The 12th International Immunology and Diabetes Workshop was held during April 1993 in Orlando, Florida, to review research progress since the 11th Immunology of Diabetes Workshop meeting in Nagasaki, Japan, one and a half years before. The NOD mouse may have as many as 10 susceptibility genes, including its novel IA major histocompatibility complex antigen and a defective interferon-γ receptor, whereas human IDDM is so far known to be encoded by cis and trans complementation products of certain DQ genes on chromosome 6q, and a gene in the insulin-like growth factor II region on chromosome 11p. A unique protein regulator of the X box promoter of the highly susceptible DQB1'0302 allele has also been found. Islet cell antibody negative siblings of IDDM patients appear to have lower than expected abilities to secrete insulin in response to intravenous glucose. Sera from patients before and/or after developing IDDM immunoprecipitate two native proteins of 64,000- and 38,000-M₄, glutamic acid decarboxylase (GAD₆₅) reacting to conformational epitopes. However, a multitude of other autoantibodies often reacting to denatured proteins through linear epitopes have also been identified. The first workshop for GAD antibody assays was successfully completed; however, the 38,000-M₄ antigen has not yet been identified. Other autoantibodies reactive to gangliosides and to sulfatides continue to be reported. Insulitis has come to be recognized as a sometimes protective event. Protective insulitis predominates in older lesions. It can be induced by as disparate means as tuberculin antigen administration, by interleukin-4 treatments, by transfer of T-cell lines generated in autologous mixed lymphocyte responses, and by immunization to insulin B-chain, whereas oral islet cell antigens, such as insulin, can delay diabetes onset in the NOD mouse. Although Th2 cells may be important in protective insulitis, the NOD may actually have a deficiency of Th1 cells. Encapsulated islets can function for months after transplantation, whereas xenogeneic islet grafts appear to be rejected through a CD4⁺ T-cell-mediated mechanism like the pathogenic destruction of islets seen in NOD mice. We summarize a few of the meeting highlights. Diabetes 42:1099–104, 1993

The first research meeting devoted to the immunology of diabetes was convened in 1976 in Philadelphia under the auspices of the JDF. Some 30 participants from Europe and the U.S. were involved. Islet cell autoantibodies had just been established as characteristic of IDDM patients, a finding that documented the autoimmune nature of the disease. This past April 1993, the 12th International Immunology and Diabetes Workshop was held in Orlando, Florida, under sponsorship of the ADA. More than 350 participants from many countries of the world presented their most recent research findings as relevant to the underlying genetics, pathogenesis, predictability, and eventual prevention of the disease. Progress in islet cell and pancreatic transplantation was a new feature of this meeting. We present a synopsis of the meeting emphasizing areas that proved to be the most active foci for discussion and enlightenment.

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JDF, Juvenile Diabetes Foundation; ADA, American Diabetes Association; IDDM, insulin-dependent diabetes mellitus; GAD, glutamic acid decarboxylase; MHC, major histocompatibility complex; IL, interleukin; IgG, immunoglobulin G; IGF-II, insulin-like growth factor II; HLA, human leukocyte antigen; IAA, insulin autoantibody; AMLR, autologous mixed lymphocyte response; TNF-α, tumor necrosis factor-α; IFN-γ, interferon-γ; CFA, complete Freund's adjuvant; ICA, islet cell antibody; BSA, bovine serum albumin; PARP, polyadenylated RNA polymerase; PARP, polyadenylated RNA polymerase; PARP, polyadenylated RNA polymerase; TGF, transforming growth factor; GLUT, glucose transporter; ALX, alloxan.
TABLE 1
IDDM and its genes

<table>
<thead>
<tr>
<th>Candidate gene</th>
<th>Species</th>
<th>Chromosome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Class II MHC</td>
<td></td>
<td></td>
</tr>
<tr>
<td>DQA/DQB</td>
<td>Human</td>
<td>6</td>
</tr>
<tr>
<td>Regulator X box</td>
<td>Human</td>
<td>6</td>
</tr>
<tr>
<td>? TAP2 (transporter gene)</td>
<td>Human</td>
<td>6</td>
</tr>
<tr>
<td>IAβ</td>
<td>NOD mouse</td>
<td>17</td>
</tr>
<tr>
<td>Others</td>
<td></td>
<td></td>
</tr>
<tr>
<td>High affinity Fc receptor</td>
<td>NOD mouse</td>
<td>3 telomeric</td>
</tr>
<tr>
<td>?IL-2</td>
<td>NOD mouse</td>
<td>3 centromeric</td>
</tr>
<tr>
<td>INF-γ receptor</td>
<td>NOD mouse</td>
<td>10</td>
</tr>
<tr>
<td>Insulin/IGF-II</td>
<td>Human</td>
<td>11</td>
</tr>
<tr>
<td>Known genomic intervals</td>
<td>NOD mouse</td>
<td>1, 2, 3, 6, 11, 15</td>
</tr>
</tbody>
</table>

GENETICS

Acknowledging the inherent difficulties in studying diverse outbred human populations to locate the susceptibility genes of IDDM, numerous groups studied the NOD mouse hoping to gain insights that could apply to humans. Using a variety of strategies to link microsatellite-based genomic intervals and candidate genes to the occurrence of insulitis and diabetes in a number of NOD cross-bred mice, many of as 10 separate relevant loci were identified. The most important of these were the class II antigens of the MHC on chromosome 17 and two distinct loci found on chromosome 3. The candidate genes involved were the IA locus within the MHC, the structural gene for IL-2, and the high affinity Fc receptor gene for IgG (Table 1).

The predisposition to an inflammatory infiltrate within the pancreatic islets (insulitis) in some cases was distinct from that of diabetes. This could be seen for loci on chromosomes 1 and 3 in particular. Other suggested loci were mapped to chromosomes 6, 11, and 15, and resistance genes were preliminarily located on chromosomes 1, 2, 3, 5, and 18 in congenic and outbred strains of NOD mice. As is the case with other normal mice, e.g., those of b and/or s MHC haplotypes, the NOD expresses only one class II MHC antigen, because of a regulatory genetic defect in the promoter region of the IE a-chain gene. In previous studies, it has been shown that when the NOD expresses IE a/b-dimeric molecules through the means of IE a-transgenes or through breeding NOD mice congenic for IE a-genes, diabetes does not result. The protection appears to be only relative because outbred NOD IE a+ and IE a- congenic mice developed late-onset diabetes equally well and could be shown to be susceptible to diabetes after adaptive transfer of splenocytes from NOD mice.

It has been established that the IA of the NOD is unique, resulting from a b/d haplotype recombinant event, and has a Ser in position 57 of the b-chain instead of the more common Asp. The importance of the IA of NOD probably resides in its affinity to bind to a disease-specific islet cell peptide antigen because of particular motifs within its binding cleft. Thus, immune enhancement to the antigen or loss of immune tolerance might result. Similarly, the BB rat requires its U MHC haplotype for diabetes susceptibility, but 3 other recessive genes are thought to be involved as well. The BB lymphopenia gene has been mapped to a genomic segment close to the neuropeptide Y and carboxypeptidase genes. In humans with IDDM, only one gene, located 3' to the insulin gene and proximal to the IGFBP2 gene on chromosome 11 (INS locus), in addition to the HLA DR/DQ loci on chromosome 6, has been clearly identified to convey disease susceptibility.

Among Caucasians, DQ molecules that are cis and/or trans combinations of an a-chain with an Arg at position 52 and a b-chain with an Ala at position 57 encode susceptibility to IDDM. Typically, these susceptibility alleles occur on DR3 (DQA1*0501/DQB1*0201) and DR4 (DQA1*0301/DQB1*0302) HLA haplotypes, and DQA1*0102/DQB1*0602 is dominantly protective. However, among Japanese, IDDM is associated with DR4, 8, and 9 but not DR3. In Japanese, the disease can be phenotypically divided into acute versus slowly progressive subtypes. Acute IDDM was found to be associated with two DR4* haplotypes, i.e., DQA1*0301/DQB1*0401 (Asp+) and DQA1*0301/DQB1*0302, and one DR9* haplotype, DQA1*0301/DQB1*0303, whereas slow-onset IDDM was only associated with DQA1*0301/DQB1*0401. Thus, among Japanese, IDDM susceptibility is most often actually encoded by HLA haplotypes with a residue Asp*57 DQB1; however, such haplotypes usually have an Arg 52* DQA1 and an Asp*57 DRB1.

The case for a gender-related transmission bias for IDDM-associated HLA haplotypes continues to be inconclusive. However, a Danish report suggested that DQA1*0301/DQB1*0302 was preferentially passed from fathers and DQA1*0501/DQB1*0201 from mothers to affected offspring. Of great potential importance was the finding of specific regulator protein interactions with the X box associated uniquely with the DQB1*0302 gene. Thus, quantitative expression differences between IDDM-associated versus nonassociated class II HLA alleles might prove to be involved in the predisposition to IDDM. Another study has suggested that T-cell expression of class I MHC in IDDM patients as well as NOD mice is defective, albeit, this remains controversial. Thus, it was of interest that a negative association was reported with the peptide transporter 2 (TAP2) gene among French patients. Unfortunately, similar studies among Finnish, Norwegian, and Japanese patients found such TAP2 associations to be entirely secondary to linkage disequilibria with HLA-DQ/DR alleles already known to be linked to IDDM susceptibility.

Although the INS locus has limited polymorphisms, patients with IDDM have increased rates of homozygosity for the common allele in both DR4* and DR4+ patients. A complex interaction between the INS gene and DQ alleles appears possible, as does evidence for a paternal transmission bias for the IDDM-associated allele. The possibility of a maternal imprinting effect, where the maternal INS gene becomes inactivated, has been suggested but not proven. The presence of IAAs among nondiabetic relatives of individuals with IDDM has been reported to be associated with HLA-DR4, and the association may be strengthened by determination of the associated DQA1 alleles, according to one report. Inter-
Interestingly, patients with the Stiff Man Syndrome were found to have increased frequencies of DQB1*06 and DQ81*0201 alleles, with increases of the latter especially associated with coincident IDDM.

In summary, the search for the IDDM genes in humans is an active area for investigation. Those genomic intervals found to affect insulinitis and/or diabetes in NOD mice may assist in the location of the human counterparts to genomic regions that have synteny, or similarities, in genetic composition. More likely, however, the identification of the actual candidate genes within genomic intervals in the NOD mouse may provide functional information that could lead to the identification of similar dysfunctions in humans, most likely involving different underlying genetic lesions.

IMMUNOBIOLOGY

Although it is relatively easy to transfer disease to young NOD animals (<3 wk of age), it becomes increasingly more difficult to transfer disease as the recipient mice age. For this reason, disease transfer to older prediabetic mice requires irradiation beforehand. It was shown that development of the immune system in NOD animals involves the generation of both destructive T-cells as well as another set of T-cells that expresses the CD4+ phenotype and negatively regulates the disease process. Disease transfer to nonirradiated animals is possible, provided these protective cells are eliminated by treatment of recipient NOD mice with anti-CD4 antibodies. In an interesting study, pancreatic β-cells were destroyed in early life by ALX treatment. These studies demonstrated that the presence of active insulin-secreting β-cells were required for the activation of the disease process. The self antigens expressed by these cells were shown to drive the autoimmune process leading to β-cell destruction and diabetes. Thus, the disease could not be transferred from β-cell-depleted mice.

Although treatment of disease-prone NOD mice with nondepleting anti-CD4 antibodies protects NOD mice from their development of clinical disease, these animals still develop pancreatic insulinomas. Such insulinomas are nondestructive and is associated with the accumulation of lymphocytes in a peri-islet distribution. The mice, although protected from spontaneous diabetes, are still disease prone because diabetes can be rapidly precipitated after treatment with cyclophosphamide. Stimulation of the NOD immune system with CFA or BCG either early, or late (65 days) in the disease process also results in the development of nondestructive lesions in the pancreatic islets. These animals also can be shown to be disease prone, because diabetes can still be precipitated by cyclophosphamide treatment. In contrast, when NOD mice are protected by treatment with anti-class II antibody, diabetes cannot be induced by cyclophosphamide. These findings show that diabetes in the NOD mouse can be regulated by either immunostimulation or by immunosuppression. In most cases, the mice remain diabetes prone but develop nondestructive lesions in their pancreases. This negative regulation involves CD4+ cells that can be negated by cyclophosphamide treatments.

The AMLR in NOD mice has been found to be defective by several investigators. Autoreactive CD4+TCR-α/β+ T-cell clones were developed in AMLR response to syngeneic antigen presenting cells, which on cotransfer experiments protected the animals from severe intra-islet insulinitis and diabetes. These clones inhibited proliferation of T-cells responding to allogeneic stimulator cells without affecting IL-2 production. The inhibition was the result of growth arrest but not apoptosis of responder T-cells.

Whereas CD4+ T-cells are of critical importance to regulation, the relative roles of CD4+ and CD8+ T-cells in the destruction of pancreatic β-cells remains debatable. The findings of β-cell–destructive CD4+ T-cell clones clearly indicate the CD4+ T-cell alone is sufficient for the transfer of insulinitis and diabetes to NOD-scid mice. Some of the clones generated from the NOD insulinitis lesions showed strong reactivities to insulin (porcine and rat), indicating that insulin itself may be an autoantigen of importance in the disease. The CD8+ T-cell, however, appears to be required for the initiation of the disease, albeit, the function of this cell remains one of the outstanding enigmas in disease pathogenesis.

In pancreatic biopsies taken from Japanese who developed IDDM, CD8+ T-cells were the predominant cells in the islet infiltrates. Nevertheless, in adoptive transfer studies involving transgenic mice expressing influenza hemagglutinin on β-cells, CD4+ T-cells and F4/80+ macrophages dominated the early infiltrates, and in spontaneous diabetes in NOD mice, the lymphocytic infiltrates developed around a network of VCAM-1+ ICAM+ dendritic cells. In chimeric animals, insulinitis and diabetes could occur even when the responding T-cells were unable to recognize islet-specific antigens directly on β-cells, suggesting that direct contact between pathogenic CD4+ T-cells and β-cells is not required. TNF-α, given to NOD mice from the neonatal period, accelerated diabetes development, whereas treatments with antitNF-α antibodies powerfully prevented diabetes. Treatments with IFN-γ, however, had no effects. These findings coincided with a report that IFN-γ knock-out NOD mice have essentially normal rates of diabetes. Sequence analyses of peptides presented by the unique MHC class II molecules isolated from NON splenocytes also has been undertaken. Peptides from serum albumin, fibronectin, and ribonuclear proteins were eluted and sequenced. The serum albumin peptide demonstrated allele-specific binding to IA-NOD but not IAβ. It remains to be seen whether this kind of approach will prove useful in identifying novel autoantigens in IDDM.

ISLET CELL ANTIGENS AND IDDM PREDICTION

The islet cell antigens that are targeted by the autoimmune process that leads to IDDM have been identified by T-cell or autoantibody reactivities. The number of putative islet autoantigens in IDDM has grown dramatically over the past 2–3 yr, demanding continued evaluation of their potential roles in the pathogenesis of the disease. When islet cells are metabolically labeled in vitro and their detergent lysates exposed to sera from patients with IDDM or from those who subsequently developed IDDM,
only two protein antigens of 64,000 and 38,000 \( M_r \) can be regularly immunoprecipitated. This suggests that these proteins must be in their native configuration and that the reactive epitopes are likely to be conformational in nature. Arguably, this would imply a greater likelihood for a primary pathogenic role for these autoantigens, because the formation of their respective autoantibodies might be expected to involve whole rather than denatured proteins if a T-cell response to them occurred as an inductive event in the disease (Table 2).

The 64,000-\( M_r \) antigen has been completely identifiable as the lower isoform of GAD (GAD\(_{65} \)). Several groups have identified ICAs to be often reactive to the GAD\(_{65} \) antigen where ICA absorption can be accomplished by whole recombinant GAD\(_{65} \) but not by GAD fragments, confirming reactivity to conformational epitopes. GAD antibodies also were found to react to a linear epitope; a finding that might imply a secondary autoimmunization by release of a denatured form of GAD following \( \beta \)-cell lysis. The two isoforms of GAD have an ~70% homology. However, most investigators to date have found only modest frequencies of autoantibodies to the higher molecular weight GAD\(_{67} \), in IDDM patients. In other instances, ICA cannot be removed by reacting positive sera with recombinant GAD\(_{65} \), indicating that other antigenic reactions are involved, especially in IDDM patients who are proceeding towards or have actually developed IDDM. The 38,000-\( M_r \) antigen may be one such antigen, but its structure has yet to be identified. Glycolipids have long been recognized as an ICA antigen. Recently, a GM\(_2\)-1 ganglioside has been specifically implicated as an ICA antigen. Sulfatides represent another possible candidate with high frequencies of anti-sulfatide autoantibodies reported in IDDM. Another antigen of interest is a 69,000-\( M_r \), islet cell protein with homology to BSA, recently implicated as an environmental trigger for IDDM through molecular mimicry. Yet others include a 52,000-\( M_r \) protein with homologies to a rubella capsid antigen, a 62,000-\( M_r \) heat shock protein, membrane-associated proteins of 155,000 and 160,000 \( M_r \), and a 37,000- to 40,000-\( M_r \) glycoprotein seen in trypptic digests of islet cell preparations. Peripheral blood monocytes have been found to react to GAD\(_{65} \), islet cell 38,000-\( M_r \), protein, and uncharacterized antigens of 32,000, 55,000-72,000, and 120,000-170,000 \( M_r \). In the NOD mouse, T-cell reactivity to GAD, insulin, peripherin, heat shock protein 60, and carboxypeptidase H has been found beginning near the time of weaning.

The ability to predict impending IDDM both in relatives of affected patients as well as in general populations of school children through ICA analysis has been demonstrated by many groups. The youngest antibody positive individuals, those with higher titers, those with the IDDM-associated DQ alleles, and those with more than one IDDM-associated antibody are at greatest risk of progression to clinical disease. The greatest impediment to consistency with such studies, however, remains with the variability between laboratories in their sensitivity to measure lower titers of ICA, especially of \( \leq 20 \) JDF U. Hopefully, future developments in the GAD autoantibody methodology will permit a more screening-reproducible test. To that end, the first workshop for GAD autoantibodies was held and revealed that the many different assays had fair specificities but limited sensitivities. Assays based on whole recombinant GAD\(_{65} \), especially those using eukaryotic expression systems, should have improved sensitivities in the near future. It remains unclear at what age the pathogenic process of IDDM begins or what induces it. One study indicated that IAA could appear as early as birth when the father had IDDM, whereas others had found that coxsackie virus might be an inductive agent because its P-2C protein has homologies to GAD. GAD antibodies follow coxsackie infections, and experimental immunizations against coxsackie proteins induced anti-GAD responses.

**PATHOGENESIS AND PREVENTION**

It has been shown previously that immunosuppressive drugs can be used to induce metabolic remissions in newly diagnosed patients. However, more recent studies have shown that such remissions tend not to be sustainable over time despite continuation of the therapy. An open pilot study of low dose cyclosporin in 3 of 4 high-risk ICA\(^+ \) French children showed improvement in their first-phase insulin responses to intravenous glucose. Whereas the potential for adverse side effects, especially those of viral-associated neoplasias (e.g., Epstein-Barr virus related B-cell lymphoma), inhibit widespread trials of this sort, there is increasing interest in less invasive approaches (Table 3).

The immunosuppressant mycophenolic acid is an IMP dehydrogenase inhibitor that depletes GTP, especially in activated lymphocytes, and arrests their DNA synthesis with only limited potential for neoplasia induction. When given to BB rats, the agent can both prevent diabetes and inhibit insulin. Although it had been shown that antibody therapy prevented insulin and diabetes when given to NOD mice to deplete them of CD4\(^+ \) T-cells, long-term protection from diabetes (? tolerance to islet...
cell antigens) could also be induced by nondepleting anti-CD4+ antibodies if given to young animals.

A common denominator to immunologically mediated islet cell destruction may be the damage inflicted by superoxides and nitric oxide. Both mediators inflict DNA breaks, which are repaired through the action of PARP. This repair process is accompanied by a fall in β-cell NAD. Nicotinamide is an agent that can replenish intracellular NAD and, at high doses, may inhibit PARP. It can also inhibit IL-1-induced nitric oxide formation by rat islets. High doses of nicotinamide also can delay onset of diabetes in NOD mice. The long-term inhibition of PARP by high doses of nicotinamide carries the theoretical risk of neoplasia, raising concerns that such a risk needs to be considered in human trials. After preliminary findings in ICA+ relatives of patients with IDDM that administration of the vitamin may delay onset of IDDM, a large population-based study has been initiated in New Zealand school children who have screened positive for ICA. One premise of the study is that ICA in such children will predict IDDM similarly to that among relatives, and such appears to be the case in the independent Florida studies. To date, the ICA+ New Zealand school children treated with nicotinamide do appear to have a lower than expected rate of IDDM. This rate is less than that observed in historical controls, or in that of a cohort followed in parallel that was not tested or treated. Other antioxidant inhibitors, such as probucol and its derivatives, have antidiabetic properties in rodents, but to date no human trials have been reported.

Another related approach being explored in rodents involves inhibition of the inducible form of nitric oxide synthase using aminoguanidine. β-cell production of nitric oxide can be induced with IL-1 and TNF-α, and potentially damaging levels of nitric oxide can be generated by macrophages within the insulitis lesions. Whereas aminoguanidine has minimal side effects, it may not be applicable to human IDDM because it remains unclear whether human macrophages can actually liberate the quantities of nitric oxide seen from rodent macrophages. Thus, nitric oxide might not have a pathogenic role in the IDDM seen in humans.

Polyinosine-polycytidine is a powerful inducer of IFN-α which when administered to BB rats can accelerate their diabetes, albeit, opposite effects have been reported in NOD mice. A transgenic mouse expressing IFN-α in islets develops diabetes accompanied by insulitis, an outcome that is inhibitable by administration of an antibody to the cytokine. It has been reported previously that β-cells of newly diagnosed IDDM patients often express IFN-α, a finding that led to the hypothesis that viruses could be initiating the lesions. Taken together, these findings could argue the case for the therapeutic potential for antibodies to IFA-α to prevent human IDDM.

In several laboratories, it has become clear that not all insulitis is destructive. Such was evident in murine genetic studies where insulitis could be mapped separately from diabetes, as well as after the administration of CFA to NOD mice. CFA is a lipoid adjuvant containing tuberculosis antigen that has been shown to induce an increase in intra-islet IL-4 producing T-cells (Th2), and a reduction in potentially damaging IFN-γ T-cells (Th1 and CD8+ cells). Thus, the concept began to emerge that β-cell injury might be linked to a Th1 (IL-2, IFN-γ, delayed hypersensitivity) function and protection with Th2 cells (IL-4, IL-7, IL-10, TGF-β, antibody formation) property. However, one group reported that the insulitis lesions of the NOD were relatively devoid of T-cells expressing mRNA for IL-2, indicating an age-related predominance of Th2 cells in the infiltrate. In this regard, it was interest that immunization of NOD mice with B-chain insulin but not A-chain insulin in IFA could powerfully prevent diabetes but not insulitis. The protection so induced could be transferred by splenocytes in cotransfer experiments. The study evoked images of future trials of vaccination by islet cell antigen fragments to prevent IDDM. Treatments of NOD mice with IL-4 could also be used to prevent diabetes. It has also been shown by two groups that oral feedings of insulin or GAD can induce a delay in the onset of diabetes in NOD mice. The effect is thought to be attributable to an active antigen-specific tolerogenesis that induces regulatory T-cells to traffic to the islet to encounter their target antigen. By so doing, they may release inhibitory cytokines such as IL-4, TGF-β, and/or IL-10, and inhibit local ongoing inflammatory responses through a nonantigen specific bystander effect. One study even suggested that oral feedings of glucagon may be effective, suggesting that the tolerogenic antigen needs be organ specific but not necessarily involved in the disease process.

TRANSPLANTATION BIOLOGY

There are qualitative differences between the hosts' reaction to allografts versus xenografts. In the case of the allograft response, both CD4+ and CD8+ T-cells are involved, but in some particular cases, CD4+ T-cells are not required. In contrast, in xenogeneic responses, the CD8+ T-cell plays a less prominent role. For example, the CD8+ T-cell is not involved in the destructive process of rat islets when transplanted into mice. Thus, allogeneic graft rejection is CD8+ T-cell-dependent, whereas that of xenogeneic graft rejection is CD4+ T-cell-dependent. Thus, it would appear that the xenograft reaction is akin to the disease process underlying IDDM, and is the result of nonclass I MHc-restricted inflammatory tissue damage.

This shift in emphasis concerning xenogeneic reactions is important because in the past, the xen reaction has been considered a more intense but otherwise similar form of the allograft response. It has been clear for years that immunological tolerance can be induced in animals by their treatment at the perinatal phase of development. This has been most readily demonstrated in rodents where a brief tolerogenic window after birth has been shown. However, tolerance may also be induced in adult animals as well. Tolerance can be maintained by a passive process, too, such as clonal deletion of potentially reactive T-cells or by their being rendered anergic. Tolerance can also be maintained by active mechanisms by which the destructive function of the immune system is negatively regulated or suppressed.
This latter form of tolerance is observed when antigen is introduced into the immune system of adult animals in an appropriate form to facilitate it. When fibroblasts are made transgenic for the expression of class I MHC antigen, a suppressive form of tolerance can be induced in adult animals when the cells are grafted into them. These studies demonstrate that an active suppressive form of tolerance can be induced in adult animals and that the way the antigen is introduced can dramatically affect the outcome. That is to say, the means of antigen presentation to the immune system may determine whether an immunizing or tolerizing response results. The antigen dose, the route of injection, and the form of antigen presentation are all factors that influence the outcomes in such situations. The fact that tolerance can be induced even in adult animals offers much hope for the treatment of diabetes by islet grafting in the future, but much more work is needed to elucidate the mechanisms responsible for the maintenance of tolerance once induced.

The relative roles of macrophages versus dendritic cells in antigen presentation in inflammatory responses versus graft rejection mechanisms appear to be important. It was shown that pig islets could function in NOD mice long term, provided that the mice were treated with anti-CD3 and CD4 antibodies and the grafts were exposed to high concentrations of oxygen before their transplantation. When the outcome was successful, the native islets were still often infiltrated, whereas the xenografts appeared to be relatively resistant to autoimmune attack. In BBZ/Wor rats, diabetes appeared to result from reduced \( \beta \)-cell mass from the autoimmune process as well as reduced \( \beta \)-cell responsiveness to glucose as a result of GLUT2 loss. It is not known whether the finding would extend to other models or to islet cell transplant situations. IL-1 has been shown to induce an inducible form of nitric oxide synthase within islet cells, raising the possibility that toxic levels of nitric oxide may arise within these cells when undergoing islet rejection or autoimmune attack. Mice expressing the EBV receptor CR2 in their \( \beta \)-cells were found to develop a peri-islet infiltrate but not an intra-islet inflammation or diabetes, suggesting that another signal event might be required for this to occur.

Progress continues in allogeneic and xenogeneic islet transplantation using hybrid pancreatic devices in diabetic rats and dogs. Such transplantations exclude the immune system from direct attack and have functioned for as long as a year. One group reported long-term success with pig islets in a single coiled acrylic capillary implant in ALX-treated rabbits. Of the studies reported, the predominant long-term hope for the restitution of islet cell function in patients with longstanding IDDM may lie with xenogenic grafts.

**SUMMARY**

The number and complexity of genetic susceptibilities in the NOD mouse and patients with IDDM is growing rapidly, with a dichotomy seen in respect to the predispositions to insulitis and/or diabetes. Insulitis in many circumstances was seen to be protective, a type of response that appeared to increase relatively with age in the NOD mouse and could be induced by several means, such as treatment with BCG/CFA; by perturbations of the phenotype of invading cells, especially in respect to their expressed cytokines; through orally induced tolerance to islet antigens; or even by tolerogenesis induced by active immunization to islet antigens in IFA. Much progress has been made in respect to the identification of islet cell antigens targeted by the autoimmune process, and their relative pathogenic roles need examination. Various assays for GAD have appeared and hold promise of improved methods of screening persons susceptible to IDDM. The 38,000-M, antigen, however, still needs elucidation. Many potential treatments to prevent IDDM have appeared, and human trials have begun. Progress in islet cell transplantation continues to be made, but functional xenogeneic grafts in diabetic humans still appear distant. We look forward to further progress to be reported at the 13th meeting scheduled to take place in Paris, France, in June 1994.

**ACKNOWLEDGMENTS**

This synopsis was culled from a large number of research presentations that cannot be individually recognized here. Our thanks is extended to all of them for their original contributions.