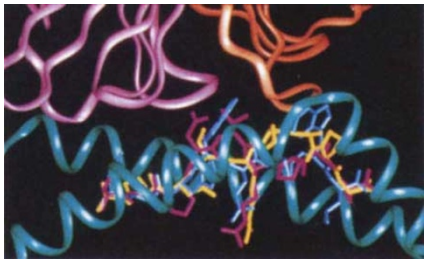
Developing transgenic frogs

The frog, *Xenopus laevis*, is a valuable model organism for examining gene expression, but the difficulty in regulating tissue-specific expression has limited its use as a developmental model. Although transgenic expression is easily accomplished by injecting mRNA directly into single cell embryos, the RNA is immediately translated and relatively unstable–factors that do not allow for temporal and spatial regulation. In general, DNA injection has not resulted in the sufficiently controlled regulation required for the analysis of the effect of heterologous gene expression on development. Fu et al. (see pp. 233, 253) have used a small genetic element from the adeno-associated virus—that enhances transgene expression in mammalian cells—to develop a strategy for the efficient and stable expression of transgenes under the control of either ubiquitous or tissue-specific promoters. The viral sequence apparently enhances the segregation of the injected DNA in a manner that allows tissue specific gene expression.

Vaccination against cholera, an infectious diarrheal disease, has been hampered by the difficulty of vaccine administration in the developing countries where it is most needed. Arakawa et al. (see p. 292) have engineered potatoes that can synthesize and deliver the bacterial antigen—cholera B toxin protein—in a manner in which its biological activity as well as tertiatry structure are maintained. Oral vaccination of mice using the potato vaccine prevented disease upon challenge with cholera toxin.

Sweet peptide mimetic



Tumor specific antigens are attractive targets for cancer immunotherapy. MUC1 is an epithelial mucin that is aberrantly expressed on human tumor cells in both its distribution and its glycosylation pattern. These differences make it a tumor-specific protein that is capable of generating a CTL response in mice that protects them from developing MUC1 expressing tumors. Apostolopoulos et al. demonstrate that a peptide mimic of a carbohydrate epitope of MUC1 is able to generate the same protective response, against the tumor-specific MUC1, in a mouse model (see pp. 236, 276).

Research briefs written by Philip Bernstein.

The veto of CD4⁺ T lymphocytes

Although enhancing an immune response has therapeutic value in treating pathogen infections or cancer, suppression of an immune response is needed to combat autoimmune disease or to facilitate organ transplantation. Most immunosuppressive strategies use drugs that target the entire immune system, which can compromise patient's immune response against pathogens. To target a specific arm of the immune system—$CD4^+$ T lymphocytes—Qi and Staerz have used hybrid antibodies to mimic the so-called veto effect (see pp. 237, 271). The hybrid antibody recognizes both MHC class II as well as CD4. Presumably, the single antibody recognizes both epitopes on the same cell, which somehow interferes with—vetoes—the activation of T-helper cells.

Transcriptional regulation is controlled, in large part, by the interactions of protein transcription factors with promoter elements in a gene. Protein–DNA interactions can be examined by techniques that do not interfere with this association, such as mobility shift gel assays. Although the technique is easy, it involves the use of labels—usually radioactive—and is slow. Using electrospray ionization mass spectrometry, Veenstra et al. (see p. 262) have been able to examine the metal requirements for the association of the vitamin D receptor with its cognate DNA binding domain.

Attracting antigen-specific T-cells

The adoptive transfer of T-cells for the treatment of cancer or infection requires the ability to isolate low frequency antigen-specific lymphocytes from a heterogeneous cell population. To this end, Luxembourg and colleagues (see p. 281) have used magnetic beads coated with MHC–peptide complexes that would normally interact with a specific T-cell receptor. Rare T-cells, or those with low affinity for the antigen, were isolated from mice and were functional both in vitro and in vivo.

Neurodegenerative disease may result from prolonged activation of excitatory neurotransmitter receptors. The unregulated influx of calcium through *N*-methyl-D-aparate (NMDA) gluatamate receptors leads to a variety of cellular responses, including apoptosis making these channels obvious drug targets. Using random peptide libraries, Montal and colleagues (see p. 286) have isolated peptide leads that prevent neuronal cell death elicited by excitotoxic compounds on hippocampal cultures.

A StEP up in molecular evolution

As the genomes from an increasing number of organisms are being sequenced and the structure of a variety of the encoded proteins are being solved, the rules for the design of proteins with enhanced characteristics are slowly being uncovered. While rational design approaches require understanding of structure–function relationships, molecular evolution—the selection of enhanced proteins in vitro—requires only the gene sequence. By forcing a PCR reaction to switch templates, a vast number of mutations (either naturally occurring or artificially introduced) can be incorporated into a single template in a random manner (see pp. 234, 258). Using this technique, Zhao et al. have evolved a mesophilic subtilisin into a thermophilic counterpart.

Sugimura and colleagues have isolated peptides from a random library expressed on phage that mimics the structure of the binding site of the T-cell costimulatory molecule CTLA4 (see p. 267). The peptide mimic, in the presence of antigen, is able to stimulate appropriately primed T cells and thus may serve as a platform for the development of immunostimulatory drugs.