

MEETING ABSTRACT

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Food allergy and anaphylaxis – 2046. Cloning and characterization of the gene for acidic thaumatin-like protein, an important allergen from sapodilla plum (*manilkara zapota*)

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Background

Allergic reactions to sapodilla ingestion are rare; a 21 kD protein was recognized as an allergen in sapodilla extracts by IgE-immunoblot [1]. Further, the allergen was identified as a basic thaumatin-like protein (TLP) by its N-terminal sequence (ATFDIQNNC-) and isoelectric point. The major purpose of this study was to identify additional allergens from this tropical fruit.

Methods

A case of oral allergy syndrome to sapodilla and custard apple was investigated following approval by Institutional Ethics Committee. Sapodilla allergy was confirmed by diagnostic tests (SPT and allergen-specific IgE). Sapodilla proteins were separated on SP-Sepharose by adsorption at pH 4 followed by step elution at pH 5 (SP1), and with increasing NaCl – 0.1 M (SP2) and 0.2 M (SP3). Forward primers and nested reverse primers specific to the SP1 component were designed based on its N-terminal sequence and conserved regions of homologous plant TLPs. PCR was performed using sapodilla (cv. cricket ball) leaf genomic DNA as template.

Results

SPT and allergen-specific IgE were positive. ELISA revealed that IgE from allergic serum recognized two 21 kD proteins – one in the SP1 pool and the other in the SP2 pool; the 21 kD protein in SP2 was identified as basic TLP. The N-terminal sequence of SP1 component was found to be ATFDVVNQCTFTVWAGASPGGGKQL-

which was identified as an additional TLP. Sequence analysis of overlapping PCR products revealed an almost full-length gene (603 bases; GenBank accession JN624813.1) corresponding to acidic TLP (residues 8–207 of sapodilla acidic TLP and lacking the N-terminal 7 residues). Phylogenetic analysis shows that sapodilla acidic TLP is evolutionarily related to the allergenic TLPs from olive and kiwi fruits, all belonging to the order Ericales.

Conclusions

A partial gene coding for sapodilla acidic TLP representing 97% of the mature sequence has been cloned. Sapodilla acidic TLP has weak β -1,3-glucanase activity and is an important allergen belonging to the TLP family of pollen and fruit allergens [2].

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References

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2. Breiteneder H: Thaumatin-like proteins – a new family of pollen and fruit allergens. *Allergy* 2004, **59**:479-481.

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