Association of T-cell receptor repertoire diversity with L-asparaginase hypersensitivity in childhood acute lymphoblastic leukemia

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Background

Asparaginase is an indispensable component of therapy of acute lymphoblastic leukemia (ALL). Treatment with asparaginase is associated with a plethora of adverse effects, amongst which the occurrence of hypersensitivity is one of the most common. Hypersensitivity affects the ability to maintain dose intensity which in turn results in poorer outcomes, especially if there is no suitable replacement.

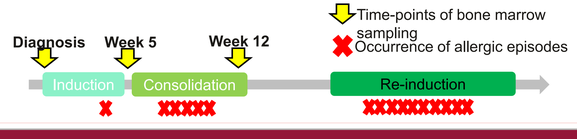
The basis of L-asparaginase hypersensitivity is undoubtedly immune-mediated. T-cell recognition of antigens is arguably one of the most important facets for establishing an immune response, of which a crucial component is the repertoire and diversity of T-cell receptors (TCR). **The exact aspects of T-cell immunity underpinning association with asparaginase allergy has yet to be characterized.**

Objective

To evaluate the associations of TCR repertoire and its diversity with asparaginase hypersensitivity in children with ALL.

Methodology

We longitudinally profiled the TCR repertoires in N=180 samples from 67 children with B-ALL treated on the frontline Ma-Spore ALL 2010 trial, all of whom receive native L-asparaginase in induction, consolidation, and reinduction. Only patients who developed grade 2 or higher reactions (CTCAE 3.0) were included in this study. These children had bone marrow cells sampled at 3 time-points – diagnosis, week 5 (post-induction), and week 12 (post-consolidation). We then evaluated TCR characteristics with clinical features and occurrence of allergy. DNA-based TCR-sequencing was performed on the TCR beta and TCR gamma repertoires using the LymphoTrack TRB and TRG Assay Panels, Invivoscribe; on NextSeq 500 Illumina system using the NextSeq 500/550 Mid Output Kit v2.5 (300 Cycles) with paired end read lengths of 2 × 151 bp.

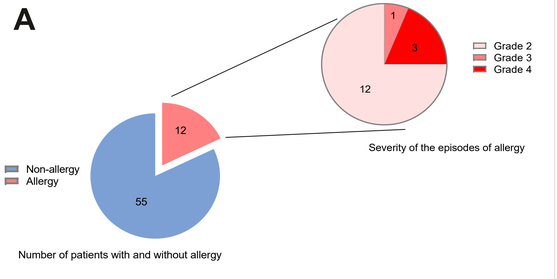


Conclusions

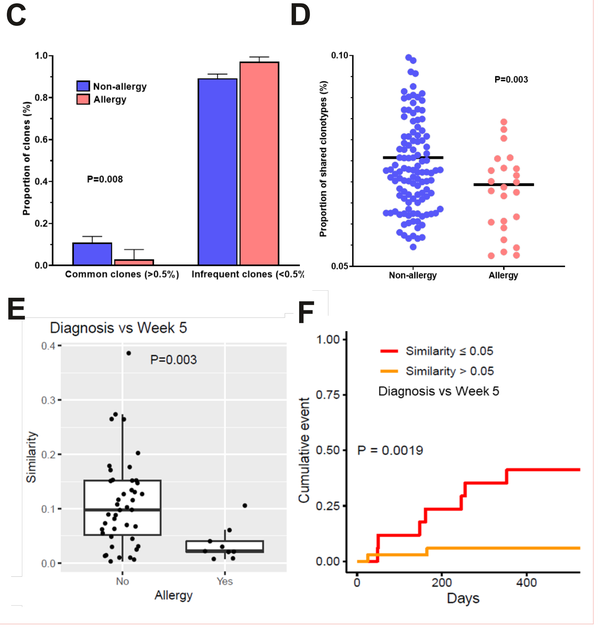
Higher TCR diversity and a more dynamically changing TCR-repertoire is associated with the risk of L-asparaginase hypersensitivity. This more diverse immunological mileau is characterized by infrequent, dissimilar, and less shared clonotypes, which increases the chance of a reactive clonotype matching to the allergen. Understanding the immunological basis of T-cell response in allergy may help in developing strategies to mediate this toxicity, to further improve outcomes in children with ALL.

Results

12 out of 67 children (17.9%) had allergy in our cohort, representing 16 episodes of varying severity (**Figure 1A**). First, we evaluated the association of TCR-repertoire diversity early on in treatment with the occurrence of subsequent allergy. **We found that a higher TCR diversity at all time-points was significantly associated with late allergy** (P=0.03 for Shannon's entropy, P=0.01 for inverse Simpson's index, **Figure 1B**).



This TCR diversity was characterized by a higher proportion of infrequent clones occurring <0.5% (P=0.008, **Figure 1C**). **Patients with allergy had significantly lower proportion of shared clonotypes** (P=0.003, **Figure 1D**). Examining the dynamic changes of the TCR repertoire between diagnosis and week 5 for each patient, we found that **patients with allergy had a much less similar clonotypic set (i.e. changed more) between timepoints compared to non-allergic patients** (P=0.003, **Figure 1E**). In fact, patients who demonstrated a higher variability in their clonotypes between timepoints (i.e. similarity coefficient of <0.05) had an 8.1-fold risk of allergic event (95% CI 1.7 – 39.1, P=0.001, **Figure 1F**).



Evaluating the TCR-repertoire before and **after an allergy, we found that there was convergence of TCR towards a common antigen** (**Figure 1G**), where clonotypes became more closely related after an allergy (**Figure 1H**). Finally, we found that **allergic patients had a lower proportion of public clonotypes** (i.e. public clonotypic sequences found on VDJDB database) compared to non-allergic patients (P=0.02, **Figure 1I**), supporting the "hygiene hypothesis" as a predisposing factor in development of allergy.

