



# ICA2018

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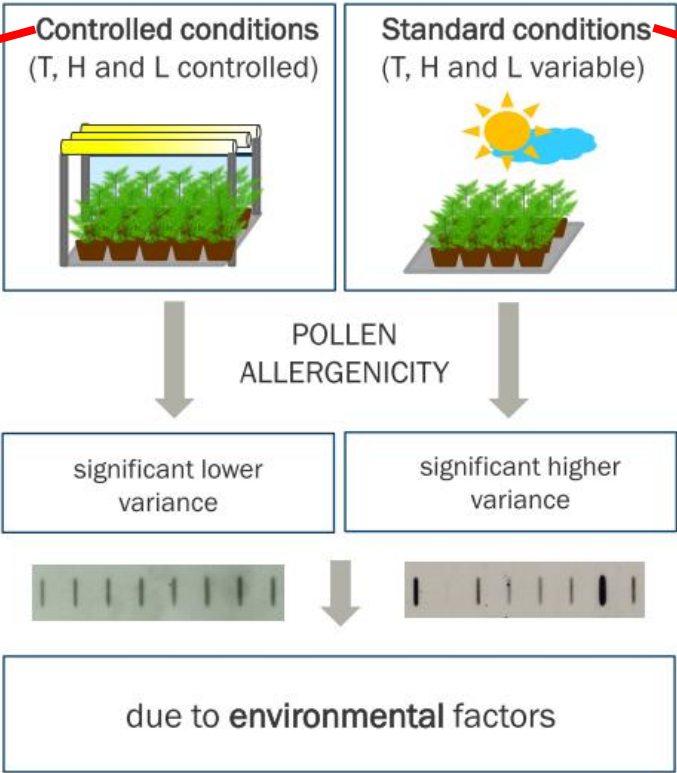


## Is pollen allergenicity a temperature-responsive trait in common ragweed (*A. artemisiifolia* L.)?

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# Previous experiments....



**Pollen allergenicity**  **Epigenetic controlled trait governed by environmental conditions (T, H and L)**

**OPEN** Is ragweed pollen allergenicity governed by environmental conditions during plant growth and flowering?

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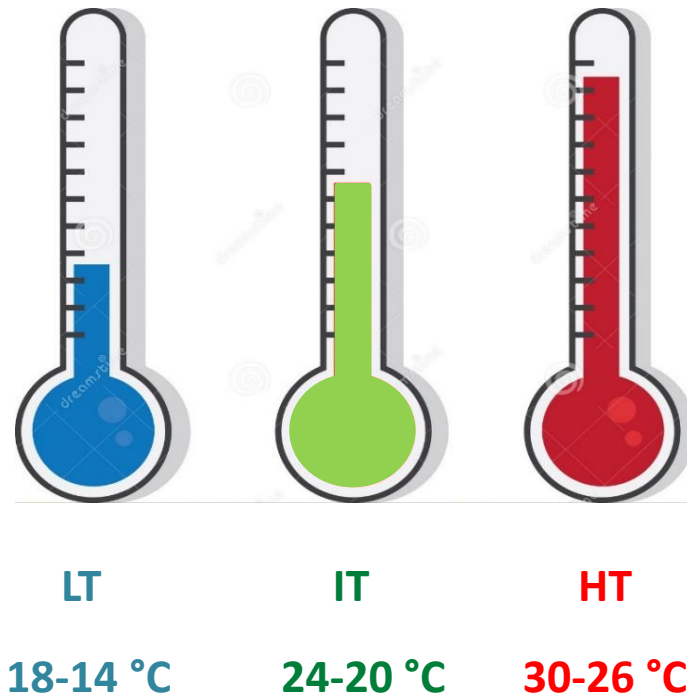
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# Determination of temperature effect: germination and growth conditions

3 growth rooms:

- constant photoperiod (15-9 h light-dark)
- constant intensity of light (Osram Dulux L 36W/840 Lumilux, 2900 lm, total 300-350 LUX).
- 3 thermal regimes:



51 plants per temperature



a total of 153 plants

# Determination of temperature effect: pollen collection

- During flowering:
  - ❖ Male inflorescences were covered with a modified ARASYSTEM to collect pollen grains
- Sampled pollen was stored in 2 ml eppendorf in boxes containing silica gel at room temperature
- Pollen from each single plant was used to obtain soluble protein extracts to determine pollen allergenic potency (by Slot blot, 1D/2D immunoblotting and LC-MS-MS)

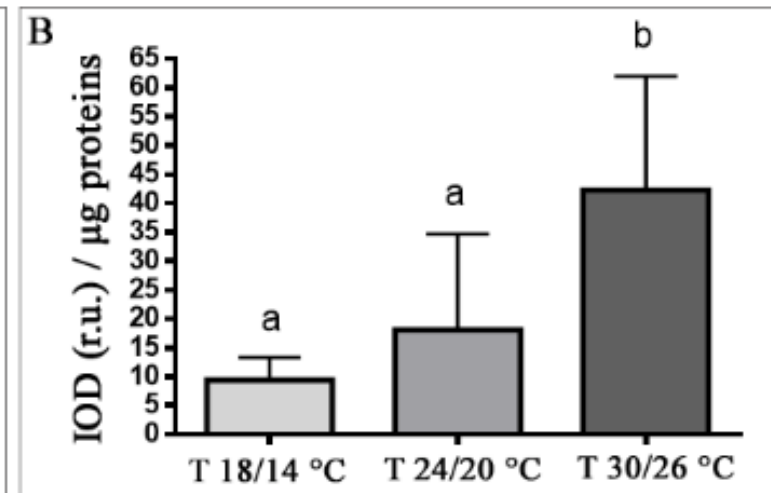
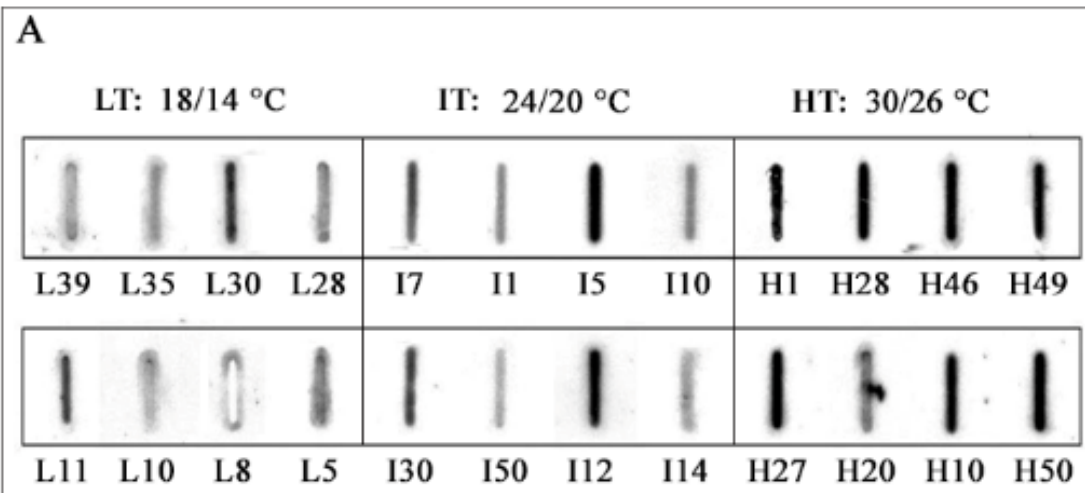


# Determination of temperature effect:

## pollen total allergenicity by Slot blot analysis

### Representative Slot blot membrane

Each spot corresponds to a immunoreactive reaction with a pool of sera (patients sensitized to common ragweed).



highest mean potential allergenicity

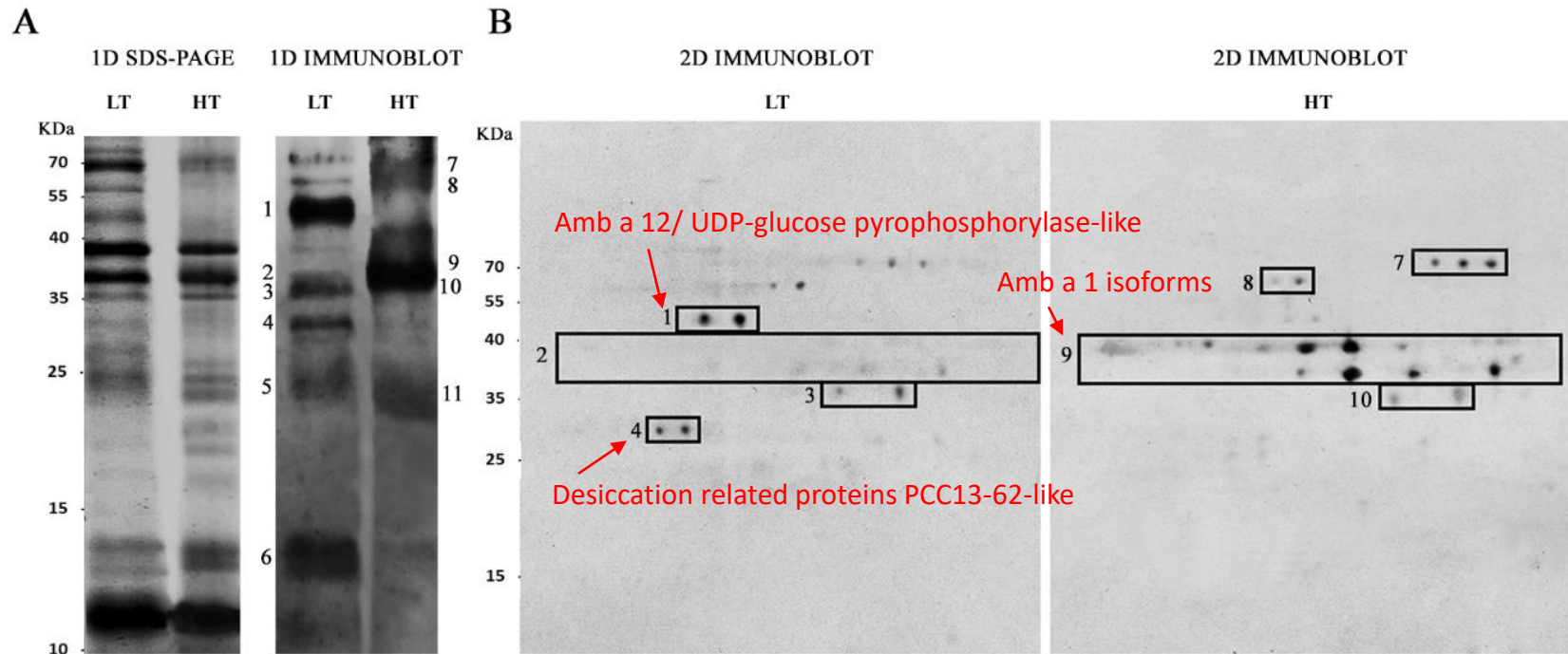


in pollen from plants grown at HT



# Determination of temperature effect:

## what can explain the different total pollen allergenicity? Allergen profile?

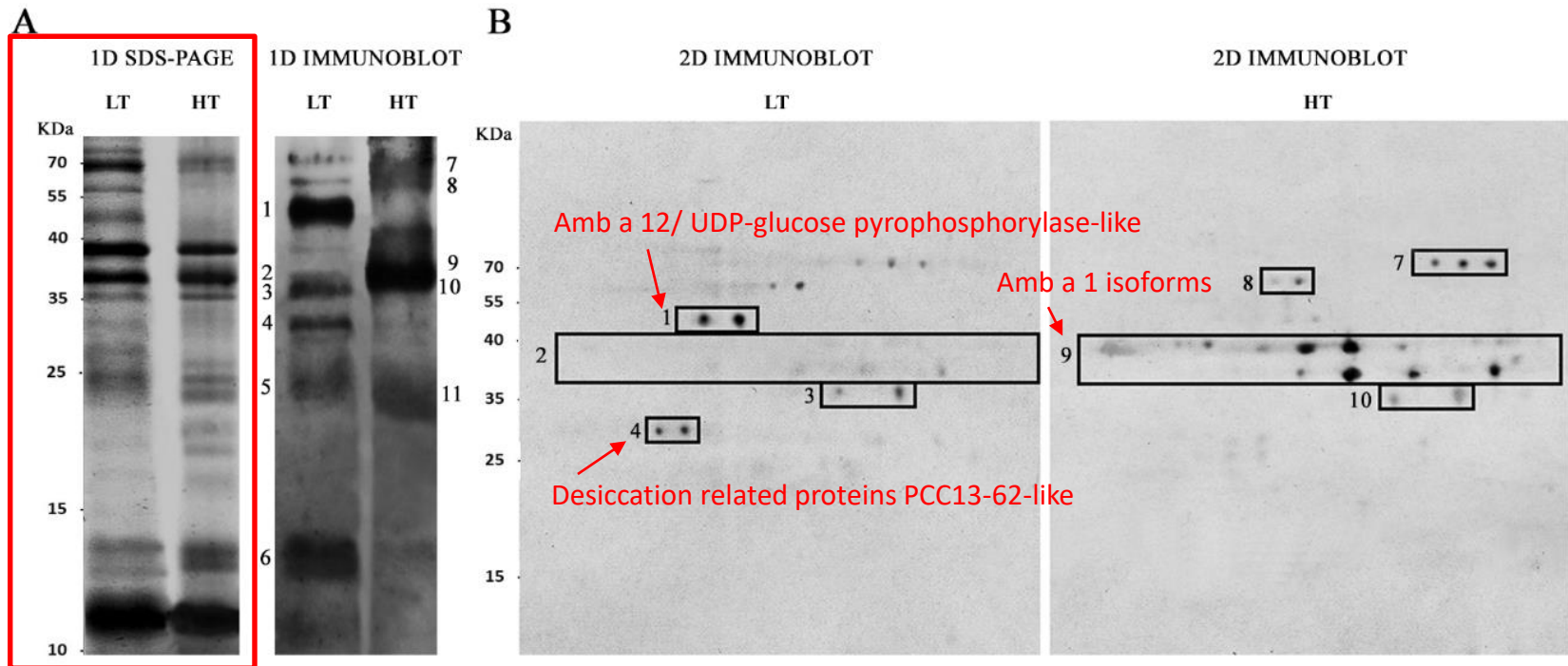


(1) Amb a 12 and UDP-glucose pyrophosphorylase-like, (2) Amb a 1.03, (3) Amb a 11, (4) Desiccation related protein PCC13-62-like, (5) triosephosphate isomerase like protein and Amb a 1.05, (6) Amb a 1 beta chain and Amb a 3, (7) berberine bridge enzyme-like 21, (8) glyoxal oxidase enzyme N-terminus like, (9) Amb a 1 isoforms, (10) Amb a 11, (11) triosephosphate isomerase like protein and Amb a 1.05.

### LT and HT show different allergenic profiles:

- ❖ at HT the allergenicity is mainly related Amb a 1 isoforms .
- ❖ at LT the allergenicity is mainly related to Amb a 12/ UDP-glucose pyrophosphorylase-like and to a desiccation related protein PCC13-62-like

# Determination of temperature effect: what can explain the different allergen profile?



## ➤ Allergen expression

Amb a 12/ UDP-glucose pyrophosphorylase-like and Desiccation related protein PCC13-62 like are only expressed in LT samples

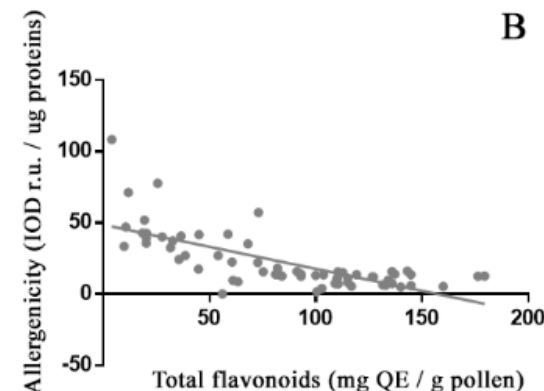
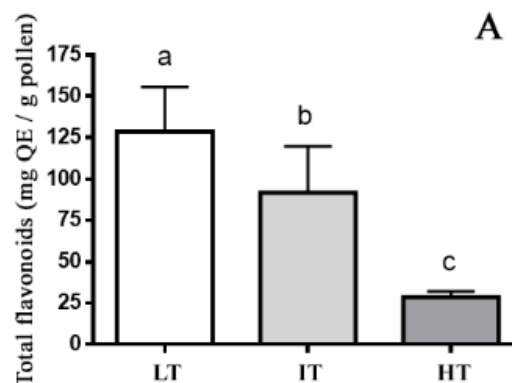
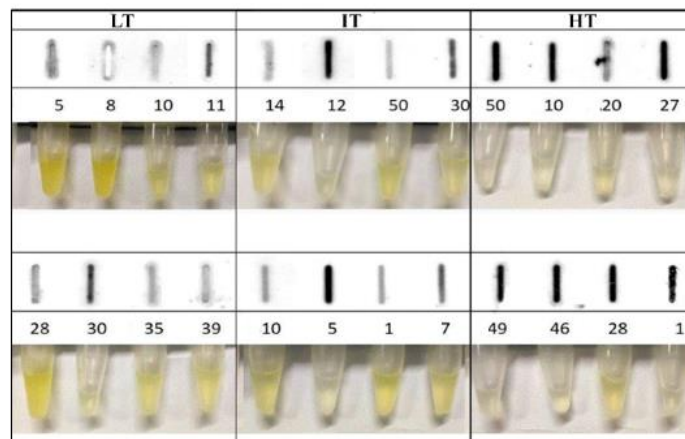
## ➤ Allergen- IgE binding

HT affects Amb a 1- IgE binding

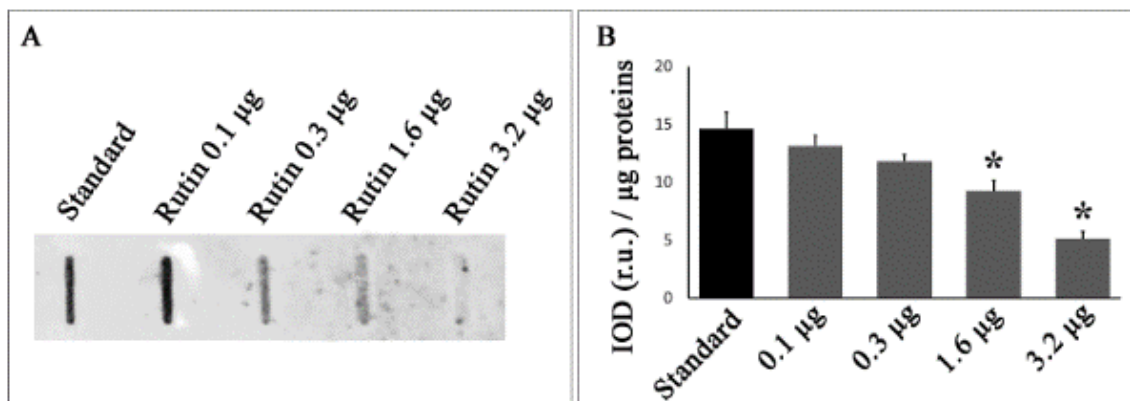
# Determination of temperature effect:

## what can explain the different Amb a 1 – IgE binding? Pollen flavonoids?

- negative relationship between pollen allergenicity and flavonoid content



- Quercetin-type flavonols addition to pollen extracts affects allergenicity



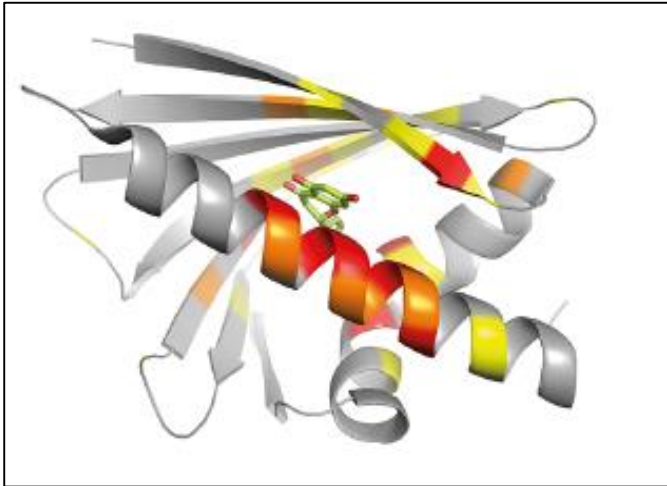
Flavonoids affect the allergen – IgE binding



## Determination of temperature effect:

### what can explain the different Amb a 1 – IgE binding? Pollen flavonoids?

Flavonoids are physiological ligands of PR10 proteins such as Bet v 1 and Fra a allergens



Natural binding between Bet v 1 (major allergen of birch, PR10, pathogenesis-related protein) and quercetin-3-O-sorbose.

Seutter von Loetzen et al. 2014

**However...**

**the relevance of these interactions for allergenicity is still to be determined**



THANK

YOU