

CCD-Blocking-Solution

A new feature in the AllergyScreen® / AlleisaScreen® system



Many allergens are glycoproteins, which consist of a protein part and one or more glycan chains. Glycan chains are composed of different sugars linked together and are bound via an amino-group (N-glycane) or a hydroxyl-group (O-glycane) to the protein part. N-glycans are particularly immunogen and can induce the production of IgE (anti-CCD IgE).

The IgE antibodies are highly cross-reactive for glycoproteins of plants, insects and molluscs. Therefore, glycan chains are named "cross-reactive carbohydrate determinants" (CCDs). CCDs have been found in allergen extracts of plant or animal origins like tree, weed and grass pollen, vegetables, fruits, seeds and nuts, latex, insect venoms, naile and parasites.

Approximately 25% of allergic patients produce anti-CCD IgE. The sensitisation of patients against CCDs results probably from pollen allergens or insect stings. While the occurrence of CCDs in allergen extracts induce usually no positive results in skin tests, they can lead to false-positive results or increase a positive result in *in vitro* test systems. For this reason, especially in *in vitro* test systems, the possible occurrence of specific anti-CCD IgE must be considered.

The CCD-Blocking-Solution is a mixture of three important plant glycoproteins with different N- and O-glycan chains:

- Bromelain (from pineapple)
- Peroxidase (from horseradish)
- Ascorbat Oxidase (from courgette)



To exclude false positive or increased positive results in our AllergyScreen®/AlleisaScreen® system, a patient serum can be mixed with the new CCD-Blocking-Solution, which leads to an inhibition of such glycan-specific IgE and is an improvement in *in vitro* allergy diagnosis.

Before testing it in our AllergyScreen®/AlleisaScreen® system the patient serum is mixed thoroughly with 10% CCD-Blocking-Solution (e.g. 270 µl serum + 30 µl CCD-Solution) and incubated at room temperature for 30 minutes.

After incubation the CCD-serum mix can be directly used in the AllergyScreen® (250 µl) or AlleisaScreen® (300 µl) assay.



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Example of the use of CCD-Blocking-Solution in the routine:



Panel 30 RespA

| | |
|-------------------------|---|
| Control | |
| Derm. pteron. (d1) | |
| Derm. farinae (d2) | |
| Acarus siro (d70) | |
| Cockroach (i6) | |
| Alder (t2) | + |
| Birch (t3) | |
| Hazel (t4) | |
| Oak (t7) | |
| Mixed grasses (gx) | |
| Rye pollen (g12) | |
| Ragweed (w1) | |
| Mugwort (w6) | |
| Oxeye daisy (w7) | |
| Plantain (w9) | |
| Nettle (w20) | |
| Camomile flowers (w206) | |
| Bee venom (i1) | |
| Wasp venom (i3) | |
| Latex (k28) | |
| Cladosporium herb. (m2) | |
| Alternaria altern. (m6) | |
| Penicillium not. (m1) | |
| Aspergillus fum. (m3) | |
| Cat (e1) | |
| Horse (e3) | |
| Dog (e5) | |
| Guinea Pig (e6) | |
| Rabbit (e82) | |
| Golden hamster (e84) | |



Panel 30 FoodA

| | |
|--------------------------------|--|
| Control | |
| Bromelain (CCD 1) | |
| Horseradish Peroxidase (CCD 2) | |
| Ascorbat Oxidase (CCD 3) | |
| Peanut (f13) | |
| Soy bean (f14) | |
| Hazelnut (f17) | |
| Walnut (f256) | |
| Crab/Shrimp (f23/f24) | |
| Cod fish/Salmon (f3/f41) | |
| Cuttlefish/Mussel (f59/f207) | |
| Pea (f12) | |
| Carrot (f31) | |
| Celery (f85) | |
| Apple (f49) | |
| Orange (f33) | |
| Kiwi (f84) | |
| Potato (f35) | |
| Wheat flour (f4) | |
| Rye flour (f5) | |
| Tomato (f25) | |
| Bell pepper (f218) | |
| Onion (f48) | |
| Mango (f91) | |
| Banana (f92) | |
| Egg white (ff1) | |
| Egg yolk (f75) | |
| Milk (f2) | |
| α -Lactalbumine (f76) | |
| β -Lactoglobuline (f77) | |
| Casein (f78) | |

The left figure shows Panel 30 Resp A (inhalative allergens) worked off with a serum without CCD-Blocking-Solution and after preincubation of the same serum with 10% CCD-Blocking-Solution. The right figure shows the same for Panel 30 Food A (food allergens). The CCD-inhibition reveals that alder, birch, hazel, mugwort and cat are the only relevant inhalative allergens.

In case of the food panel the CCD-inhibition shows that the patient was not sensitized against any of the tested allergens.

The reduction of positive allergen lines after inhibition of a serum with CCD-Blocking-Solution indicates that the positive response is completely or partly due to a reaction of anti-CCD IgE in patients sera with glycoproteins in allergens.

The use of the CCD-Blocking-Solution makes it now possible to distinguish between a "false" glycan-based sensitisation against CCD's or a "true" protein-based sensitisation.



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