

Betula verrucosa may be taken as a marker allergen for tx5 tree mix

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Background: Allergen extract mixes are often used in routine diagnostics. Reflex testing after a positive mix result is poorly performed to identify the responsible allergen. Tx5 contains Hazel and Grey alder extracts being known to have IgE cross-reactivity between them as well as to Common Silver birch. Elm, Willow and Cottonwood extracts are also included in tx5. The aim of the study was to investigate if one tree pollen allergen extract can be used instead of the mix.

Method: 30 serum samples from Northern European routine patients (biobank) were tested simultaneously on the ImmunoCAP tx5 mix (ThermoFisher), the tree pollen extracts included in tx5 and MUXF (cross reactive carbohydrate determinants, CCD). Samples were tested on Alder and Hazel with the IDS Specific IgE assays (formerly Allersys®, Omega Diagnostics). The allergen extract Birch and Bet v 1 were included additionally on both systems. Anti-CCD IgE antibodies were blocked with the inhibitor from HÄMOSAN Life Science Services® GmbH according to the instructions for use

Agreement between allergen mix tx5 and single allergens

	Allergen mix tx5		Sample nr	Alder T002	Birch T003	Hazel T004	rBet v 1 T215	Mix tx5	Alder t2	Birch t3	Hazel t4	rBet v 1 t215	Elm t8	Willow t12	Cottonw t14	MUXF o214
	positive agreement	negative agreement														
Alder	94.4 %	91.7 %	6													
Birch	94.4 %	83.3 %	1													
Hazel	94.4 %	91.7 %	9													
rBet v 1	88.9 %	83.3 %	8													
Alder	100 %	91.7 %	10													
Birch	100 %	83.3 %	3													
Hazel	100 %	91.7 %	7													
Elm	33 %	100 %	5													
Willow	16.7 %	100 %	2													
Cottonwood	16.7 %	100 %	4													
rBet v 1	94.4 %	83.3 %	20													
			21													
			19													
			23													
			22													
			12													
			18													
			26													
			11													
			16													
			24													
			27													
			28													
			25													
			13													
			15													
			14													
			17													
			30													
			29													

Grey shaded, black font: IDS allergy assays; Dark grey shaded, white font: ImmunoCAP allergy assays

Using 0.35 kUa/L as a cut-off 12 samples were negative (red) and 18 samples positive (green) for tx5. 2 samples were positive for MUXF.

Sample 4:

The mix result is negative and the single allergens included in the mix too. The birch allergens (extract and molecule) were positive and the patient suffered from rhinitis.

Sample 21:

The mix result is negative whereas the single allergens for alder, hazel (in tx5) and birch are positive on both platforms. The clinical history confirmed OAS to PR-10 related foods.

Sample 19:

Disagreement between both platforms. MUXF positive sample.

Most of the patient samples have been negative for Elm, Willow and Cottonwood.

Blocking of anti-CCD antibodies converts positive result into negative one

Sample nr	Alder T002	Birch T003	Hazel T004	rBet v 1 T215	Mix tx5	Alder t2	Birch t3	Hazel t4	rBet v 1 t215	Elm t8	Willow t12	Cottonw t14	MUXF o214
19: 1 st run	0.19	0.25	0.10	<0.1	0.85	0.85	1.12	0.90	0.36	1.11	1.04	0.94	0.90
19: 2 nd run	0.13	not tested				0.88	not tested						0.88
19: 2 nd - BLOCK	<0.1	not tested				0.02	not tested						0.01

Grey shaded, black font: IDS allergy assays; Dark grey shaded, white font: ImmunoCAP allergy assays

Conclusion: Positive results for tx5 were due to Alder and Hazel allergens in the mix for this population. There was also a high concordance between tx5 results and the individual Birch assay. SIgE for Elm, Willow, Cottonwood was rarely detected in tx5 positive samples, which might be explained by the low presence of those trees in Belgium. One sample was negative in tx5 but positive for single tree allergen assays across both platforms and the clinical history confirmed that the mix missed the sensitisation to the tree pollen allergens for this patient. It is very likely that any of the 3 single allergens, i.e. alder, birch and hazel could be an alternative for tx5. Worth mentioning that one sample being positive in tx5 and in all single ImmunoCAP assays was negative in all IDS assays. This sample gave a positive result in the MUXF assay, and blocking of anti-CCD IgE antibodies resulted in a negative ImmunoCAP result for the tested allergen suggesting that there might be differences between both platforms relating to interference by CCD.