

7th International Conference on

Allergy, Asthma and Clinical Immunology

September 14-15, 2016 Amsterdam, Netherlands

Bisbenzylisoquinoline alkaloids: New perspectives on anti-allergic drug development

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Allergic diseases are major public health problem worldwide. Allergic asthma is a chronic inflammatory airway disease, characterized by intense eosinophil recruitment and activation and airway hyper-responsiveness (AHR). The anti-allergic therapy includes the use of drugs that cause significant side effects or are not effective under specific conditions, justifying the search for novel, safe and effective drugs. Bisbenzylisoquinoline alkaloids (BBA) are known for their significant biological effects on inflammation and allergy and recent findings placed BBA as inhibitors of calcium influx in many cell types. Our research group have demonstrated that BBA such as warifteine and curine present significant anti-inflammatory and anti-allergic properties in different models, including: Inhibition of eosinophil recruitment and activation; inhibition of mast cell dependent responses; modulation cytokine and lipid mediator production both *in vivo* and *in vitro*; protection against anaphylactic shock in mice; inhibition of mucus production and AHR development; inhibition of and modulation of B-cell functions. Our findings suggest that these effects are due to calcium influx inhibition, resulting in modulation of many steps of leukocyte activation and function. In conclusion, BBA exhibit anti-allergic effects in models of lung, skin and systemic allergy in the absence of significant toxicity and as such have the potential for anti-allergic drug development.

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The use of *Pichia pastoris* in the production of recombinant proteins for the diagnosis of allergic diseases

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Allergic diseases are a major public health problem. Respiratory allergies, mainly allergic rhinitis, asthma and allergic dermatitis are the most common manifestations. Allergy to house dust mites is the most common cause of asthma and allergic rhinitis. Our goal is to determine the prevalence of group 5 allergens from *Dermatophagoides pteronyssinus* (Der p 5) and from *Blomia tropicalis* (Blo t 5), Recombinant proteins produced by the use of *Pichia pasturis*. DNA encoding these proteins has been cloned into cloning and expression vectors, expressed in *Pichia pastoris* SMD 1168. Both allergens Der p r 5 and r Blo t 5 were purified, characterized by mass spectrometry and circular dichroism. IgE reactivity was measured by means of indirect ELISA tested to measure specific IgE levels in sera of 64 asthmatic patients. The results obtained by circular dichroism show that rDer p 5 and rBlo t 5 are helical protein with a predominance of alpha-helical secondary structure. By mass spectrometry it was demonstrated that these two allergens are monomeric under physiological conditions. The recognition rate of rDer p 5 and t 5 rBlo by specific IgE asthmatic patients is 28% and 6%, respectively; the results obtained by indirect ELISA showed that these two proteins pose no cross-reactivity between them. Both species of mite, *Dermatophagoides pteronyssinus* and *Blomia tropicalis* are answered in Algeria, class 5 recombinant proteins of these two species can be used for the diagnosis of allergic diseases and specific immunotherapy.

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