ARQUIVOS DE ASMA, ALERGIA E IMUNOLOGIA

Review Article

New perspectives in immunotherapy: the importance of dendritic cells in allergen-specific immunotherapy

Novas perspectivas em imunoterapia: a importância das células dendríticas na imunoterapia alérgeno-específica

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ABSTRACT

Allergen-specific immunotherapy is the only treatment capable of altering the natural course of allergic disease. Clinical trials have shown that immunotherapy is safe and effective for many patients. However, it still faces problems related to efficacy, safety, long treatment duration and poor patient compliance. In this context, there has been intense research into the development of adjuvant treatments that increase safety, optimize treatment regimens, and improve patient compliance. Allergens were modified (glycoconjugated) with carbohydrates derived from *Saccharomyces cerevisae* to increase their uptake and presentation through carbohydrate receptors in dendritic cells, benefiting from their ability to induce tolerance and initiate immune response. In light of the new evidence, these cells are a key therapeutic target for adequate response to allergen-specific immunotherapy and can drive innovation in the field of immunotherapy.

Keywords: Allergen-specific immunotherapy, allergy, dendritic cells.

RESUMO

A imunoterapia alérgeno-específica é o único tratamento capaz de alterar o curso natural da doença alérgica. Ensaios clínicos mostram que a imunoterapia é segura e eficaz para muitos pacientes. No entanto, ainda enfrenta problemas relacionados à eficácia, segurança, longa duração do tratamento e baixa adesão dos pacientes. Neste contexto, tem havido intensa pesquisa no desenvolvimento de adjuvantes com objetivo de aumentar a segurança, otimizar os esquemas de tratamento e melhorar a adesão dos pacientes. Alérgenos foram modificados (glicoconjugados) com carboidratos derivados de *Saccharomyces cerevisae* para aumentar sua captação e apresentação através dos receptores de carboidratos presentes nas células dendríticas, beneficiando-se da capacidade de atuarem na indução de tolerância para iniciar respostas imunes. À luz de novas evidências, essas células constituem alvo terapêutico chave para se obter uma resposta adequada à imunoterapia alérgeno-específica, com potencial de contribuição na inovação do campo da Imunoterapia.

Descritores: Imunoterapia alérgeno-específica, alergia, células dendríticas.

Introduction

Although allergic diseases may be controlled with symptomatic or emergency treatment, allergen-specific immunotherapy is the only curative treatment option with proven efficacy and safety described by several studies and meta-analyses.[1,2]

However, some limiting factors include long treatment durations, costs, poor patient adherence,

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and the risk of serious, life-threatening adverse reactions. The development of immunotherapy with modified allergens with increased antigenicity and decreased allergenicity, in combination with novel adjuvant molecules via new routes, may shorten treatment durations and possibly reduce these disadvantages.[3]

Mechanisms of allergen-specific immunotherapy

Contrary to what was previously believed, the shift from Th2 to Th1 immune response is not the key to successful treatment. Recent advances in the knowledge of regulatory T (Treg) and B cells and peripheral tolerance mechanisms were essential to explain immune alterations resulting from immunotherapy.

Immunotherapy was believed for many years to induce a shift from Th2 to Th1 immune response by reducing the levels of inflammatory cytokines (IL)-4, IL-5, and IL-13 and, consequently, increasing the levels of interferon-γ. However, this theory does not completely explain why patients undergoing immunotherapy do not have a higher incidence of diseases related to the Th1 lymphocyte population.[4]

The first studies demonstrating the role of Tregs in the mechanism of allergen-specific immunotherapy were published in 2004. Since then, immune tolerance induction has become the main target in the prevention and treatment of diseases related to immune system dysfunctions, such as allergies.[5]

Cell subsets with regulatory capabilities are induced during allergen-specific immunotherapy. IL-10 and transforming growth factor β (TGF-β) are the main suppressor cytokines, in addition to surface molecules such as cytotoxic T lymphocyte-associated antigen-4 (CTLA-4) and programmed cell death protein-1 (PD-1) within the microenvironment. Modified T- and B-cell responses and antibody isotypes, increased activity thresholds for eosinophils, basophils and mast cells, and consequent limitation of inflammatory cascades induce and maintain a state of sustained allergen-specific unresponsiveness. Established tolerance is reflected on clinical perspectives as improvement of allergy symptoms together with reduced medication requirements and progression of disease severity.[5]

New adjuvants

Allergen-specific immunotherapy is the only treatment capable of altering the natural course of allergic disease. Although clinical trials have shown that immunotherapy is safe and effective for many patients, it has some limitations.

Although it has been evolving for more than 100 years, immunotherapy with allergen extracts is often inconvenient for patients due to disadvantages such as long-lasting treatment regimens and concerns about efficacy, treatment safety, and longevity of induced effects. For some allergies, immunotherapy is still only partially effective and may be hampered by undesirable side effects. Therefore, many research projects aim to improve immunotherapy by creating new vaccine candidates and adjuvants that increase efficacy while decreasing undesirable adverse effects.[6]

An extensive literature review found recent publications reporting new and innovative approaches aimed at increasing safety, maintaining or even increasing efficacy, and improving treatment regimens in allergen immunotherapy. To increase the effectiveness of immunotherapy, allergens were coupled to immunostimulants, and new adjuvants were introduced. Allergens were modified to increase their uptake and presentation. Hypoallergenic molecules were developed to improve the safety profile of vaccines, including peptides derived from allergens, recombinant allergens, receptor agonists and other adjuvants, among which we highlight the new adjuvants obtained from *Saccharomyces cerevisae.*[7,8]

Mannan from *Saccharomyces cerevisiae* is a polysaccharide consisting of mannose residues derived from yeast. The potential of mannan as an adjuvant for the treatment of different diseases was described by studies demonstrating enhanced dendritic cell (DC) maturation and antigen presentation, as well as enhanced immune responses with mannan.[9,10] Carbohydrate conjugation to allergens is a well-described approach to targeting antigen-presenting cells (APCs) and rendering the allergen hypoallergenic.[11]

Mannan (*Saccharomyces cerevisiae*) has been used in several studies for targeting allergens to APCs. Weinberger et al. demonstrated that mannan conjugates were efficiently taken up by DCs *in vivo*, inducing a switch from IgE to IgG production.[11] The strategy of conjugating the antigens (from mites, grass

pollens, etc.) with a carbohydrate source (mannose) extracted from the cell wall of a known yeast called *Saccharomyces cerevisiae*, which is composed of three main structures (mannan, chitin, and glucan) and is well described in the literature.[12]

The targeting of antigens to DCs to increase cellular uptake has the potential to result in more effective and efficient immunotherapy. Antigens coupled to yeast mannan, as a source of mannose, are suitable for this purpose, given that mannose-binding receptors are expressed on these cells.[12]

APC system – DCs – C-type lectin receptors

DCs are the therapeutic target of glycoconjugates that are rich in yeast mannose. These preparations benefit from carbohydrate receptors found in DCs. They are targeted to C-type lectin receptor (CLR) agonists, in which the adjuvant acts both as an immunostimulant and as an antigen delivery vector system – directing allergens to greater uptake by DCs.

Described by Ralph Steinmen, the 2011 Nobel Prize-winning immunologist, DCs are the main professional APCs. They are located in all lymphoid tissue, in primary and secondary lymphoid organs, and in the blood. They are responsible for initiating and maintaining immune responses.[13]

Glycoconjugate antigens (*Saccharomyces cerevisiae*) are suitable vaccines for allergen-specific immunotherapy, with a growing number of important publications, and may be extremely relevant for the development of therapeutic interventions in other diseases related to immune tolerance.[14] They induce potent blocking antibodies and are captured by human DCs much more efficiently than native antigens, which rely on non-integrin-mediated internalization of the mannose receptor and the specific DC. In addition, they activate human DCs to generate functional forkhead box P3 (FOXP3) Treg cells through PD-L1.[14] These adjuvant characteristics may be explained by their ability to activate CLRs, which are pattern recognition receptors normally expressed in DCs.[14]

Glycoconjugate antigens form an antigen-mannan complex that is more easily captured and internalized. Subsequently, in the presence of IL-10, DCs are activated and acquire a tolerogenic phenotype to form a new population of Treg tolerant cells.[15] Glycoconjugation has been shown to promote the generation of tolerogenic DCs capable with ability to induce FOXP3, functional Tregs both *in vivo* and *in vitro*. The presence of alum was shown to impair the tolerogenic properties of allergenic vaccines with glycoconjugate antigens (*Saccharomyces cerevisiae*).[16]

The Mannose receptor – Internalization and activation

Although there is evidence for carbohydrate conjugation to allergens (glycoconjugation) and *Saccharomyces cerevisiae* can be easily found in Brazil, the use of the mannose-rich adjuvant mannan is linked to an international patent (INMUNOTEK, Spain). However, mannose can be obtained from carbohydrate structures on the cell wall of *Saccharomyces cerevisiae*, for which the general principle of antigen glycoconjugation to mannose may be applied.

Saccharomyces cerevisiae (native) preserves its macrostructures (chitin, glucan, and mannan). The cell wall structure of *Saccharomyces cerevisiae* mainly consists of chains of glucose residues and mannoproteins. Despite having a high degree of purity, the β-glucan fraction of yeast has a mannose-rich fraction, with short or long chains with a high concentration of mannan. Thus, glycoconjugation with this fraction of *Saccharomyces cerevisiae* is capable of maintaining the carbohydrate structure intact (mannan, glucose) and its potentially associated tolerogenic properties, with adjuvant capacity for targeted delivery of allergens to DCs.

Differentials, safety, and effects in immunotherapy

The use of polysaccharides in immunotherapy regimens with one of the mannose-rich fractions of *Saccharomyces cerevisiae* was studied by Oliveira & Binnotti.[17] Although the authors considered the practice to be promising, they described it as experimental.

Current studies demonstrate that glycoconjugates (*Saccharomyces cerevisiae*) target DCs via the mannose receptor and the DC-SIGN, increasing allergen uptake, increasing IL-10 production and PD-L1 expression, and promoting the generation of allergen-specific FOXP3 T cells, both *in vitro* and *in vivo*, which are impaired by the conventional adjuvant alum.[16]

A recent literature review showed that these conjugates also reprogram monocyte differentiation and generate tolerogenic DCs through epigenetic and metabolic rewiring. Unprecedented molecular mechanisms have been discovered, through which these glycoconjugates can restore allergen tolerance during allergen-specific immunotherapy.[18]

Conclusions and future perspectives

Although the use of polysaccharides, currently best described as glycoconjugate antigens with mannose from *Saccharomyces cerevisiae*, in immunotherapy regimens has been poorly understood in the past, several international studies currently supporting its benefits, which are superior to that of native antigens, constituting a recent and important evolution in the field of immunotherapy.

The essential role of DCs is highlighted in the literature. After capturing and internalizing antigens, DCs are activated and acquire a tolerogenic phenotype to form T cells – a fundamental mechanism of allergen-specific immunotherapy. Studies have demonstrated the benefits of optimizing and improving the safety of the therapeutic regimen, as well as of increased promotion of Treg tolerant cells.[15]

DC absorption increases bioavailability and absorption, improving the dosing scheme by suppressing the induction phase and leading to increased spacing between applications, with intervals every 5 weeks. Vaccines for pollen and dust mite allergies have already been developed, but the same concept is also being studied for other allergens, including peanuts.[15]

Glycoconjugate antigens (*Saccharomyces cerevisiae*) represent a new generation of allergy vaccines, as they optimize the uptake of allergens by DCs and increase the bioavailability of administered doses while promoting safe immune responses.[15]

The emergence of new evidence elucidates the immunostimulatory activity and the allergen delivery vector system to DCs, potentiated by glycoconjugation of antigens to high mannose adjuvant from *Saccaromyces cerevisae* and its potential contribution to innovation in the field of immunotherapy.

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Conflicts of interest: Maria Angela Vigoritto declares no conflicts of interest. Gustavo Pradez is CEO of IMUNO Center.

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