune system by environmental antigens, bacteria and parasites may lead to abnormal responses of the immune system [3–6]. These abnormal responses may result in sensitization of the respiratory tract to otherwise innocuous antigens. This sensitization could lead to production of allergen-specific immunoglobulin E (IgE) that could attach to mast cells in tissues and to basophils in the circulation. Allergens could then interact with the IgE in a way that would cause the cells to release histamine and other pharmacological mediators with resultant symptoms of allergic rhinitis [1,7].

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Chang and Pan [1] consider that the environment and not genes is often the dominant factor in determining whether or not an individual develops an allergy. This suggestion is consistent with reports that after the fall of the communist systems in Europe, the prevalence of hay fever, asthma and airway hyper-responsiveness was significantly higher in children aged 9–11 in West Germany than in East Germany [6]. Follow-up studies have shown that the incidence of allergic disease has increased in East Germany as the population has adopted "western lifestyles" [1,3].

It has not been determined what natural routes of exposure to microorganisms and environmental antigens would be most effective in stimulating immune responses that would protect against allergic rhinitis. Infectious agents and antigens that enter the body via the respiratory tract may cause disease or may sensitize the tissues, leading to allergic conditions. Exposure via the oral route may lead to killing or denaturising of the agents in the digestive tract without stimulating an immune response. Alternatively, the antigens may induce a state of immune tolerance [8–10]. Current immunotherapy practices that may give relief from allergic rhinitis consist of a series of injections of allergen into subcutaneous tissues [7] or application of preparations containing allergen to the sublingual mucosa [11].

Although the common cold is not usually considered life threatening, it is responsible for a significant portion of the days that children miss school and that adults are absent from work [12–16]. Furthermore, the common cold may exacerbate or precipitate allergic conditions affecting the respiratory tract and may predispose to bacterial sinusitis, ear infections and other disease conditions in school-age children [13]. In the early stages of a cold, the nasal discharge may contain viruses that are easily spread by sneezing, nose blowing and nose wiping [14]. Viruses that cause cold-like symptoms include: rhinovirus, coronavirus, respiratory syncytial virus, adenovirus and influenza virus.

Rhinoviruses have been estimated to cause about 50% of all cases of the common cold and may be the most common cause of acute infectious illness in humans [14]. Symptoms typical of the common cold are frequently confined to the upper respiratory tract and may occur 2 or 3 days following infection, with full recovery within 1 to 3 weeks. It has not been considered feasible to develop vaccines against the infection since many different serotypes of the virus may coexist in a population and immunity may be short-lived [13,16]. Turner and Couch [14] consider that "developing effective control of rhinovirus infections constitutes the most important need for future disease-related research".

Coronaviruses are also a common cause of mild to moderate upper respiratory illness, and natural infections do not induce levels of immunity that protect against re-infection or disease [16,17]. The respiratory syncytial virus is a serious pediatric respiratory disease and almost all children become infected during the first few years of life. However, the virus infects individuals of all ages and is an important cause of lower respiratory tract infections. The need for a vaccine to protect against the respiratory syncytial virus has been given high priority [15].

Influenza epidemics continue to be a major cause of morbidity and mortality worldwide and rapid transmission of the viruses can occur in schools and other places where large numbers of people are in close contact. Killed vaccine has been widely used to protect against influenza and formulation of the vaccine is changed each year to incorporate serotypes of virus that would be most likely to circulate in the environment [12,18]. However, the potential risk of adverse reactions has resulted in lower than expected acceptance of these vaccines by health care workers in hospitals [19]. Live attenuated influenza virus vaccines have been developed for administration into the nasal passages but have had limited use [16,18,20].

Most respiratory viruses will not survive passage through the digestive tract but an orally administered adenovirus vaccine has, in the past, been administered to US military recruits. The vaccine caused an infection in the intestinal tract that stimulated immunity to protect the respiratory tract [21]. These findings, along with those from allergen-specific immunotherapy [7,11], give evidence that induction of immune responses in the digestive tract mucosa and in skin can lead to protection of the respiratory tract. The focus of this hypothesis is that immunization against respiratory virus infections and allergic rhinitis could be achieved by topical application of antigenic agents to the anal region, consisting of the anal mucosa and adjacent skin.

Background to the hypothesis

The pioneering studies on vaccine development for the poultry industry in the 1930s included an innovative approach that might be applied for immunizing humans. It was shown that chickens could be protected against a viral disease with a tropism for the larynx and trachea by applying vaccine to the cloacal mucosa, located at the terminal end of the digestive tract. Initially the vaccine consisted of mucous taken from the respiratory tract of diseased chickens that would have contained the causative herpesvirus [22]. This experience suggests that the virus in secretions collected from the human respiratory tract in the early stages of a common cold, might serve as a crude vaccine if applied to the anal region. Having considered this, a healthy adult male, referred to herein as "the principal", collected secretions from his respiratory tract on tissue paper and applied this to his anal region. This became a routine daily practice and was carried out for 4 years. The principal followed this practice with the understanding that low quantities of respiratory virus may occasionally be found in the secretions in the absence of respiratory symptoms [16]. Secretions from the respiratory tract would also contain environmental antigens and a variety of bacteria, including potential pathogens. During the 4 year trial period, the principal experienced no ill effects that could be attributed to this practice. To the contrary, he experienced a marked diminution in problems attributed to the common cold. Unexpectedly, he also experienced a marked reduction in problems due to seasonal allergic rhinitis. These observations suggested the possibility that respiratory viruses, environmental antigens and other antigenic agents in the respiratory secretions applied to the anal region had stimulated immune responses in tissues at the site of application that had protected the respiratory tract. However, the principal's observations do not have scientific validity. Other factors, of which he was unaware, may have contributed to his improved health. Nevertheless, by applying respiratory secretions containing antigenic agents to the perianal skin and anal mucosa, the principal was performing both transcutaneous [23,24] and mucosal immunization [25,26]. These forms of vaccination stimulate immune responses that can extend to the respiratory tract.

The hypothesis

From man's beginning until the 19th century, it was common practice to cleanse the anal region following defecation with vegetation, stones and other natural materials that would have been contaminated with environmental antigens and infectious agents. This practice was a crude form of transcutaneous and mucosal vaccination and would have brought antigens from the environment into contact with the immune defences in both the perianal skin and anal mucosa. Today, in developed countries, this natural form of exposure to antigenic agents from the environment has almost been eliminated by the move to indoor toiletry, where toilet paper

and purified water are used for cleansing purposes. Also, until the 1960s, it was common practice in North America to use cloth diapers and to dry these and other clothing, towels and bedding on lines outdoors where they would have collected a variety of environmental allergens and infectious agents. Thus, from birth the skin was exposed to antigenic agents that could have penetrated into the tissue and elicited immune responses. The increased use of disposable diapers and clothes dryers since the 1960s has also reduced exposure of the skin to antigenic agents from the environment.

Mazmanian et al. [27] propose that "the mammalian genome does not encode for all functions required for immunological development but rather that mammals depend on critical interactions with their microbiome (the collective genomes of their microbiota) for health". In accord with this proposal, others have noted that the changed or reduced exposure of the immune system to microorganisms and environmental antigens may result in disordered regulation of the immune system with resultant health problems that include: allergic and autoimmune diseases, inflammatory bowel disease, some forms of cancer, type 1 diabetes and multiple sclerosis. In light of the changes in hygienic practices since the 1960s, it is noteworthy that the prevalence of these health conditions has greatly increased during the past 50 years [4,5,27,28].

The anal region is well suited as a safe site for topical application of vaccines because it is a contained anatomic space where antigens and infectious agents could stimulate the immune system in both the anal mucosa and adjacent skin. Studies on transcutaneous immunization support this claim and Glenn et al. [23] consider that "the skin represents an ideal route to the immune system: by traveling the microbial footpath of the multiple, repeated daily challenges on the surface of the skin, vaccines can achieve effective and robust immune responses". It has been shown that disruption and wetting of the skin's protective stratum corneum can greatly facilitate the passage of antigens and infectious agents into the epidermis where they can be trapped by dendritic Langerhans cells and transported to lymph nodes for presentation to T cells. The chain of events could lead to both cellular and humoral immune responses [23,24]. Adjuvants, such as bacterial toxins, may be applied to the skin along with antigens and can greatly augment the immune response [23,24]. The perianal skin should be well suited for the uptake and response to foreign antigens as the stratum corneum is always moist and is frequently disrupted during cleaning. Furthermore, the stratum corneum would be covered with bacterial products that may have an adjuvant effect on the applied vaccines. Concerning the role for mucosal immunization, Seipp [25] states that "if an immune response is generated in the gastrointestinal lining, T cells produced there can travel to other mucosal sites, for example, the lungs or nasal cavity, providing protection over a large surface area".

Immunization against allergic rhinitis

Various reports suggest that beginning early in life, the immune system requires frequent stimulation by a broad range of microbes and environmental antigens in order to prevent disordered regulation that could lead to development of allergic conditions, including allergic rhinitis [1,3,6,28]. To achieve this, it is proposed that preparations composed of microbes and antigens be applied daily to the anal mucosa and adjacent skin, beginning soon after birth. This would simulate exposure of the body to the microbes and antigens in the environment as it occurred prior to implementation of modern hygienic practices.

The preparations applied to the anal region could contain microbes, components of microbes and antigens found in the respiratory tract. Sources for the desired broad range of antigenic agents could include soil, natural water and air in various seasons [1,28]. Specific organisms that may have an anti-allergic effect, such as the environmental saprophyte *Mycobacterium vaccae*, could be added to vaccines [29]. In addition, components of pathogens like *Mycobacterium tuberculosis*, or of parasites, including helminths or *Toxoplasma gondii*, could be included in vaccines, since infections or infestations with the live organisms are believed to stimulate immune responses that could inhibit the development of allergic immune responses [1,6].

To provide immunotherapy for persons suffering from allergic rhinitis, specific allergens could be applied to the anal region. This would be a non-invasive approach and many allergens could be combined into a single vaccine. These allergens would require less purification than those intended for administration by subcutaneous injection [7] or by application to the sublingual mucosa [11] since pollens, dust mites, animal dander and other primary sources of allergen could be broken apart in the anal mucosal fluids, thus exposing the allergens [1]. This approach to immunotherapy would be less likely to cause adverse reactions than would preparations of purified allergens administered by injection into subcutaneous tissues [30]. To maintain protection, frequent application of specific allergens throughout life may be necessary and this could probably be achieved in a more convenient and acceptable manner via the anal mucosa rather than via the sublingual mucosa. While subcutaneous immunotherapy has been practiced for many years, the risk to benefit ratio has not always been favourable [30] and immunity may only endure for one year [7]. Nevertheless, the success from both subcutaneous and sublingual immunotherapy in relieving symptoms of allergic rhinitis [7,11], support the expectation that effective immunotherapy against this condition could be achieved by topical application of allergens to the anal mucosa and adjacent skin.

Immunization against respiratory infections

It is proposed that live and fully virulent respiratory viruses applied to the anal region would establish localized infections in the anal mucosa and adjacent skin and thereby elicit immune responses that could protect the respiratory tract. While some of these viruses may readily propagate in tissues of the anal region, others may require modification and adaptation [15,31]. The use of natural viruses as vaccines would be a departure from current vaccination practices, since "apart from vaccinia, no other completely natural organism has ever come into standard use" [8].

The feasibility of stimulating immunity in the digestive tract mucosa to protect the respiratory tract is supported by findings from oral adenovirus vaccination programs [21]. However, most respiratory viruses would not be expected to survive passage through the digestive tract and to date there is no data to support the hypothesis that common cold viruses or influenza viruses, in a natural or altered state, could infect the anal mucosa or perianal skin. Yet, there appears to be no data to the contrary. In a study by Cate et al. [31], rhinovirus that was inoculated into the duodenum of volunteers, failed to survive passage through the intestinal tract. This failure was attributed in part to body temperature, as viruses of this group replicate best in the upper respiratory tract at temperatures of about 33 °C. However, in the same study, a volunteer that was infected with rhinovirus via the respiratory tract developed respiratory symptoms. An unexplained finding was that the virus was detected in a stool specimen but not in the rectal mucosa of that volunteer. Although speculative, it is proposed that the tissue paper used by the volunteer to capture his respiratory secretions contained virus that was inadvertently applied to his anal region, resulting in an infection at that site. This would explain why the quantity of virus detected in the stool specimen exceeded what

would occur from mechanical contamination. Conditions may be more favourable for growth of respiratory viruses at the terminal end rather than in more proximal portions of the digestive tract. This expectation gives a rationale for applying vaccines to the anal region.

Most respiratory viruses that cause the common cold can be propagated in cell culture systems [13–15], and these could be incorporated into vaccines. Influenza viruses for vaccine purposes are usually propagated in embryonated chicken eggs [18]. A major deterrent to the development of vaccines against the common cold has been the large number of viruses that can circulate in a population at the same time [13]. However, a live virus vaccine applied to the anal region could be composed of a mixture of many groups and strains of virus. Reverse genetics techniques have increased the possibility of rapidly constructing strains of respiratory syncytial virus or of influenza viruses that could have potential as live virus vaccines to be administered to the nasal mucosa [15,18,32]. It is expected that the same techniques could be used to construct viruses that could propagate and stimulate immune responses in the anal mucosa. The respiratory syncytial virus tends to be cell-associated, making it difficult to obtain high titres of purified cell-free virus [15]. However, topical administration of cellular debris containing virus to the anal mucosa should not pose a problem. Immunity induced by viruses in the respiratory tract may be short-lived, whereas the frequent self-administration of viruses to the anal region should make it possible to stimulate and maintain levels of immunity required for protection. A safety feature in applying respiratory viruses to the anal region of infants is that the area would be covered by a diaper. Also, the respiratory syncytial virus is noted for its instability [15].

Although mucosal vaccination via the nasal route can be successful [20], adverse reactions include the increased possibility of developing reactive airway disease [18] and the chance of an infection passing from the nasal mucosa to the brain through the cribriform plate [8]. In contrast, respiratory viruses applied to the anal region would be expected to remain localized and would be less likely to have undesirable side effects. Killed vaccines require much larger quantities of virus than live vaccines. In the case of influenza virus, 10 billion virus particles produced in a single embryonated chicken egg may constitute a single dose of the vaccine [18]. In comparison, the amount of virus produced in a single egg may be sufficient to vaccinate thousands of people with a live virus vaccine. In the face of an influenza pandemic, there would be an urgent need for safe vaccines that could be applied to prevent widespread morbidity and mortality [32]. Production of live virus vaccines that could be self-applied to the anal region may make this possible. In addition, vaccine applied to the anal region may have therapeutic value in the case of persons suffering from an acute respiratory infection.

Vaccines applied to the anal region may play a dual role in protecting against disease. For example, incorporation of *Streptococcus pneumoniae* or its components into a vaccine intended for immunization against allergic rhinitis, may protect the respiratory tract against infection by this organism [12]. The immune responses induced by vaccines intended to protect against respiratory virus infections, may reduce susceptibility to certain allergic disorders [1,2,13]. While special procedures would be required to assure safe handling of vaccines composed of live microorganisms, most of these infectious agents are ubiquitous and appropriate hand washing should prevent the mechanical spread of infection. It is expected that vaccines administered via the anal route could have advantages over current methods for preventing or treating respiratory virus infections and allergic rhinitis in terms of safety, economics, convenience and efficacy.

Conclusions

It should not be difficult to conduct properly controlled studies to determine the validity of the approach to immunization proposed herein. Replication and survival of viruses applied to anal region could be assessed by polymerase chain reaction technology and the immune response to the viruses and various antigens could be monitored. Studies lasting for several years should be sufficient to assess the potential for protection against respiratory virus infections and allergic rhinitis. However, long term studies would be required to establish the full benefits to health that could be achieved from daily exposure of the immune system to immunogenic preparations, topically applied to the anal region. This approach to disease prevention would complement and not compromise hygienic measures that have been shown to be effective in preventing the spread of disease. This is a hypothesis for hygienic self-immunization that would use microbes and other antigenic agents from the environment to benefit human health.

Conflicts of interest statement

None declared.

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