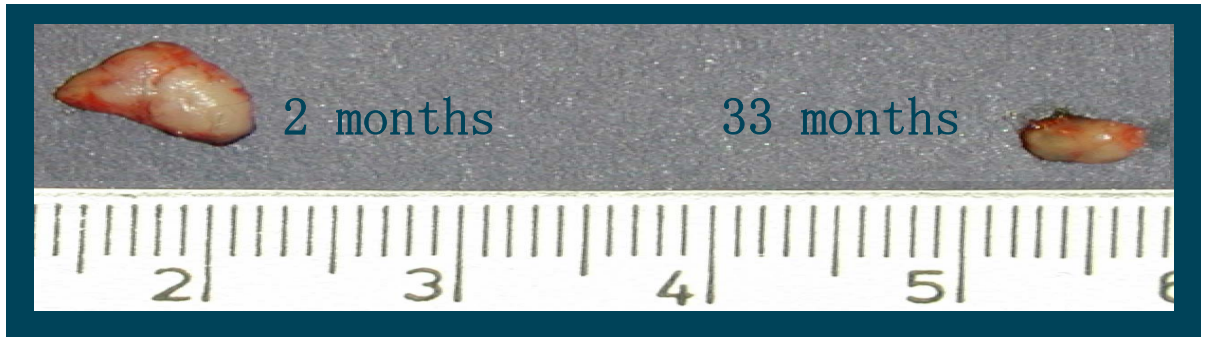
Modification of IL-7 and the Reversal of Thymic Involution in Aged Mice

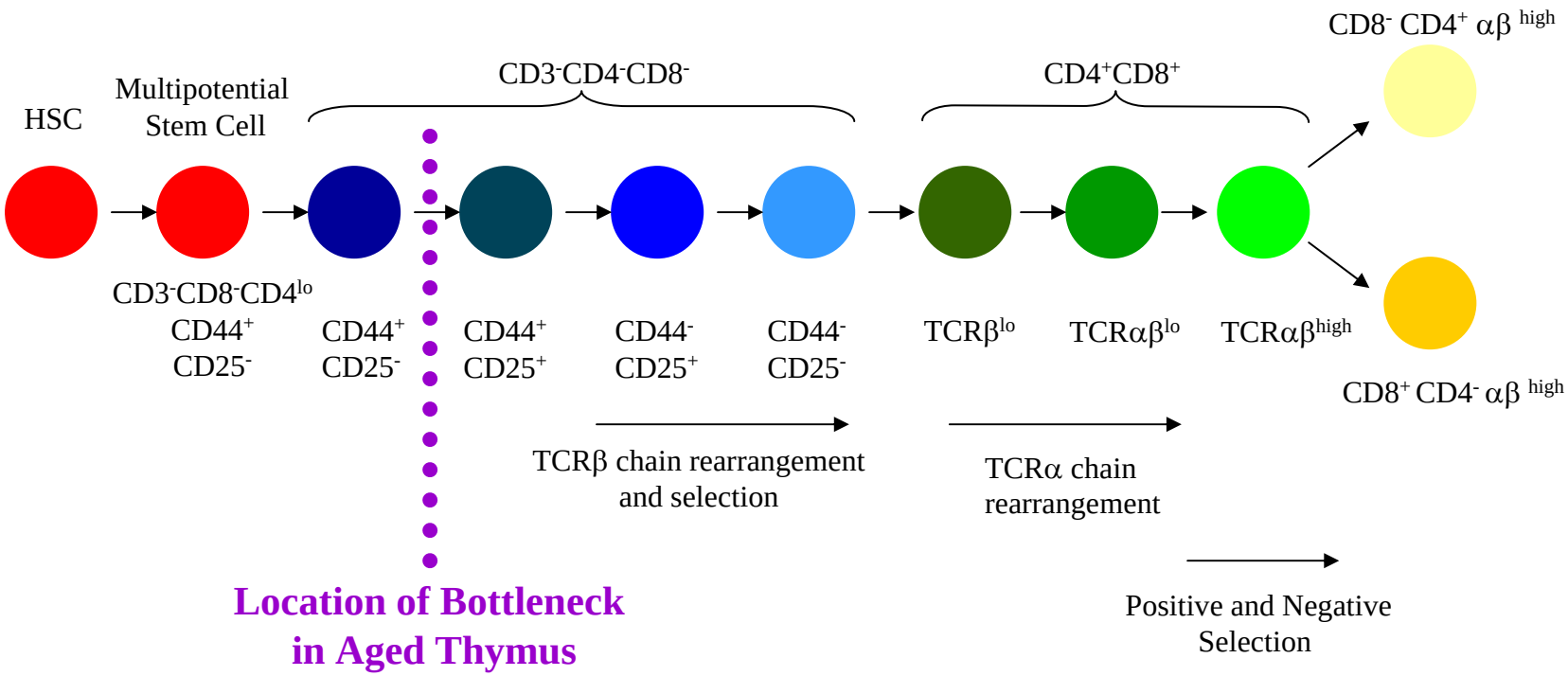
Dr Siân Henson

Effect of Age on the Thymus



Age	n	Total	$CD4^-CD8^-$	$CD4^+CD8^+$	$CD4^+CD8^-$	$CD4^-CD8^+$
3 mo	9	$1.64\pm0.3\times10^8$ (100%)	$3.1\pm1.5\times10^6$ (2±1%)	$1.4\pm0.3\times10^8$ (86 ± 3%)	$1.2\pm0.4\times10^7$ (7 ± 2%)	$8.5\pm4.7\times10^6$ (5 ± 2%)
20 mo	8	$2.75\pm0.6\times10^7$ (100%)	$8.6\pm3\times10^5$ (3 ±1%)	$2.2\pm0.5\times10^7$ (83±19%)	$2.6\pm0.89\times10^6$ (10±3%)	$1.1\pm0.4\times10^6$ (4±1%)
Difference		1.36×10^8	2.23×10^6	1.17×10^8	9.32×10^6	7.38×10^6

Effect of Age on T Cell Development



Problems With IL-7 Therapy

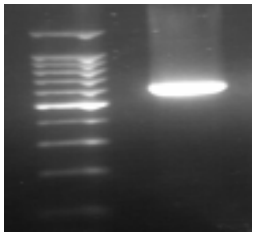
- Half life of IL-7 is short, 115 minutes in vivo
- Frequent injection at a dose high enough to reach the thymus in sufficient quantity to have an effect

Modification of IL-7 to Improve Effectiveness

- Targeting

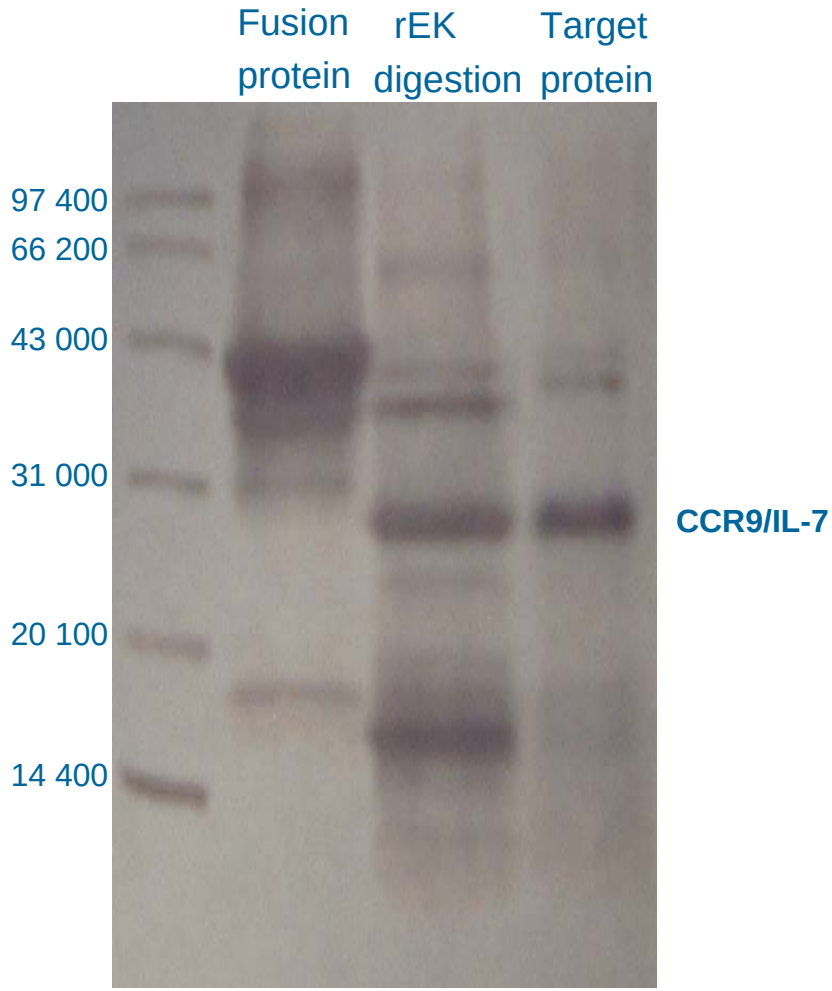
  create a fusion protein between IL-7 and a chemokine receptor whose ligand is organ specific

CCR9/IL-7 Fusion Clone

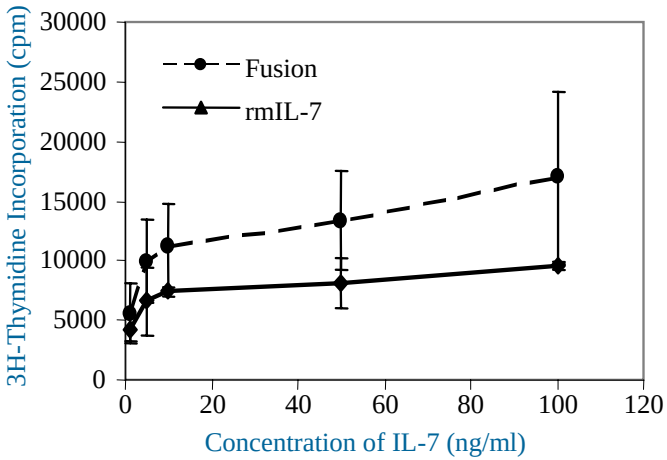


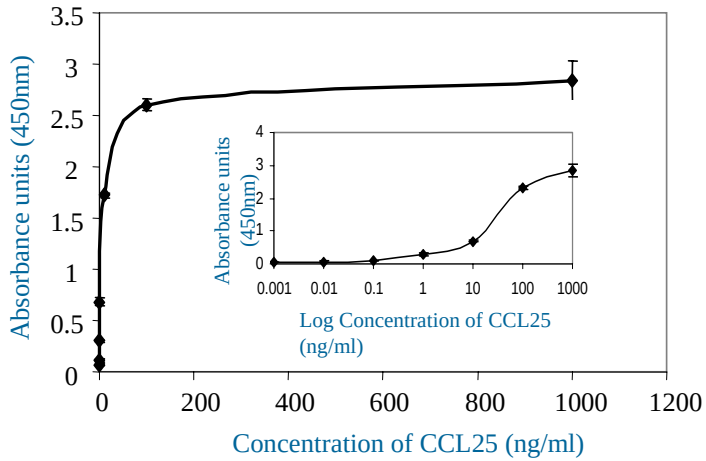
612 bp

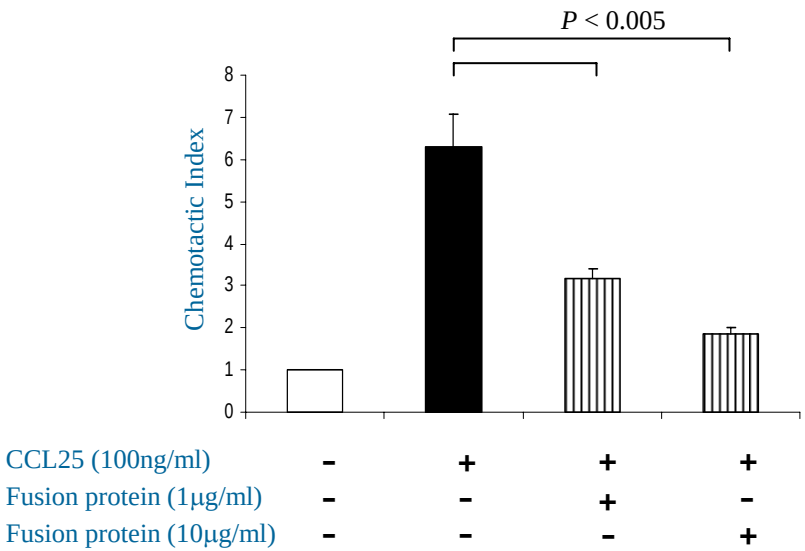
```
atgatgcccacagaactcacaagccttattcctggcatgtttgatgacttcagctatgac
 M  M  P  T  E  L  T  S  L  I  P  G  M  F  D  D  F  S  Y  D
tccactgcttccacagatgactacatgaatttgaatttcagtagcttcttctgtaagaaa
 S  T  A  S  T  D  D  Y  M  N  L  N  F  S  S  F  F  C  K  K
aataatgtcaggcagtttgcacgcggccgcatgttccatgtttcttttagatatatcttt
 N  N  V  R  Q  F  A  R  G  R  M  F  H  V  S  F  R  Y  I  F
ggaattcctccactgatccttgttctgctgcctgtcacatcatctgagtgccacattaaa
 G  I  P  P  L  I  L  V  L  L  P  V  T  S  S  E  C  H  I  K
gacaaagaaggtaaagcatatgagagtgtactgatgatcagcatcgatgaattggacaaa
 D  K  E  G  K  A  Y  E  S  V  L  M  I  S  I  D  E  L  D  K
atgacaggaaccgatagtaattgcccgaataatgaaccaaacttttttagaaaacatgta
 M  T  G  T  D  S  N  C  P  N  N  E  P  N  F  F  R  K  H  V
tgtgatgatacaaaggaagctgcttttctaaatcgtgctgcccgcaagttgaagcaattt
 C  D  D  T  K  E  A  A  F  L  N  R  A  A  R  K  L  K  Q  F
ctttaaaatgaatatcagtgaagaattcaatgtccacttactaacagtatcacaaggcaca
 L  K  M  N  I  S  E  E  F  N  V  H  L  L  T  V  S  Q  G  T
gaaactactggtgagctgcacaagtaaggaagaaaaaaacgtaaaggaacagaaaaagaat
 Q  T  L  V  S  C  T  S  K  E  E  K  N  V  K  E  Q  K  K  N
gatgcatgtttcctaaagagactactgagagaaataaaaacttgttggaataaaatttttg
 D  A  C  F  L  K  R  L  L  R  E  I  K  T  C  W  N  K  I  L
aagggcagtata
 K  G  S  I
```



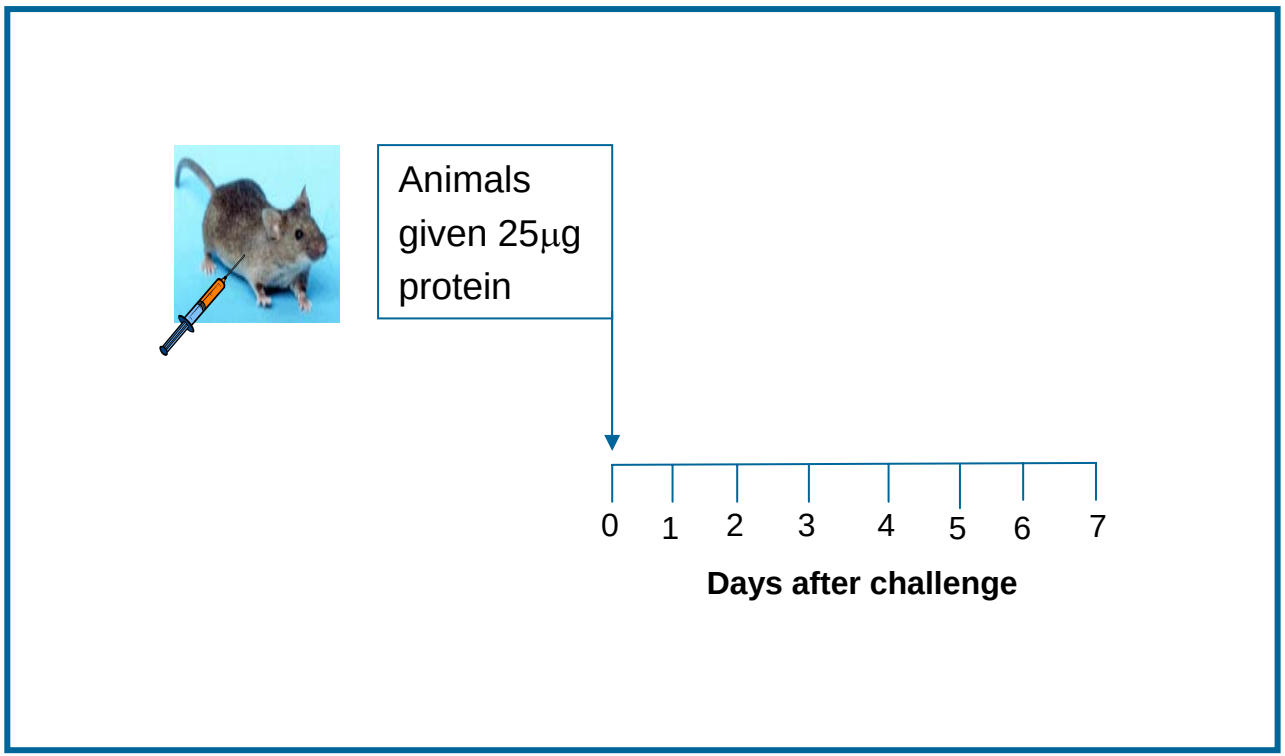
In Vitro Activity of the CCR9/IL-7 Fusion Protein



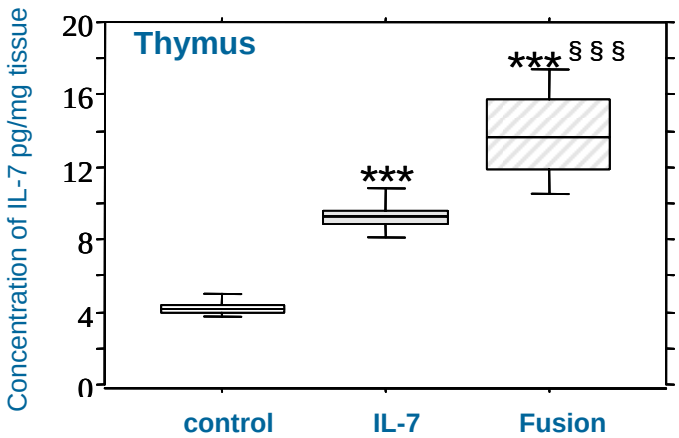


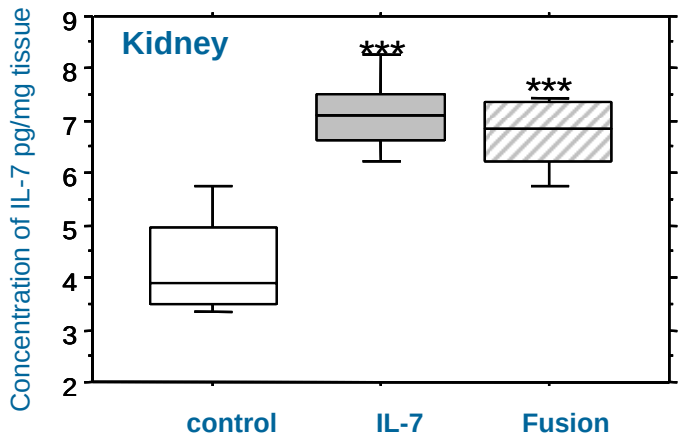


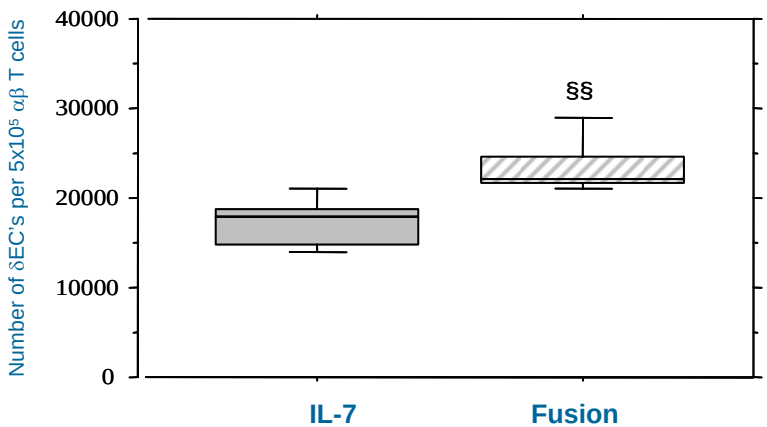
Treatment with IL-7/CCR9 Fusion Protein



In Vivo Activity of the CCR9/IL-7 Fusion Protein

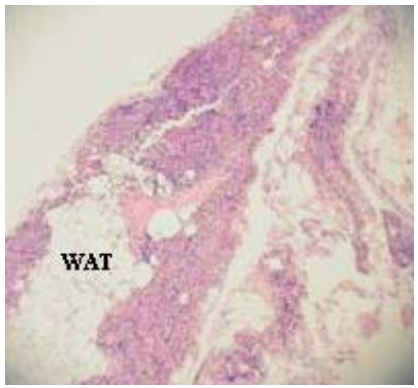




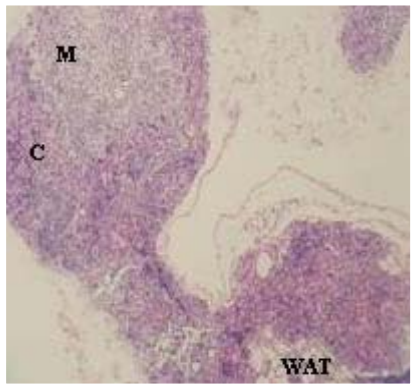


Restoration of Thymic Architecture After Treatment with the Fusion Protein

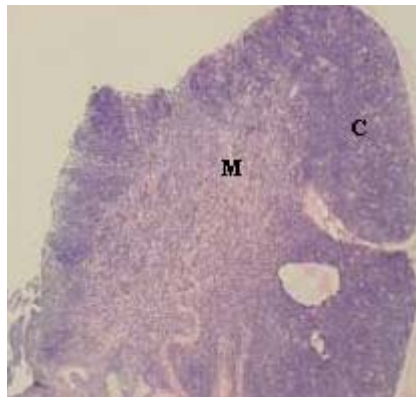
Control



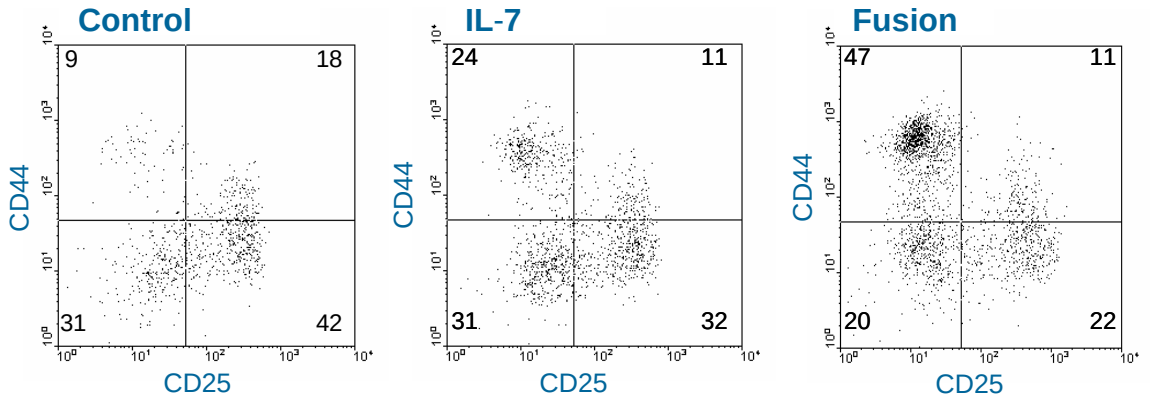
IL-7

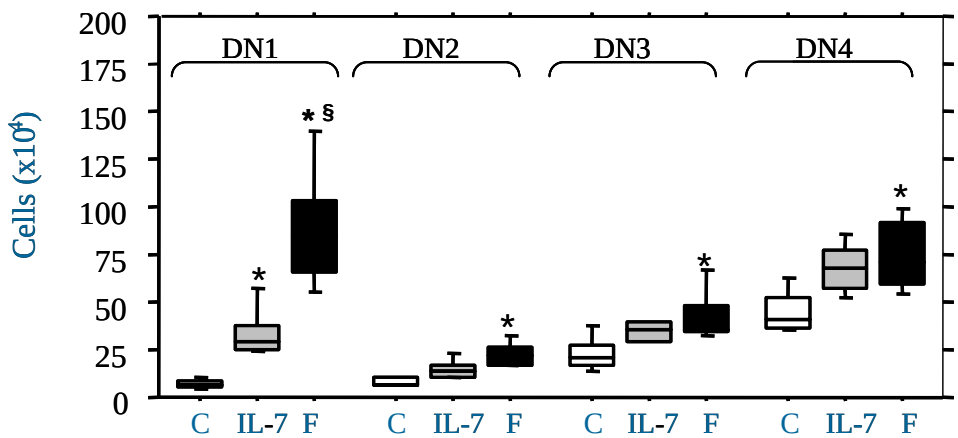


Fusion

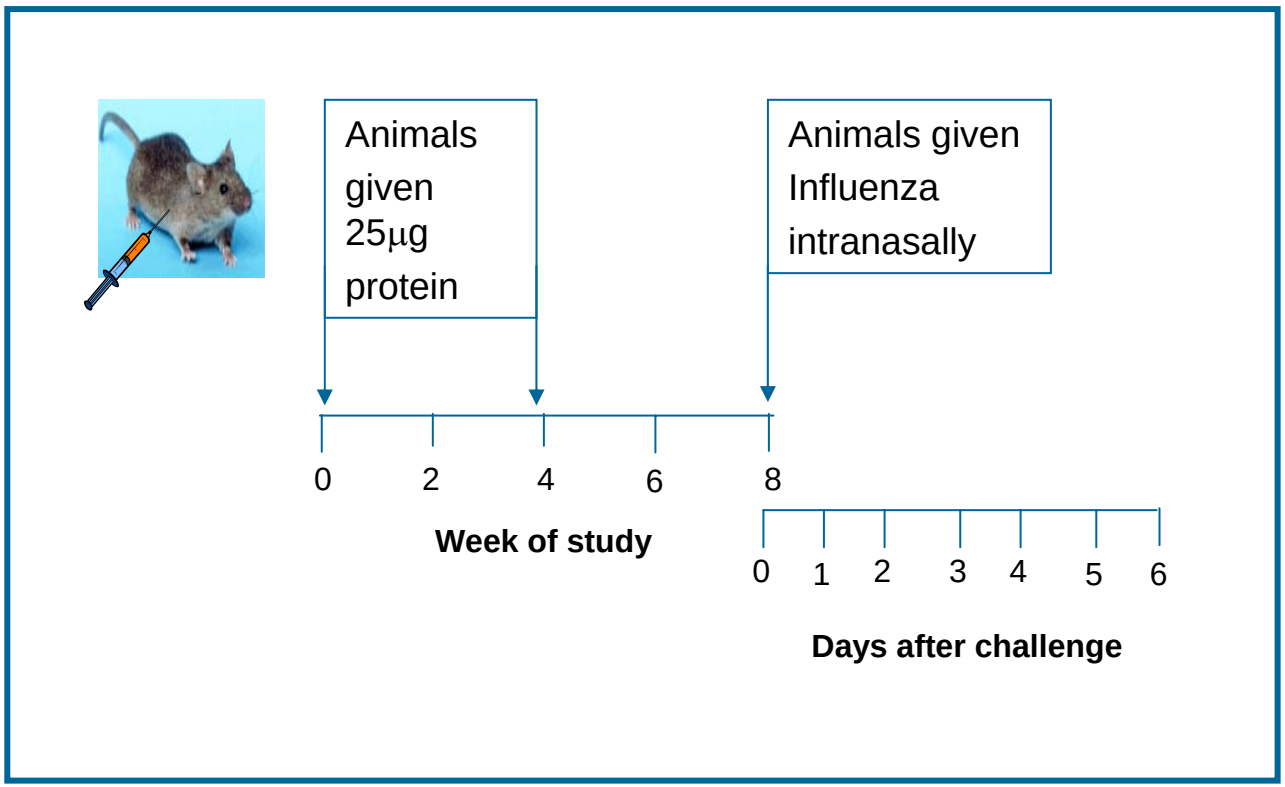


Increased Thymocyte Numbers After Treatment with the Fusion Protein

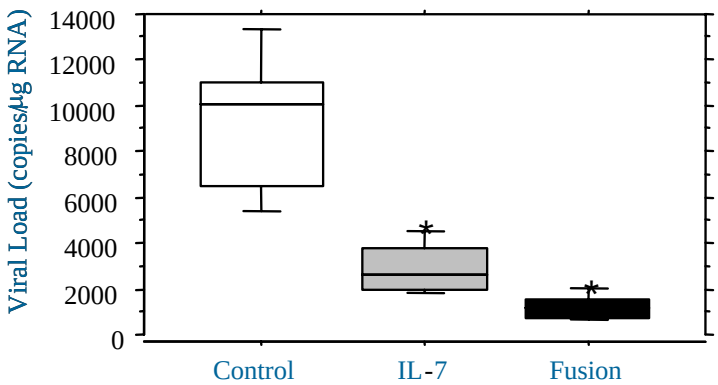


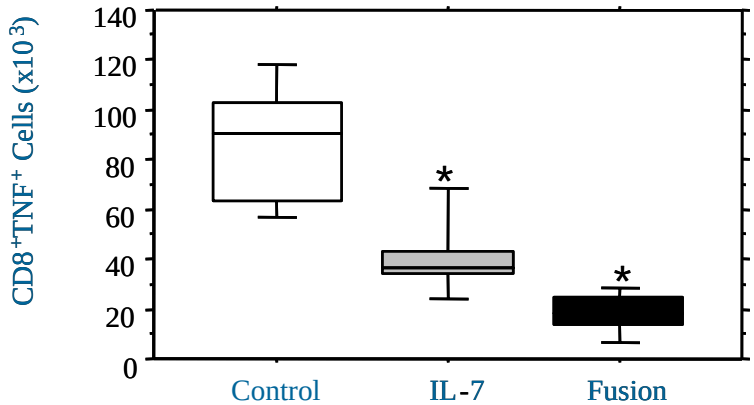


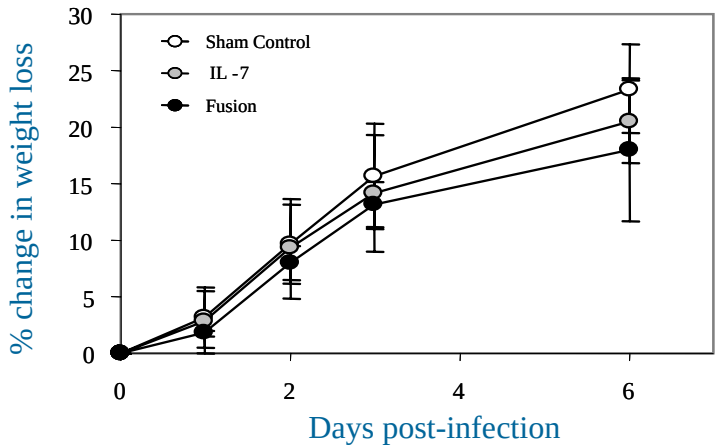
Challenge with Influenza



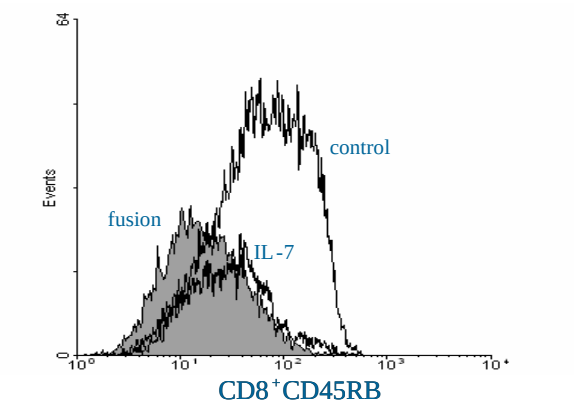
Fusion Treatment Causes Reduced Influenza Disease Progression

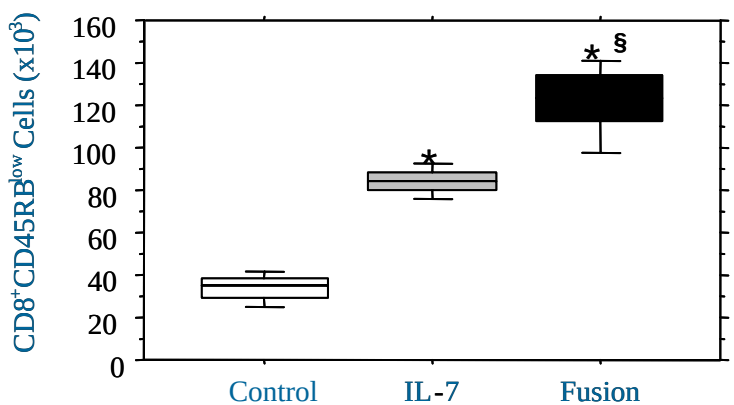


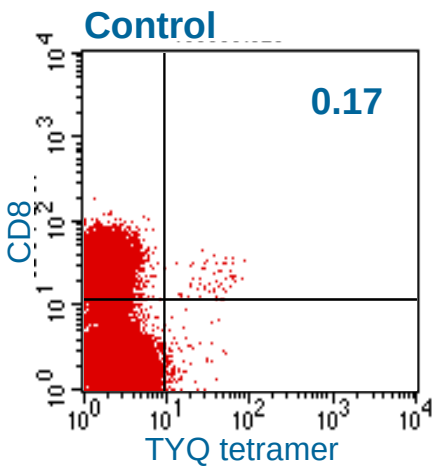


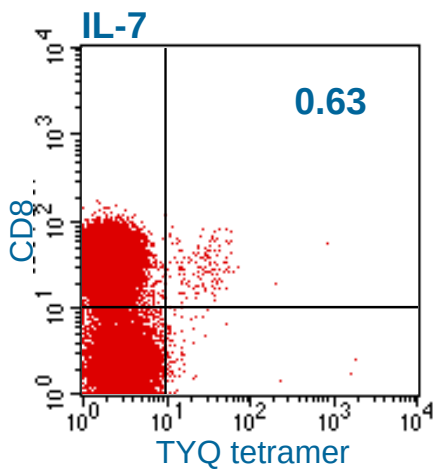


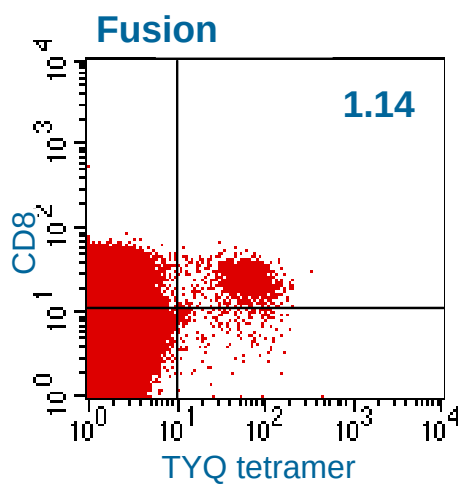
CD45RB^{low} Expression in CD8^{+} Cells is Greater in Fusion Treated Animals After Receiving Influenza











Conclusions

- I have created a novel fusion protein between CCR9 and IL-7 which retains both its IL-7 activity and CCL25 binding ability
- It shows an enhanced affinity for the thymus and causes an improvement in de novo T cell production with an improvement in the ability to respond to challenge with influenza
- This fusion protein is a more effective immunorestorative agent than treatment with IL-7 alone

Acknowledgments

IL-7 Work
Imperial College

Richard Aspinall

Influenza Work
Kennedy Institute

Tracy Hussell
Robert Snelgrove

Support

bbsrc
biotechnology and biological sciences research council

Research into Ageing
Help the Aged

sparc
epsrc
bbsrc