

LIST OF FIGURES

Figures	Title	Page
Figure 1	Cellular effect and signalling of TNF α in immune/ normal cells.	10
Figure 2	Structure of antibody molecule	14
Figure 3	Human antibody techniques	16
Figure 4	Different format of antibody fragments	21
Figure 5	Structure of a filamentous phage displaying scFv fragments on its surface	22
Figure 6	Selection of specific antibodies from combinatorial antibody phage display libraries using biopanning	31
Figure 7	Yeast surface display	35
Figure 8	Polyclonal phage ELISA	77
Figure 9	Selection of binders based on Avidity	79
Figure 10	Map of phagemid vector pIT2	80
Figure 11	Expression of anti-TNF α scFv (Recab)	85

Figure 12	EC50 value of C2 and G2 clones MoSUMO and	86
Figure 13	Protein-A affinity purification of Recab and western blot analysis	89
Figure 14	ELISA to investigate stability of binding of Recab to hTNF α at different concentration of NaCL, Urea, DMSO, NaSCN, and different pH on antigen binding	92
Figure 15	Affinity maturation for Recab using Yeast Display System	94
Figure 16	Screening of mutated Recab clones	98
Figure 17	Bivariate flowcytometric analysis	99-100
Figure 18	Multiple sequence alignment of anti-TNF clones	102
Figure 19	Construction of Recab expression construct for <i>P. pastoris</i>	129
Figure 20	Screening and Southern blot analysis of Recab transformants	131
Figure 21	Selection of Recab transformants based on ELISA and Southern blot analysis	135

Figure 22	Effect of different methanol concentration on Recab expression	137
Figure 23	Effect of temperature on Recab expression	138
Figure 24	Cell based assay for Recab	139