CAP inhibition in hymenoptera venom allergy

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Abstract

Citation: Savi Eleonora, Peveri Silvia, Montagni Marcello CAP inhibition in hymenoptera venom allergy. **protocols.io** dx.doi.org/10.17504/protocols.io.ht7b6rn
Published: 03 May 2017

Protocol

Step 1.

Annual validation: 17/01/2017

Step 2.

BACKGRUOND A common issue in diagnosis of Hymenoptera venom allergy (HVA) is the occurrence of multiple positive results to the different venoms, mainly due to cross-reactivity between phospholipase A1 for vespids, and hyaluronidase, that may concern all venoms; another important cross-reactivity source is common cross-reactive carbohydrate determinants (CCD)

Step 3.

AIM Provide the laboratory technician with the instructions for the in vitro test

Step 4.

FIGURES INVOLVED laboratory technician and allergologist

Step 5.

ACTIVITY DESCRIPTION

- Eligible patients: CAP-inhibition is mandatory in the correct choice of the venom to be used for VIT in patients with apparent poly-sensitization

- Blood sample: Blood sample is collected in a dry tube with a Separator Gel. No fasting is required before exam.

Step 6.

EXECUTION

Two 100 mcl aliquots of patient’s serum were incubated separately overnight at 4 °C with 200 mcl of two venoms at increasing dilutions (0; 25 mcg; 50 mcg; 100 mcg 200 mcg/ml). Subsequently sIgE

against each of the venoms were determined in the samples prepared as above using ImmunoCap tecnique. The CAP-inhibition test was carried out with a specific programme in UniCap 250 (Thermofisher) with a calibration curve between 0.1 to 100 ku/l sIgE concentrations that allows a good evaluation of low values.The extent of homologous (blockage of venom-sIgE by the same venom) and heterologous (blockage of the venom-sIgE by the other venom) inhibition was computed with the following formula:% inhibition = 100 − [IgE inhibited sample (kU/l) × 100/ IgE anti-venom (kU/l) at zero concentration of venom].A percentage of homologous inhibition >70 % was required to perform heterologous inhibition. The same percentage was considered suggestive of cross-reactivity among venoms. The venom preparations tested were the same used for skin tests

Step 7.

COMPLICATIONS Having a small quantity of blood sample can cause problems in running the test that can not be conducted until the end.

Step 8.

RECOMMENDATIONS Take a test tube full enough of blood.It’s recomanded to use a IgE specific serum concentration below 25-28 ku/l to have a good rate between sIgE concentration and venom concentration. Higher values are adjusted with a suitable serum dilution.