

KEY SUPPORTING RESEARCH

Key concepts of the Faustman lab have been confirmed or expanded upon by the scientific literature.

A growing body of literature supports the concept that TNF- α , or induction of TNF (via BCG or CFA), might beneficially change the disease course in some forms of murine autoimmunity:

1. Tran S, Kodama S, Mezey EM. Treatment success and regenerative mechanisms influenced by age of NOD mice and target organ of autoimmune attack [Abstract]. *Diabetes* 2006, 1202-P.
2. Faustman D, Tran S, Kodama S, Lodde B, Szalayova I, Key S, Toth Z, Mezey E. Comment on Papers by Chong et al, Nishio et al and Suri et al on Diabetes Reversal in NOD Mice. *Science* 2006 314:1243-4.
3. Okubo Y, Kanazawa Y, Oikawa Y, Miyazaki J, Shimada A. Islet hypertrophy observed in “reversed” diabetic NOD mouse after pancreatic beta cell line administration [Abstract]. *Diabetes* 2006, 1193-P.
4. Nishio J, Gaglia JL, Turvey SE, Campbell C, Benoist C, Mathis D. Islet recovery and reversal of murine type 1 diabetes in the absence of any infused spleen cell contribution. *Science* 2006 311:1775-8.
5. Chong AS, Shen J, Tao J et al. Reversal of diabetes in nonobese diabetic mice without spleen cell-derived beta cell regeneration. *Science* 2006 311:1774-5.
6. Suri A, Calderon B, Esparza TJ, Frederick K, Bittner P, Unanue ER. Immunological reversal of autoimmune diabetes without hematopoietic replacement of beta cells. *Science* 2006 311:1778-80.
7. Qin HY, Chaturvedi P, Singh B. In vivo apoptosis of diabetogenic T cells in NOD mice by IFN- γ /TNF- α . *Int Immunol.* 2004 16(12):1723-32.
8. Kodama S, Kuhlreiber W, Fujimura S, Dale EA, Faustman DL. Islet regeneration during the reversal of autoimmune diabetes in NOD mice. *Science.* 2003 Nov 14; 302(5648):1223-7.
9. Ryu S, Kodama S, Ryu K, Schoenfeld DA, Faustman DL. Reversal of established autoimmune diabetes by restoration of endogenous beta cell function. *J Clin Invest.* 2001 Jul; 108(1):63-72.
10. Qin HY, Singh B. BCG vaccination prevents insulin-dependent diabetes mellitus (IDDM) in NOD mice after disease acceleration with cyclophosphamide. *J Autoimmunity* 1997; 10: 271-8.
11. Shehadeh N, Etzioni A, Cahana A, et al. Repeated BCG vaccination is more effective than a single dose in preventing diabetes in non-obese diabetic (NOD) mice. *Isr J Med Sci* 1997; 33: 711-5.
12. Kalechman Y, Gafter U, Da JP, Albeck M, Alarcon-Segovia D, Sredni B. Delay in the onset of systemic lupus erythematosus following treatment with the immunomodulator AS101: association with IL-10 inhibition and increase in TNF- α levels. *J Immunol* 1997; 159: 2658-67.
13. Rabinovitch A, Suarez-Pinzon WL, Sorensen O, Rajotte RV, Power RF. TNF- α down-regulates type 1 cytokines and prolongs survival of syngeneic islet grafts in nonobese diabetic mice. *J Immunology* 1997; 159:6928-303.
14. Grewal IS, Grewal KD, Wong FS, Picarella DE, Janeway CA, Flavell RA. Local expression of transgene encoded TNF- α in islets prevents autoimmune diabetes in non-obese diabetic (NOD) mice by preventing the development of autoreactive islet specific T cells. *J Exp Med* 1996; 184:1963-74.

15. Bras A, Aguas AP. Diabetes-prone NOD mice are resistant to Mycobacterium avium and the infection prevents autoimmune disease. *Immunology* 1996; 89:20-5.
16. Lakey JR, Singh B, Warnock GL, Rajotte RV. BCG immunotherapy prevents recurrence of diabetes in islet grafts transplanted into spontaneously diabetic NOD mice. *Transplantation* 1994; 57: 1213-7.
17. Qin HY, Sadelain MW, Hitchon C, Lauzon J, Singh B. Complete Freund's adjuvant-induced T cells prevent the development and adoptive transfer of diabetes in nonobese diabetic mice. *J Immunol* 1993; 150:2072-80.
18. Wang T, Singh B, Warnock GL, Rajotte RV. Prevention of recurrence of IDDM in islet-transplanted diabetic NOD mice by adjuvant immunotherapy. *Diabetes* 1992; 41: 114-7.
19. McInerney MF, Pek SB, Thomas DW. Prevention of insulinitis and diabetes onset by treatment with complete Freund's adjuvant in NOD mice. *Diabetes* 1991; 40: 715-725
20. Harada M, Kishimoto Y, Makino S. Prevention of overt diabetes and insulinitis in NOD mice by a single BCG vaccination. *Diabetes Res Clinic Prac* 1990; 8:85-89.
21. Sadelain MWJ, Hui-Yu Q, Lauzon J, Singh B. Prevention of type 1 diabetes in NOD mice by adjuvant immunotherapy. *Diabetes* 1990; 39:583-9.
22. Satoh J, Seino H, Shintani S, Tanaka S, Ohteki T, Masuda T, et al. Inhibition of type 1 diabetes in BB rats with recombinant human tumor necrosis factor-alpha. *J Immunology* 1990; 145:1395-9.
23. Jacob CO, Aiso S, Michie SA, McDevitt HO, Acha-Orbea H. Prevention of diabetes in nonobese diabetic mice by tumor necrosis factor; similarities between TNF-alpha and interleukin 1. *PNAS* 1990; 87:968-972.
24. Satoh J, Seino H, Abo T. Recombinant human tumor necrosis factor-alpha suppresses autoimmune diabetes in nonobese diabetic mice. *J Clin Invest* 1989; 84:1345-8.
25. Jacob CO, McDevitt HO. Tumour necrosis factor-alpha in murine autoimmune 'lupus' nephritis. *Nature* 1988; 331:356-8.

A growing body of literature using anti-TNF- α drugs that reduce circulating TNF worsen or induce new forms of autoimmunity; TNF itself or TNF receptor mutations may be contributory to autoreactivity

Human literature:

1. Enayati PJ, Papadakis KA. Association of Anti-tumor Necrosis Factor Therapy With the Development of Multiple Sclerosis. *J Clin Gastroenterol* 2005; 39: 303-306.
2. Levine A, Shamir R, Wine E, Weiss B, Karban A, Shaoul RR, Reif SS, Yakir B, Friedlander M, Kaniel Y, Leshinsky-Silver E. TNF promoter polymorphisms and modulation of growth retardation and disease severity in pediatric Crohn's disease. *Am J Gastroenterol* 2005 100:1598-1604.
3. Thomas CW, Jr., Weinshenker BG, Sandborn WJ. Demyelination during anti-tumor necrosis factor alpha therapy with infliximab for Crohn's disease. *Inflamm Bowel Dis* 2004; 10: 28-31.
4. Vermeire S, Noman M, Van Assche G, et al. Autoimmunity associated with anti-tumor necrosis factor alpha treatment in Crohn's disease: a prospective cohort study. *Gastroenterology* 2003; 125: 32-9.
5. Food and Drug Administration (FDA). Update on the TNF-alpha blocking agents. 2003. http://www.fda.gov/ohrms/dockets/ac/03/briefing/3930B1_01_B-TNF.briefing.pdf.
6. Swale VJ, Perrett CM, Denton CP, Black CM, Rustin MH. Etanercept-induced systemic lupus erythematosus. *Clin Exp Dermatol* 2003; 28: 604-7.
7. Jarrett SJ, Cunnane G, Conaghan PG, et al. Anti-tumor necrosis factor-alpha therapy-induced vasculitis: case series. *J Rheumatol* 2003; 30: 2287-91.
8. Van Heel DA, Udalova IA, DeSilva AP, McGovern DP, Kinouchi Y, Hull J, Lench NJ, Cardon LR, Carey AH, Jewell DP, Kwiatkowski D. Inflammatory bowel disease is

- associated with a TNF polymorphism that affects an interaction between the OCT1 and NF- κ B transcription factors. *Human Mol Genet* 2002; 11:1281-9.
9. Shakoor N, Michalska M, Harris CA, Block JA. Drug-induced systemic lupus erythematosus associated with etanercept therapy. *Lancet* 2002; 359: 579-80.
 10. Cairns AP, Duncan MK, Hinder AE, Taggart AJ. New onset systemic lupus erythematosus in a patient receiving etanercept for rheumatoid arthritis. *Ann Rheum Dis* 2002; 61: 1031-2.
 11. Shtauvere-Brameus A, Dabadghao P, Rumba I Sanjeevi CB Tumor necrosis factor-alpha allele 2 shows an association with insulin dependent diabetes mellitus in Latvians 2002; 958, 357.
 12. Sicotte NL, Voskuhl RR. Onset of multiple sclerosis associated with anti-TNF therapy. *Neurology* 2001; 57: 1885-8.
 13. Bleumink GS, ter Borg EJ, Ramselaar CG, Ch Stricker BH. Etanercept-induced subacute cutaneous lupus erythematosus. *Rheumatology (Oxford)* 2001; 40: 1317-9.
 14. Sandborn WJ. Strategies targeting tumor necrosis factor in Crohn's disease. *Acta Gastroenterol Belg* 2001; 64: 170-2.
 15. Lipsky PE, van der Heijde DM, St. Clair EW, et al. Infliximab and methotrexate in the treatment of rheumatoid arthritis. Anti-Tumor Necrosis Factor Trial in Rheumatoid Arthritis with Concomitant Therapy Study Group. *N Engl J Med* 2000; 343: 1594-602.
 16. Charles PJ, Smeenk RJ, De Jong J, Feldmann M, Maini RN. Assessment of antibodies to double-stranded DNA induced in rheumatoid arthritis patients following treatment with infliximab, a monoclonal antibody to tumor necrosis factor alpha: findings in open-label and randomized placebo-controlled trials. *Arthritis Rheum* 2000; 43: 2383-90.
 17. Galaria NA, Werth VP, Schumacher HR. Leukocytoclastic vasculitis due to etanercept. *J Rheumatol.* 2000; 2041-4.
 18. Schaible TF. Long term safety of infliximab. *Can J Gastroenterol* 2000; 14 (Suppl C): 29C-32C.
 19. Bloom BJ. Development of diabetes mellitus during etanercept therapy in a child with systemic-onset juvenile rheumatoid arthritis. *Arthritis Rheum* 2000; 43: 2606-8.
 20. Moreland LW, Schiff MH, Baumgartner SW, et al. Etanercept therapy in rheumatoid arthritis. A randomized, controlled trial. *Ann Intern Med* 1999; 130: 478-86.
 21. The Lenercept Multiple Sclerosis Study Group and The University of British Columbia MS/MRI Analysis Group. TNF neutralization in MS: results of a randomized, placebo-controlled multicenter study. *Neurology* 1999; 53: 457-65.
 22. Jurewicz AM, Walczak AK, Selmaj KW. Shedding of TNF receptors in multiple sclerosis patients. *Neurology* 1999; 53: 1409-14.
 23. Sandborn WJ, Hanauer SB. Antitumor necrosis factor therapy for inflammatory bowel disease: a review of agents, pharmacology, clinical results, and safety. *Inflamm Bowel Dis* 1999; 5: 119-33.
 24. Jurewicz AM, Walczak AK, Selmaj KW. Shedding of TNF receptors in multiple sclerosis patients. *Neurology* 1999; 53: 1409-14.
 25. Gabay C, Cakir N, Moral F, et al. Circulating levels of tumor necrosis factor soluble receptors in systemic lupus erythematosus are significantly higher than in other rheumatic diseases and correlate with disease activity. *J Rheumatol* 1997; 24: 303-8.
 26. van Oosten BW, Barkhof F, Truyen L, Boringa JB, Bertelsmann FW, von Blomberg BM, et al. Increased MRI activity and immune activation in two multiple sclerosis patients treated with the monoclonal anti-tumor necrosis factor antibody cA2. *Neurology* 1996; 47: 1531-4.
 27. Aderka D, Wytensbeek A, Engelmann H, et al. Correlation between serum levels of soluble tumor necrosis factor receptor and disease activity in systemic lupus erythematosus. *Arthritis Rheum* 1993; 36: 1111-20.

28. Cope AP, Aderka D, Doherty M, et al. Increased levels of soluble tumor necrosis factor receptors in the sera and synovial fluid of patients with rheumatic diseases. *Arthritis Rheum* 1992; 35: 1160-9.

Mouse/Rat literature:

1. Kontoyiannis D, Kollias G. Accelerated autoimmunity and lupus nephritis in NZB mice with an engineered heterozygous deficiency in tumor necrosis factor. *Eur J Immunol* 2000; 30: 2038-47.
2. Fujimura T, Hirose S, Jiang Y, Kodera S, Ohmuro H, Zhang D, et al. Dissection of the effects of tumor necrosis factor-alpha and class II gene polymorphisms within the MHC on murine systemic lupus erythematosus (SLE). *Int Immunol* 1998; 10: 1467-72.
3. Rothe H, Schuller I, Richter G, et al. Abnormal TNF production in prediabetic BB rats is linked to defective CD45R expression. *Immunology* 1992; 77: 1-6.

A growing body of literature supports the concept that the NF- κ B pathway is deranged in autoimmune cells in murine and human models of autoimmunity:

Human literature (includes proteasome gene mutations that alter NF- κ B):

1. Maeda S, Hsu LC, Liu H, Bankston LA, Iimura M, Kagnoff MF, Eckmann L, Karin M. NOD2 mutation in Crohn's disease potentiates NF- κ B activity and IL-1 β processing. *Science* 2005 307:734-8.
2. Guo D, Li M, Zhang Y, et al. A functional variant of SUMO4, a new I κ Ba modifier, is associated with type 1 diabetes. *Nat Genet* 2004; 36:837-41.
3. Kessel A, Rosner I, Rozenbaum M, et al. Increased CD8+ T cell apoptosis in scleroderma is associated with low levels of NF- κ B. *J Clin Immunol* 2004; 24:30-6.
4. Abbott DW, Wilkins A, Asara JM, Cantley LC. The Crohn's disease protein, NOD2, requires RIP2 in order to induce ubiquitinylation of a novel site on NEMO. *Curr Biol* 2004 14(24):2217-27.
5. Mitterski B, Bohringer S, Klein W, Sindern E, Haupts M, Schimrigk S, Epplen JT. Inhibitors in the NF- κ B cascade comprise prime candidate genes predisposing to multiple sclerosis, especially in selected combinations. *Genes Immun* 2002 3(4):211-9.
6. Hugot JP, Chamaillard M, Zouali H, et al. Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. *Nature* 2001; 411: 599-603.
7. Ogura Y, Bonen DK, Inohara N, et al. A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature* 2001; 411: 603-6.
8. Wong HK, Kammer GM, Dennis G, Tsokos GC. Abnormal NF-kappa B activity in T lymphocytes from patients with systemic lupus erythematosus is associated with decreased p65-RelA protein expression. *J Immunol* 1999; 163: 1682-9.

Mouse literature:

1. Eckmann L, Karin M. NOD2 and Crohn's disease: loss or gain of function? *Immunity* 2006 22(6):661-7.
2. Karban AS, Okazaki T, Panhuysen CI, Gallegos T, Potter JJ, Bailey-Wilson JE, Silverberg MS, Duerr RH, Cho JH, Gregersen PK, Wu Y, Achkar JP, Dassopoulos T, Mezey E, Bayless TM, Novet FJ, Brant SR. Functional annotation of a novel NF- κ B1 promoter polymorphism that increases risk for ulcerative colitis. *Human Mol Genet* 2004 13:35-45.
3. Valero R., Baron ML, Guerin S, et al. A defective NF-kappa B/RelB pathway in autoimmune-prone New Zealand black mice is associated with inefficient expansion of thymocyte and dendritic cells. *J Immunol* 2002; 169: 185-92.
4. Kammer GM, Tsokos GC. Abnormal T lymphocyte signal transduction in systemic lupus erythematosus. *Curr Dir Autoimmun* 2002 5:131-50.

5. Hegazy DM, O'Reilly DA, Yang BM, Hodgkinson AD, Millward BA, Demaine AG. NF- κ B polymorphisms and susceptibility to type 1 diabetes. *Genes Immun* 2001 2:304-8.
6. Hayashi T, Faustman DL. Selected contribution: Association of gender-related LMP2 inactivation with autoimmune pathogenesis. *J Appl Physiol* 2001; 91: 2804-15.
7. Hayashi T, Faustman D. Essential role of HLA-encoded proteasome subunits in NF- κ B activation and prevention of TNF- α induced apoptosis. *J Biol Chem* 2000 275:5238-47.
8. Hayashi T, Faustman D. NOD mice are defective in proteasome production and activation of NF- κ B. *Mol Cell Biol* 1999; 19: 8646-59.

A growing body of literature supports the concept that TNF- α induction or administration directly kills the pathogenic T cells of autoimmunity:

1. Kodama S, Davis M, Faustman DL. The therapeutic potential of tumor necrosis factor for autoimmune disease: a mechanistically based hypothesis. *Cell Mol Life Sci*. 2005;62(16):1850-62.
2. Qin HY, Chaturvedi P, Singh B. In vivo apoptosis of diabetogenic T cells in NOD mice by IFN-g/TNF-a. *Int Immunol*. 2004 Dec;16(12):1723-1732.
3. Christen U, Von Herrath M. Apoptosis of autoreactive CD8 lymphocytes as a potential mechanism for the abrogation of type 1 diabetes by islet-specific TNF-a expression at a time when autoimmune process is already ongoing. *Ann NY Acad Sci* 2002; 958: 166-169.
4. Ryu S, Kodama S, Ryu K, Schoenfeld DA, Faustman DL. Reversal of established autoimmune diabetes by restoration of endogenous beta cell function. *J Clin Invest* 2001; 108: 63-72.
5. Hayashi T, Kodama S, Faustman DL. Reply to 'LMP2 expression and proteasome activity in NOD mice'. *Nat Med* 2000; 6: 1065-6.
6. Hayashi T, Faustman D. NOD mice are defective in proteasome production and activation of NF- κ B. *Mol Cell Biol* 1999, 19: 8646-59.

A body of literature supports the concept that MHC class I and self peptide is an education complex, can recognize pathogenic cells, is necessary to prevent self-reactivity and altered levels correlates with autoimmunity:

1. Elliot J, Higgins C. Major histocompatibility complex class I shedding and programmed cell death stimulated through the proinflammatory P2X₇ receptor *Diabetes* 2005; 53:212.
2. Lieberman SM, Evans AM, Han B, et al. Identification of the beta cell antigen targeted by a prevalent population of pathogenic CD8⁺ T cells in autoimmune diabetes. *PNAS* 2003; 100:8384-8.
3. Palermo B, Campanelli R, Garbelli S, et al. Specific cytotoxic T lymphocyte responses against Melan-A/MART1, Tyrosinase and Gp100 in vitiligo by the use of major histocompatibility complex/peptide tetramers: the role of cellular immunity in the etiopathogenesis of vitiligo. *J Invest Dermatol* 2001; 117(2):326-32.
4. Markiewicz MA, Girao C, Opferman JT, et al. Long-term T cell memory requires the surface expression of self-peptide/major histocompatibility complex molecules. *PNAS* 1998; 95: 3065-70.
5. Fu Y, Nathan DM, Li F, Li X, Faustman DL. Defective major histocompatibility complex class I expression on lymphoid cells in autoimmunity. *J Clin Invest* 1993; 91: 2301-7.
6. Faustman D, Li XP, Lin HY, et al. Linkage of faulty major histocompatibility complex class I to autoimmune diabetes. *Science* 1991; 254(5039):1756-61.
7. Kingsbury DJ, Mear JP, Witte DP, Taurog JD, Roopenian DC, Colbert RA. Development of spontaneous arthritis in β 2-Microglobulin-deficient mice without expression of HLA-B27. *Arth Rheum* 43:2290-2296, 2000.

8. Nakanishi K, Kobayashi T, Komatsu Y, Kogawa N, Hagihara M, Tsuji K Synchronous decline of serum-soluble HLA class I antigen and beta cell function in insulin dependent diabetes mellitus. *Clinical Immunology and Immunopathology* 1997; 85: 246-52.

A body of literature supports the concept that the interruption in MHC class I and self peptide for T cell tolerance could be due to risk conferred by TAP1 and TAP2 genes.

Human literature:

1. Yu MC, Huang CM, Wu MC, Wu JY, Tsai FJ. Association of TAP2 gene polymorphisms in Chinese patients with rheumatoid arthritis. *Clin Rheumatol* 2004; 23(1):35-9.
2. Cesari M, Hoarau JJ, Caillens H, Robert C, Rouch C, Cadet F, Pabion M. Is TAP2*0102 allele involved in insulin-dependent diabetes mellitus (type 1) protection? *Hum Immunol* 2004; 65(8):783-93.
3. Runstadler JA, Saila H, Savolainen A, Leirisalo-Repo M, Aho K, Tuomilehto-Wolf E, Tuomilehto J, Seldin MF. HLA-DRB1, TAP2/TAP1, and HLA-DPB1 haplotypes in Finnish juvenile idiopathic arthritis: more complexity within the MHC. *Genes Immun* 2004; 5(7):562-71.
4. Takizawa K, Takeuchi F, Nabeta H, Hirohata S, Takeuchi A, Matsumura Y, Yamamoto K. Association of transporter associated with antigen processing genes with Behcet's disease in Japanese. *Autoimmunity* 2003; 36(3):161-5.
5. Pyo CW, Hur SS, Kim YK, Kim TY, Kim TG. Association of TAP and HLA-DM genes with psoriasis in Koreans. *J Invest Dermatol* 2003; 120(4):616-22.
6. Kanagawa S, Morinobu A, Koshiba M, Kageyama G, Hayashi N, Yoshino S, Tokano Y, Hashimoto H, Kumagi S. Association of the TAP2*Bky2 allele with presence of SS-A/Ro and other autoantibodies in Japanese patients with systemic lupus erythematosus. *Lupus* 2003; 12(4):258-265.
7. Correa PA, Molina JF, Pinto LF, Acros-Burgos M, Herrera M, Anaya JM. TAP1 and TAP2 polymorphisms analysis in northwestern Colombian patients with systemic lupus erythematosus. *Ann Rheum Dis* 2003; 62(4):363-5.
8. Zhang SL, Chabod J, Penfornis A, Reviron D, Tiberghien P, Wendling D, Toussiro E. TAP1 and TAP2 gene polymorphism in rheumatoid arthritis in a population in eastern France. *Eur J Immunogenet* 2002; 29(3):241-9.
9. Penfornis A, Tuomilehto-Wolfe E, Faustman DL, Hitman GA, DiMe (Childhood Diabetes in Finland) Study Group. Analysis of TAP2 polymorphisms in Finnish individuals with type 1 diabetes. *Hum Immunol* 2002; 63(1):61-70.
10. Penfornis A, Tuomilehto-Wolfe E, The DiMe (Childhood Diabetes in Finland) Study Group, Faustman DL, Hitman GA. Analysis of TAP2 polymorphisms in Finnish individuals with type 1 diabetes. *Hum Immunol* 2002; 63:61-70.
11. Fox RI, Tornwall J, Michelson P. Current issues in the diagnosis and treatment of Sjogren's syndrome. *Curr Opin Rheumatol* 1999; 11(5):364-71.
12. Fu Y, Yan G, Shi L, Faustman D. Antigen processing and autoimmunity. Evaluation of mRNA abundance and function of HLA-linked genes (Primary data not published elsewhere). *Ann NY Acad Sci* 1998; 842:138-55.
13. Martin-Villa JM, Martinez-Laso J, Moreno-Pelayo MA, Castro-Panete MJ, Martinez-Quiles N, Alvarez M, de Juan MD, Gomez-Reino JJ, Arnaiz-Villena A. Differential contribution of HLA-DR, DQ, and TAP2 alleles to systemic lupus erythematosus susceptibility in Spanish patients: role of TAP2*01 alleles in Ro autoantibody production. *Ann Rheum Dis* 1998; 57(4):214-9.
14. Kumagai S, Kanagawa S, Morinobu A, Takada M, Nakamura K, Sugai S, Maruya E, Saji H. Association of a new allele of the TAP2 gene, TAP2*Bky2 (Val577), with susceptibility to Sjogren's syndrome. *Arthritis Rheum* 1997; 40(9): 1685-92.

15. Ma L, Penforinis A, Wang X, Schoenfeld D, Tuomilehto-Wolf E, Metcalfe K, Hitman G, Faustman D. Evaluation of TAP1 polymorphisms with insulin dependent diabetes mellitus in Finnish diabetic patients. The Childhood Diabetes in Finland (DiMe) Study Group. *Hum Immunol* 1997; 53(2):159-66.
16. Yan G, Shi L, Fu Y, Wang X, Schoenfeld D, Ma L, Penforinis A, Gebel H, Faustman DL. Screening of the TAP1 gene by denaturing gradient gel electrophoresis in insulin-dependent diabetes mellitus: detection and comparison of new polymorphisms between patients and controls. *Tissue Antigens* 1997; 50(6):576-85.
17. Rau H, Nicolay A, Usadel KH, Finke R, Donner H, Walfish PG, Badenhop K. Polymorphisms of TAP1 and TAP2 genes in Graves' disease. *Tissue Antigens* 1997; 49(1):16-22.
18. Shi L, Yan G, Fu Y, Ma L, Penforinis A, Faustman D. Human Tap1 polymorphisms detected by denaturing gradient gel electrophoresis. *Tissue Antigens* 1997; 49:421-6.
19. Ma L, Penforinis A, Wang X, Schoenfeld D, Tuomilehto-Wolf E, Metcalfe K, Hitman G, Faustman D. Evaluation of Tap1 polymorphisms with insulin dependent diabetes mellitus in Finnish diabetic patients. The Childhood Diabetes in Finland (DiMe) Study Group. *Hum Immunol* 1997; 53:159-66.
20. Hohler T, Weinmann A, Schneider PM, Rittner C, Schopf RE, Knop J, Hasencelver P, Meyer zum Buschenfelde KH, Marker-Hermann E. TAP-polymorphisms in juvenile onset psoriasis and psoriatic arthritis. *Hum Immunol* 1996; 51(1):49-54.
21. Moins-Teisserenc H, Semana G, Alizadeh M, Loiseau P, Bobrynina V, Deschamps I, Edan G, Birebent B, Genetet B, Sabouraud O, et. al. TAP2 gene polymorphism contributes to genetic susceptibility to multiple sclerosis. *Hum Immunol* 1995; 42(3):195-202.
22. Gonzalez-Escribano MF, Morale J, Garcia-Lozano JR, Castillo MJ, Sanchez-Roman J, Nunez-Roldan A, Sanchez B. TAP polymorphism in patients with Behcet's disease. *Ann Rheum Dis* 1995; 54(5):386-8.
23. Wang F, Li X, Annis B, Faustman DL. Tap-1 and Tap-2 gene therapy selectively restores conformationally dependent HLA class I expression in type 1 diabetic cells. *Hum Gene Ther* 1995; 6:1005-17.
24. Wang F, Li X, Annis B, Faustman D. *In vitro* genetic therapy with Tap-1 or Tap-2 restores HLA class I presentation of self peptide in IDDM B cells. *Autoimmunity* 1995; 21:11.
25. Ploski R, Undlien DE, Vinje O, Forre O, Thorsby E, Ronningen KS. Polymorphism of human major histocompatibility complex-encoded transporter associated with antigen processing (TAP) genes and susceptibility to juvenile rheumatoid arthritis. *Hum Immunol* 1994; 39(1):54-60.
26. Djilali-Saiah I, Caillat-Zucman S, Schmitz J, Chaves-Vieira ML, Bach JF. Polymorphism of antigen processing (TAP, LMP) and HLA class II genes in celiac disease. *Hum Immunol* 1994; 40(1):8-16.
27. Middleton D, Megaw G, Cullen C, Hawkins S, Darke C, Savage DA. TAP1 and TAP2 polymorphism in multiple sclerosis patients. 1994; 40(2):131-4.
28. Nakanishi K, Kobayashi T, Murase T, Kosaka K. Lack of association of the transporter associated with antigen processing with Japanese insulin-dependent diabetes mellitus. *Metabolism: Clin Exp* 1994; 43(8):1013-7.

Mouse literature:

1. Yan G, Gu Y, Faustman DL. Reduced expression of Tap1 and Lmp2 antigen processing genes in the nonobese diabetic (NOD) mouse due to a mutation in their shared bi-directional promoter. *J Immunol* 1997; 159:3068-80.

2. Li F, Guo J, Fu Y, Yan G, Faustman D. Abnormal class I assembly and peptide presentation in the nonobese diabetic mouse. PNAS 1994; 91:11128-32.
3. Faustman D, Li XP, Lin HY, et al. Linkage of faulty major histocompatibility complex class I to autoimmune diabetes. Science 1991; 254(5039):1756-61

A growing body of literature supports the concept that dysfunction of the proteasome may contribute to autoimmunity:

Human literature (proteasome disruption in NF- κ B signaling):

1. Kuroda N, Mitani T, Takeda N, Ishimaru N, Arakaki R, Hayashi Y, et al. Development of autoimmunity against transcriptionally unrepressed target antigen in the thymus of AIRE-deficient mice. J Immunol. 2005 Feb 15;174(4):1862-70.
2. Uchida D, Hatakeyama S, Matsushima A, Han H, Ishido S, Hotta H, et al. AIRE functions as an E3 ubiquitin ligase. J Exp Med 2004; 199(2):167-72.
3. Guo D, Li M, et. al. A functional variant of SUMO4, a new I κ B α modifier, is associated with type 1 diabetes. Nat Genet 2004; 36(8):837-41.
4. Uchida D, Hatakeyama S, Matsushima A, Han H, Ishido S, Hotta H, Kudoh J, Shimizu N, Doucas V, Nakayama KI, Kuroda N, Matsumoto M. AIRE functions as an E3 ubiquitin ligase. J Exp Med. 2004 Jan 19;199(2):167-72.
5. Jury EC, Kabouidis PS, Aba A, Mageed RA, Isenberg DA. Increase ubiquitination and reduced expression of LCK in T lymphocytes from patients with systemic lupus erythematosus. Arth Rheum. 2003; 48:1343-1354.
6. Ding H, Cheng H, Fu Z, Yan L, Yang G. Relationship of large multifunctional proteasome 7 gene polymorphism with susceptibility to type 1 diabetes mellitus and DR3 gene. Chin Med J 2001 114(12):1263-6.
7. Fu Y, Yan G, Shi L, Faustman D. Antigen processing and autoimmunity. Evaluation of mRNA abundance and function of HLA-linked genes. Ann NY Acad Sci 1998 842:138-55.
8. Maskymowch WP, Jhangri GS, Gorodezky C, Luong M, Wong C, Burgos-Vargas R, Morenot M, Sanchez-Corona J, Ramos-Remus, C, Russell AS. The LMP2 polymorphism is associated with susceptibility to acute anterior uveitis in HLA-B27 positive juvenile and adult Mexican subjects with ankylosing spondylitis. Ann Rheum Dis. 1997; 56:488-492.
9. Deng GY, Muir A, Maclaren NK, She JX. Association of LMP2 and LMP7 genes within the major histocompatibility complex with insulin-dependent diabetes mellitus: population and family studies. Am J Human Genet 1995 46(2):528-34.

Mouse literature:

1. Hayashi T, Faustman D. Association of gender-related LMP2 inactivation with autoimmune pathogenesis. J Appl Physiol 2001; 91:2804-15
2. Hayashi T, Kodama S, Faustman D. LMP2 expression proteasome activity in NOD mice. Nat Med 2000; 6:1065-6.
3. Hayashi T, Faustman D. NOD mice are defective in proteasome production and activation of NF- κ B. Molec Cell Biol 1999; 19:8646-59.
4. Yan G, Gu Y, Faustman DL. Reduced expression of Tap1 and Lmp2 antigen processing genes in the nonobese diabetic (NOD) mouse due to a mutation in their shared bi-directional promoter. J Immunol 1997; 159:3068-80.

A growing scientific literature shows that the islets can regenerate in the pancreas, even in adult animals:

1. Meier JJ, Bhushan A, Butler AE, Rizza RA, Butler PC. Evidence for Sustained Islet Turnover in Humans with Long-Standing Type 1 Diabetes. Sustained beta cell apoptosis in patients with long-standing type 1 diabetes: indirect evidence for islet regeneration? *Diabetologia*. 2005 Oct 5; [Epub ahead of print].
2. Dor Y, Brown J, Martinez OI, Melton DA. Adult pancreatic beta-cells are formed by self-duplication rather than stem-cell differentiation. *Nature*. 2004; 429(6987):41-6.
3. Mathews V, Hanson PT, Ford E, et al. Recruitment of bone marrow-derived endothelial cells to sites of pancreatic beta-cell injury. *Diabetes*. 2004; 53(1):91-8.
4. Kodama S, Kuhlreiber W, Fujimura S, Dale EA, Faustman DL. Islet regeneration during the reversal of autoimmune diabetes in NOD mice. *Science* 2003; 302: 1223-1227.
5. Hess D, Li L, Martin M, et al. Bone marrow-derived stem cells initiate pancreatic regeneration. *Nat Biotechnol*. 2003 Jul;21(7):763-70.
6. Ianus A, Holz GG, Theise ND, Hussain MA. In vivo derivation of glucose-competent pancreatic endocrine cells from bone marrow without evidence of cell fusion. *J Clin Invest*. 2003 Mar;111(6):843-50.
7. Zorina TD, Subbotin VM, Bertera S, et al. Recovery of the endogenous beta cell function in the NOD model of autoimmune diabetes. *Stem Cells*. 2003;21(4):377-88.
8. Yang L, Li S, Hatch H, et al. In vitro trans-differentiation of adult hepatic stem cells into pancreatic endocrine hormone-producing cells. *PNAS* 2002; 99(12):8078-83.
9. Ryu S, Kodama S, Ryu K, Schoenfeld DA, Faustman DL. Reversal of established autoimmune diabetes by restoration of endogenous beta cell function. *J Clin Invest* 2001; 108: 63-72.

The spleen of diabetic mice contains precursor cells that can form insulin positive cells in diabetic animals. The spleen of mice can serve as a reservoir for islet cells or the spleen indirectly promotes pancreatic islet regeneration:

1. Yin D, Tao J, Lee DD, Shen J, Hara M, Lopez J, Kuznetsov A, Philipson LH, Chong AS. Recovery of islet beta-cell function in streptozotocin-induced diabetic mice: an indirect role for the spleen. *Diabetes* 2006 55(12):3256-63.
2. Tang D-Q, Cao L-Z, Sun Y-P, Yang L-J. In Vitro Generation Insulin-Producing Cells from Adult Spleen. American Diabetes Association's 65th Annual Scientific Sessions. 2005; Abstract # 1642-P
3. Kodama S, Davis M, Faustman DL. Diabetes and stem cell researchers turn to the lowly spleen. *Sci Aging Knowledge Environ*. 2005 Jan 19;2005(3):pe2.
4. Tang DQ et al. In vivo and in vitro characterization of insulin-producing cells obtained from murine bone marrow. *Diabetes*. 2004 Jul;53(7):1721-32.
5. Kojima H, Fujimiya M, Matsumura K, Nakahara T, Hara M, Chan L. Extrapancreatic insulin-producing cells in multiple organs in diabetes. *PNAS*. 2004 Feb 24;101(8):2458-63.
6. Kodama S, Kuhlreiber W, Fujimura S, Dale EA, Faustman DL. Islet regeneration during the reversal of autoimmune diabetes in NOD mice. *Science* 2003; 302: 1223-1227.
7. Krapp A, Knofler M, Ledermann B, et al. The bHLH protein PTF1-p48 is essential for the formation of the exocrine and the correct spatial organization of the endocrine pancreas. *Genes Dev*. 1998;12(23):3752-63.

The spleen of adult animals and humans contains precursor stem cells that have the potential to form new cells such as islets, salivary glands or bone:

1. Faustman D, Tran S, Kodama S, Lodde B, Szalayova I, Key S, Toth Z, Mezey E. Comment on Papers by Chong et al, Nishio et al and Suri et al on Diabetes Reversal in NOD Mice. *Science* 2006 314:1243-4.

2. Tran S, Kodama S, Mezey EM. Treatment success and regenerative mechanisms influenced by age of NOD mice and target organ of autoimmune attack. *Diabetes* 2006 Abstract #1202-P.
3. Kodama S, Davis M, Faustman DL. Diabetes and stem cell researchers turn to the lowly spleen. *Sci Aging Knowledge Environ*. 2005 Jan 19;2005(3):pe2.
4. Kodama S, Kuhlreiber W, Fujimura S, Dale EA, Faustman DL. Islet regeneration during the reversal of autoimmune diabetes in NOD mice. *Science* 2003; 302: 1223-1227.
5. Derubeis AR, Mastrogiacomo M, Cancedda R, Quarto R. Osteogenic potential of rat spleen stromal cells. *Eur J Cell Biol*. 2003 Apr;82(4):175-81.
6. Chadburn A. The spleen: anatomy and anatomical function. *Semin Hematol*. 2000 Jan;37(1 Suppl 1):13-21.
7. Hutchins RR, Hart RS, Pacifico M, Bradley NJ, Williamson RC. Long-term results of distal pancreatectomy for chronic pancreatitis in 90 patients. *Ann Surg*. 2002 Nov;236(5):612-8.
8. Govil S, Imrie CW. Value of splenic preservation during distal pancreatectomy for chronic pancreatitis. *Br J Surg*. 1999 Jul;86(7):895-8.
9. Lee BW, Tan SH, Lee WK, Yap HK, Aw SE, Wong HB. Glucose tolerance test and insulin levels in children with transfusion-dependent thalassaemia. *Ann Trop Paediatr*. 1985 Dec;5(4):215-8.
10. Bannerman RM, Keusch G, Kreimer-Birnbaum M, Vance VK, Vaughan S. Thalassaemia intermedia, with iron overload, cardiac failure, diabetes mellitus, hypopituitarism and porphyrinuria. *Am J Med*. 1967;42(3):476-86.

Previous trials in diabetic animals and humans have not employed appropriate dosing of BCG.

1. Shoda LK, Young DL, Ramanujan S, Whiting CC, Atkinson MA, Bluestone JA, Eisenbarth GS, Mathis D, Rossini AA, Campbell SE, Kahn R, Kreuwel HT. A comprehensive review of interventions in the NOD mouse and implications for translation. *Immunity*. 2005 Aug;23(2):115-26.

A fetal protein (transcription factor) called Hox persists into adulthood and facilitates adult islet regeneration

1. Kodama S, Davis M, Faustman DL. Regenerative medicine; a radical reappraisal of the spleen. *Trends Mol Med*. 2005;11(6):271-276.
2. Kodama S, Davis M, Faustman DL. Diabetes and stem cell researchers turn to the lowly spleen. *Sci Aging Knowledge Environ*. 2005;2005(3):pe2.
3. Rosanas-Urgell A, Marfany G, Garcia-Fernandez J. Pdx1 related homeodomain transcription factors are distinctly expressed in mouse adult pancreatic islets. *Mol Cell Endocrinol*. 2005;237:59-66.

Positive results with BCG in mice and humans.

1. Shehadeh N, Calcinaro F, Bradley BJ, Bruchlim I, Vardi P, Lafferty KJ. Effect of adjuvant therapy on development of diabetes in mouse and man. *Lancet*. 1994 Mar 19;343(8899):706-7.