Deciphering thymic development

Harald Von Boehmer*1,2

1 Department of Microbiology and Immunobiology, Harvard Medical School, Boston, MA, USA
2 Department of Cancer Immunology and AIDS, Dana-Farber Cancer Institute, Boston, MA, USA

*Correspondence: harald_von_boehmer@dfci.harvard.edu

Edited by: Kendall A. Smith, Weill Medical College of Cornell University, USA

Reviewed by: Herman Waldmann, University of Oxford, UK

Keywords: MHC-restricted antigen recognition, T cell clones, TCR alpha/beta cDNA clones, HY antigen, TCR transgenic mice, negative thymic selection, positive thymic selection

In 1979, some of us were surprised by the (1) conclusion of Doherty and Zinkernagel on MHC-restricted antigen recognition following the lead of Katz, Hamaoka, and Benacerraf (2) describing the same for the interaction of T helper cells with B cells as well as Rosenthal and Shevach (3) describing it for the interaction of T cells with macrophages. Since Doherty and Zinkernagel offered the least complicated system, they got most of the credit. We wanted to know whether a single effector cell was involved and therefore analyzed a clone of cells, specific for the HY transplantation antigen. If it could be shown that the progeny of a single cell was MHC restricted, we had something to explain, which was not obvious. The cloning in Basel worked fine, with the competent help of Hans Hengartner before his departure to the lab of Zinkernagel in Zurich. The clone was in fact MHC restricted, telling us that MHC restriction was the property of a single cell. The same conclusion was derived from experiments with Matthias Wabl, who observed the killing of targets by single killer cell (+). The clone was also alloreactive, which was observed prior to the realization that a significant portion of T cells carried two receptors and thus it is unclear to date whether a second receptor was involved or not. This was just the first example of a clone, which was MHC restricted and HY specific as well as H-2Dd specific and this overlap in specificity was subsequently observed in other clones such that the high frequency of alloreactive T cells is not really an issue.

Follow up experiments with the Michael Steinmetz lab transferring TCR alfa and beta genes from one T cell clone to another allowed us to unequivocally conclude that the MHC-restricted specificity was encoded by a single receptor long before crystallographic studies reached the same conclusion (3). This surprised some molecular biologists somewhat who thought that the cloning of the TCR put an end to the mysteries of the immune system.

This lead automatically to the next step, the construction of TCR transgenic mice, to analyze the selection of T cells according to their specificity. Initially, we were interested to test the ideas of Burnet and Lederberg that autoaggressive cells were eliminated in primary lymphoid organs. For this reason, we used again the genes of HY specific clones since then we could easily compare female and male mice. Here, I have to tell a little tale that characterizes (some?) scientists: it was Michael Steinmetz, who had previously spoken to Fritz Melchers, who asked at the Reisensburg in the South of Germany whether there would be any interest in generating TCR transgenic mice. I answered with a clear yes saying that this would allow to test Burnets and Lederbergs ideas. So, it was concluded to go ahead and initially Georges Koehler was singled out as the scientist residing by now in Freiburg to help with the construction of mice since he had succeeded to generate immunoglobulin transgenic mice. I was therefore mildly surprised when one day Georges entered my lab and asked me whether I could give him an HY specific clone since he had the idea of testing Burnets and Lederbergs ideas. I told him that this sounded familiar, he blushed only a little and then asked Hans Georg Rammensee who was in the same office a related question. I leave it to the audience to imagine what Michael Steinmetz told Georges Koehler or better what he did not tell him, even though one cannot be completely sure of it. So Georges did not produce the mice but Anton Berns in Amsterdam cooperated and very nicely mapped what was required to express TCR beta genes in transgenic mice. Finally, the co-injection of alfa and beta genes from an HY specific clone was done by Horst Blüthmann at Hoffmann La Roche in Basel where Michael Steinmetz had moved. When the mice had grown up, we tested them with a variety of reagents prepared for this task and could report on the deletion of CD4+8+ cells in male mice even though these mice came with an anomaly, the too early expression of the transgenic TCR, which made proper quantitation difficult (6). Only recently could we address this problem and reported deletion of CD4+8+ thymocytes in the absence of TCR editing (7). This ended a long story on the deletion of autoaggressive cells at a certain stage of development, something that had not been addressed in mice expressing superantigen specific receptors, which somewhat compromised our transgenic approach since they were conducted later and yielded results earlier albeit with the limitation that the conclusions had to be restricted to superantigens (8) whereas we dealt with conventional antigens for T cells.

The next step was related to positive selection and the matching of specificity and function. Here, the first realization was that a receptor derived from a CD8+ cell...
would only be expressed on CD8+ cells in the transgenic mice (9, 10). The sec-
ond was that there was in fact positive selection as mice with inappropriate MHC
antigens not restricting the specificity of the cell from which receptor genes were
obtained, failed to generate single positive cells and thus development was arrested
at the CD4+8+ stage where cells died (9). This was then named death from neglect
as opposed to death by negative selection which eliminated likewise CD4+8+ cells,
at least when the receptor was derived from CD8+ cells (7). It was then clear that it
was the MHC molecules expressed in the thymus and the TCR specificity, which deter-
mined positive selection, which also led to the matching of specificity and func-
tion (10), such that CD8+ killer cells were generated from immature cells express-
ing a class I restricted TCR (11) and as shown later CD4+ helper cells were gen-
erated from immature cells expressing a class II restricted TCR and thus in other
words helper cells recognized as a rule peptides entering the target cell from the
outside whereas killer cells recognized peptides produced in the target cell itself.
This relates to the different modes of pep-
tide loading by class I and class II MHC
antigens (12–16).

In the meantime, the molecular details
of this matching process have been worked out mostly by the work of Dietmar Kappes
(17) as well as Dan Littman (18) who identified transcription factors guiding this
process in dependence of the signaling by the receptor expressed by immature cells.
Thus, at present we have a fairly com-
plete picture of positive selection as far as the selectable T cells are concerned while
still we know relatively little about the TCR ligands that are responsible for posi-
tive selection. Here, one wonders whether thymus-specific proteasome subunits play
an essential role (19). Thus, there are still
some secrets in T cell development even after decades of the identification of the
TCR (20).

The curiosity in T cell development is still very much alive even after retire-
ment but I trust that the remaining issues are in good hands of younger scientific
colleagues who identify the outstanding

questions and think of clever experiments
to address them.

REFERENCES

1. Zinkernagel RM, Doherty PC. MHC-restricted
cytoxic T cell studies: on the biological role of polymorphic major histocompatibility
60262-x
2. Katz DH, Hamaoka T, Benacerraf B. Cell inter-
actions between histocompatible T and B lym-
phocytes. II. Failure of physiologic cooperative
interactions between T and B lymphocytes from
3. Rosenhalh AS, Shevach EM. Function of
macrophages in antigen recognition by guinea
pig T lymphocytes. I. Requirement for histo-
4. Wahl MR, von Boehmer H. Single Killer Cells Recog-
nize Non-H-2 as well as H-2 Determinants on Target
(1976), 60 p.
H, Boehmer von H, et al. Transfer of specificity by
6. Kisselov P, Blüthmann H, Staerz UD, Steinmetz
M, Boehmer von H. Tolerance in T-cell receptor
transgenic mice involves deletion of nonmature
T, Cantor H, et al. Negative selection, not recep-
tor editing, is a physiological response of autore-
8. Kappler JW, Roehm N, Marrack P. T cell toler-
9068-x
9. Scott B, Blüthmann H, Teh HS, Boehmer von
H. The generation of mature T cells requires
10. Teh HS, Kisselov P, Scott B, Kishi H, Uematsu Y,
Blüthmann H, et al. Thymic major histocompati-
bility complex antigens and the alpha beta T-cell
receptor determine the CD4/CD8 phenotype of T
335229a0
11. Kisselov P, Teh HS, Blüthmann H, Boehmer von H,
Positive selection of antigen-specific T cells in thy-
335:736–3. doi:10.1038/335736a0
12. Townsend A, Ollén C, Bastian J, Ljunggren HG, Fos-
ter L, Kärre K. Association of class I major histo-
compatibility heavy and light chains induced by
13. Davenport EV, Gow IR, Coadwell WJ, Monaco
JJ, Butcher GW, Howard JC. MHC class II region encoding proteins related to the
multidrug resistance family of transmembrane
14. Trowsdale J, Hanson I, Mockridge I, Beck S,
Townsend A, Kelly A. Sequences encoded in the
class II region of the MHC related to the
348:741–4. doi:10.1038/348741a0
genes in the murine MHC: possible implications
16. Spies T, Bresnahan M, Bahram S, Arnold D,
Blanc G, Mellins E, et al. A gene in the human
major histocompatibility complex class II
region controlling the class I antigen presentation
348744a0
17. He X, Park K, Kappes DJ. The role of ThPOK in
control of CD4/CD8 lineage commitment. Annu
annurev.immunol.28.022006.141715
18. Egawa T, Littman DR. ThPOK acts late in spec-
fication of the helper T cell lineage and sup-
presses Runx-mediated commitment to the cyto-
H, Takahama Y, et al. Regulation of CD8+ T
cell development by thymus-specific proteasomes.
1141912
20. Boehmer von H, Hengartner H, Nabhak M, Lern-
hardt W, Schreier MF, Haas W. Fine specificity
of a continuously growing killer cell clone specific

Conflict of Interest Statement: The author declares that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 14 August 2014; accepted: 19 August 2014; published online: 08 October 2014.

This article was submitted to Immunological Tolerance, a section of the journal Frontiers in Immunology.

Copyright © 2014 Von Boehmer. This is an open-access article distributed under the terms of the Creative Com-
mons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided
the original author(s) or licensor are credited and that the original publication in this journal is cited, in accordance
with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with
these terms.