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CONCEPTS AND METHODS IN IMMUNOLOGY APPLICABLE TO THE CONTROL OF HUMAN FERTILITY

By
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ABSTRACT

The reproductive event has been operationally subdivided into three periods, respectively including gametogenesis, fertilization and pregnancy: the feasibility of manipulating each period is discussed. A note of caution arises from our present incomplete understanding of the nature of the materno-foetal immunological relationship, and from the danger of producing irreversible effects or of leading to autoimmunity.

This paper is not intended to be a review or a summary of this Karolinska Symposium. In fact, this task would be too difficult, since many of the contributors have themselves given careful reviews of their own fields. Rather, I shall try to convey the impressions, the reactions and the evaluations of an immunologist – until now an outsider in the field of fertility control – suddenly exposed to an avalanche of information, hypotheses and questions.

There is no need to emphasize either the importance of the ultimate goal – the control of fertility – or the difficulty of assessing the possible contribution of immunology to that goal. In fact, everybody has noticed the growth and expansion of this scope of our discipline in recent years. This expansion has been accompanied by substantial progress in many theoretical aspects which has clarified our views on several mechanisms of the immune response, as Brigitte Askonas (1974), J. F. A. P. Miller (1974), G. J. V. Nossal (1974) and B. Pernis (in discussion at this Symposium) have already shown. However,
there are mechanisms that seemed to be clear ten years ago that are now under question. For example, the phenomenon of tolerance is still poorly understood, and tolerance happens to be very relevant to the present task. An appropriate quotation from Nossal (to be published) dramatically states: "All the classic tolerance experiments are in the midst of a somewhat agonizing reappraisal". What is the role of T and B tolerance in non-responsiveness to a variety of antigens? What is the importance of specific humoral antibodies in blocking the cellular immune responses? What is the role of soluble antigen, or of soluble antigen-antibody complexes? The lack of unanimity in answering these questions is no doubt temporary, but is certainly an obstacle to the application of immunological methods in the near future.

Let us now examine which are the steps in the reproductive process that may be susceptible to immunological intervention.

I have divided the process between gamete formation and parturition into three periods by drawing two artificial boundaries – as shown in Fig. 1 – one through the ovulation/ejaculation events, the other through the implantation event. Period I comprises the process of oogenesis and spermatogenesis. Period II includes the transport of the ovum and the spermatozoon in the female genital tract, their meeting and fertilization and the migration of the zygote into the uterine mucosa. Period III constitutes the period of pregnancy between implantation and birth.

Since immune responses are rather slow, and since the acceleration that can be seen in a secondary response is not as impressive in cell-mediated immunity as in humoral responses, the duration of each period may have an impact on the success of immunological control.
Table 1.
Timetable of reproduction.

<table>
<thead>
<tr>
<th></th>
<th>Time (hours)</th>
<th>Possible mucosal IR</th>
</tr>
</thead>
<tbody>
<tr>
<td>SPERMATOGENESIS</td>
<td>1000</td>
<td>-</td>
</tr>
<tr>
<td>(staminal all spermatozoa)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>- OOGÉNESIS</td>
<td>2000</td>
<td>-</td>
</tr>
<tr>
<td>(diplotene oocytes → ovum)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>- OVULATION</td>
<td>6-24</td>
<td>+</td>
</tr>
<tr>
<td>(→ fertilization)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>- INSEMINATION</td>
<td>6-72</td>
<td>+</td>
</tr>
<tr>
<td>(→ fertilization)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>- FERTILIZATION → IMPLANTATION</td>
<td>120</td>
<td>+</td>
</tr>
<tr>
<td>- PREGNANCY</td>
<td>6500</td>
<td>-</td>
</tr>
</tbody>
</table>

The first Table shows a subdivision of the three periods into events that can be "timed" quite precisely. By algebraic summation of the figures pertaining to each period the following proportions result:

\[ t_1 : t_{II} : t_{III} = 14 : 1 : 31 \]

I shall deal with the three periods separately, as suggested by the trend of the reports in this meeting and by recent literature. But instead of following the chronological order, I shall arbitrarily begin with the period which is most intellectually challenging (III) and end with the period that seems to be the most promising for manipulation by immunological techniques.

THE IMMUNOLOGICAL PARADOX OF PREGNANCY

Phylogenesis

At first glance, the emergence of placental pregnancy in the early mammals (including marsupials) in phylogenesis, when one considers that the immune system had, in fact, appeared much earlier, is rather unexpected. The conceptus becomes an actual allograft to the mother and since the chances of genetic compatibility in a natural population are almost non-existent, the rejection reaction should be strong. One has to admit that the selective advantage afforded by placental pregnancy is considerable, and that therefore nature has found ways (an efficient way) to overcome the formidable stumbling block of
a cell-mediated histocompatibility reaction. These considerations help characterize the effort of an immunologist trying to control fertility by tampering with period III. Pasteur and Salk merely extended or amplified the normal immune response; a kidney transplantor suppresses immune reaction. In this case the task is far more complicated – one has to remove a bypass mechanism which has arisen during a million years of evolution, without weakening the potential response.

Looking more deeply into the phylogenesis of pregnancy one is surprised to discover that well before the mammals and certain sharks, a number of cases of true pregnancy exist. They include Onichophora (Figs. 2 and 3 illustrate a Peripatus and the scheme of its placenta), certain rare insects without apparent order and certain scorpions (see Table 2).

As members of the placentata the Onichophora are particularly striking for their primitiveness. They are related to the trilobites, look somewhat like myriapods, slugs and earthworms and possess 14–43 pairs of rudimental legs equipped with tiny nails.
Table 2.
Placenta in phylogenesis.

1) ONICHO PHORA: A branch of the Artropoda that parted at a very early date from the main stock. Seventy different species living in equatorial, humid regions.

2) SALP IDAE: Protochordates, class thaliacea.

3) SCORPIONIDEA (ARACHNIDA) Only in some genera, where eggs are small, without yolk. The young develop in lateral sacs of the uterus, attached to the mother by a kind of placenta.

4) INSECTS: Some scattered examples (without any order): eggs not endowed with deutoplasm, fed through primitive placentae.

5) SELACHIANS: A number of sharks have a vitelline placenta and are viviparous.

6) REPTILES: Some living (seps, lacertidae) and many extinct (Ichthyosaurus).

7) MAMMALS: Practically all, from insectivors and marsupials, to primates.

At such an early stage of species evolution there was no immunological obstacle to cope with, yet the generalization of placenta did not happen until much later, when the obstacle appeared.

Two explanations could be offered, i.e., either the selective advantage of pregnancy is strong only in warm-blooded terrestrial animals, or the existence of a developed immune system – once local problems are solved – is advantageous and even necessary in establishing a healthy mother-foetus relationship. One of the more obvious assets would be to prevent the mutual invasion of cells filtrating through the placenta.

The homograft reaction

The problem of how the incompatible foetus can survive in the womb of the mother is not completely solved. The problem is particularly interesting because it can serve as a model to help to explain why tumours and parasites are not rejected by the host.

Many hypotheses (see reviews by Billingham & Silvers 1972; Beer & Billingham 1974; Hellström et al. 1969) have been put forward in recent years, some of which have been discussed earlier in this Symposium (see Nossal 1974; Edidin et al. 1974). It may be useful to have a simplified list of the hypotheses, with an evaluation of their relative importance (Table 3).
Table 3.

Hypotheses that may explain the survival of the foetus as a homograft.

<table>
<thead>
<tr>
<th>Hypothesis</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>1) There is complete separation of vascular systems</td>
<td>not true</td>
</tr>
<tr>
<td>2) The foetus is antigenically immature</td>
<td>it is immunogenic</td>
</tr>
<tr>
<td>3) The immunological reactivity of the mother is weakened during pregnancy</td>
<td>doubtful</td>
</tr>
<tr>
<td>4) &quot;True tolerance&quot; of the mother versus the foetus is established</td>
<td>no</td>
</tr>
<tr>
<td>5) The uterus is a privileged site</td>
<td>no</td>
</tr>
<tr>
<td>6) A balance between humoral and cellular results in blocking or &quot;enhancement&quot; of the foetal graft</td>
<td>some evidence</td>
</tr>
<tr>
<td>7) A physiological barrier exists at the trophoblast – mother interface</td>
<td>some evidence</td>
</tr>
</tbody>
</table>

Point 1 is an old belief which is now known to be erroneous.

Point 2 does not affect the potential immunogenicity of foetal tissues; one has a different spectrum of antigens exposed, which only partially overlap with the adults, but they are immunogenic.

Point 3 is controversial. There are a number of reports suggesting that there is a somewhat weakened response to non-specific primary skin allografts in certain species (rabbit) but not in others (mouse, cattle). Studies of in vitro reactivity (e.g. lectin stimulation of cells from pregnant individuals) have also failed to yield unanimous results. These admittedly unconvincing effects have been attributed to a mediated and even to a direct (Adcock et al. 1973) action of hormones, in particular chorionic gonadotrophin.

Point 4. There is no true tolerance: a systemic mother anti-foetus response has been demonstrated.

Point 5 has been tested, as reported by Beer &Billingham (1974), by grafting skin cells to the endometrium of rats. The grafts from incompatible strains were rejected (although their survival was prolonged in the presence of a decidual response).

Point 6. A balance between homograft rejection and enhancement as an explanation of tolerance (alternative to clonal suppression) was suggested by Voisin as early as 1961 (1962), demonstrated by the Hellström in tumour growth (Hellström et al. 1969) and invoked by Ceppellini (1970) ("a subtle interplay between humoral and cell-bound immunity").
Evidence stems from (a), the observation by Beer & Billingham (1974) that runt disease, which normally appears in newborns when the prospective mother is sensitized by a skin graft against the father, can be avoided in two conditions, both of which are likely to have brought about a more efficient humoral response: when the mother has already raised a litter before sensitization, or when a period longer than 10 days elapsed between skin rejection and mating; and, (b), the work of Youtananukorn & Matangkasombut (1973), who found that blood lymphocytes from post-partum women exhibit inhibited migration in the presence of placental antigens and that plasma contains a soluble factor that specifically suppresses this phenomenon (Fig. 4). Preliminary data would indicate that the factor is an antigen (possibly HLA) or soluble antigen-antibody complex (personal communication). (c), the soluble factors

![Cell migration graph]

**Fig. 4.**

*Youtananukorn & Matangkasombut* (1973): Reactivity of peripheral leucocytes from postpartum (top) and nulligravidous (bottom) women to pooled placental antigens and PPD, and the effect of autologous plasmas.
A, no antigen; B, pooled placental antigens; C, PPD.

a, normal plasma; b, autologous plasma.

Cell migration is expressed as a weight of paper within projected cell boundaries.
Table 4.
Effect of anti-embryo and anti-tumour immunity on the pregnancy of BALB – C mice. (Data by Giorgio Parmiani).

<table>
<thead>
<tr>
<th>Prospective mothers sensitized with</th>
<th>Rate of pregnancy</th>
<th>%</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Syngeneic adult tissues</td>
<td>41/47</td>
<td>87</td>
<td></td>
</tr>
<tr>
<td>Allogeneic adult tissues</td>
<td>16/18</td>
<td>87</td>
<td></td>
</tr>
<tr>
<td>Syngeneic embryo tissues</td>
<td>11/28</td>
<td>39</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Allogeneic embryo tissues</td>
<td>15/23</td>
<td>65</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>MAC – induced fibrosarcoma</td>
<td>42/75</td>
<td>56</td>
<td>&lt; 0.005</td>
</tr>
<tr>
<td>SV40 fibrosarcoma</td>
<td>11/21</td>
<td>51</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>Spontaneous sarcoma</td>
<td>21/33</td>
<td>60</td>
<td>&lt; 0.01</td>
</tr>
</tbody>
</table>

found in pregnant or multiparous women that are able to block many combinations of Mixed Lymphocyte Culture reaction. The painstaking analysis of one of these sera by Gatti and collaborators resulted in the discovery that anti-HLA antibodies (specific for the husband) were responsible for the inhibition (unpublished).

As clinical proof of a maternal anti-conceptus immune response it is interesting to quote the findings of Kenneth S. Tung, who very recently observed immune complexes consisting of IgG and unknown antigens deposited in the glomeruli of pregnant mice and guinea pigs. It is interesting to note that in mice this phenomenon occurs also in inbred strains, implying that “foetal” antigens may be involved (1974).

Point 7. The very fact that under particular sensitization conditions (I refer to the experiment by Beer & Billingham mentioned above) immune “killer” cells can pass through the placenta and damage the conceptus, but are not able to cause serious damage to the placenta and certainly do not cause its rejection, points toward some “local” privilege.

Attention throughout the search for the site of this rejection block has been focused on the trophoblast – the layer of foetal cells in immediate contact with the maternal tissue in the placenta. Experiments have shown that, if transplanted, it does not elicit a homograft rejection and is not damaged by an immune response which is artificially stimulated. The ease of growth and dissemination of tumoral trophoblastic tissue in choriocarcinoma is another example of its peculiar resistance. Concerning the mechanism of protection, it has been suspected that the presence of the glyocalix on the trophoblast
cells serves to mask the histocompatibility antigens and also repel lymphocytes. An interesting alternative (Matangkasombut, private communication) proposes the existence of an "antigen gradient": the trophoblast cells would secrete substantial quantities of glycoprotein – polysaccharide antigens, the concentration of which would be highest near the secreting cell, and this would afford maximum protection by blocking of receptors and antibody sites or by formation of complexes in antigen excess (for description of these mechanisms in the case of tumour growth the reader is referred to Nossal 1974). There are as yet no direct proofs for this interesting hypothesis which would unify the "humoral balance" of point 6 and the local protection theory. Indirect support is provided by studies in Schistosoma immunology by Cioli and Ruppel (personal communication), suggesting that the relative impurity of the parasite may be connected with the observed rapid turnover of the surface antigens.

Possible manipulation of pregnancy

Modern immunological methods (e. g., immunodialysis against insolubilized anti-hormone heterologous antibodies) may find a somewhat "science fiction" application in interfering with the hormonal balance that is essential for gestation. Csapo, Dray & Erdős (to be published) have recently shown that while injection of rabbit anti-progesterone into rats has a moderate effect, association with anti-17β-oestradiol (E2) provokes a surprisingly high rate of abortion.

The historical review of Stevens (1974) has listed a great number of experiments, mainly centered around the passive immunization of the pregnant female against placental antigens, in particular placental hormones. It is certainly possible to cause abortion by injecting pregnant animals with hetero-antibodies directed against placental extracts, but only antibody transfer from a member of the same species or an active immunization would be eventually amenable to human application.

The first type of treatment failed in the hands of Beer et al. (1972) who observed no effects on pregnancy after the injection of antisera directed against rat trophoblast, produced in male rats. On the other hand, it looked promising when Stevens et al. (1971) obtained abortion in pregnant baboons injected with baboon antihuman placental lactogen sera.

Chorionic hormones were an obvious target. However, frequent cross-reactions (intra- and interspecies) were observed between the chorionic hormones, HPL, HCG and the mother's hormones, in particular luteinizing hormone. These cross-reactions, for example in the case of LH to the subunit of HCG, are potentially dangerous, in that both passive and active immunization attempts could cause general disturbances and in case of the latter even lead to autoimmunity. It is clear that not enough is known about the immune rela-
tionship between mother and foetus for anyone to dare to attack this central point. Although no practical applications can be hoped for for a long time, it would be of great value if this area could be clarified, since – as we heard – pregnancy may well be a model for tumour growth and parasitic life.

Another point of contact between tumour immunology and reproduction processes is the appearance, in probably all neoplastic cells, of antigenic specificities present in the embryo but absent in the mature organism. Can this remarkable effect of unmasking or derepression be utilized for cross-immunization? Baldwin (personal communication) failed to obtain tumour rejection in rats immunized against foetal antigens while other authors, e.g. Edidin (1974) were successful. In the opposite direction, Parmiani & Della Porta observed that both mice and rats when immunized against tumour antigens had decreased fertility. Table 4 shows the results of unpublished experiments of these authors, comparing the effect of sensitization with adult, embryo and tumour tissue on prospective Balb-c mothers. The rate of pregnancy after the last two treatments is significantly reduced.

There seem to be two quite obvious ways to test the attractive hypothesis of a balance between cell-bound and humoral (or between T and B) responses. The first, suggested by Nossal (1974), is to test the rate of success of pregnancy in T-depleted and B-depleted animals. The second would test the importance of soluble antibodies, soluble antigens or complexes in keeping a block on rejection. One could prepare insoluble antibodies and insoluble antigens by the techniques of Avrameas (1974), make them available to the serum but not to the cells (e.g., by a semipermeable chamber located in the peritoneal cavity) and observe whether such an in vivo absorption would upset the equilibrium and precipitate rejection.

**IMMUNE CONTROL OF GAMETOGENESIS**

The long period I is very delicate as far as immunological balance is concerned, judging from the relatively high incidence of autoimmune aggressions on early sperm cells (see Voisin et al. 1974).

This is prompted by the appearance of differentiation-specific antigens behind the shelter of a so-called blood – testis barrier, a barrier that is not full-proof.

There is no prospect of man-made immune control of fertility during this period, although it would be very important to know how to prevent and cure autoimmunity.
The Local Reaction

The great majority of the contributions at this Symposium aim at mobile targets, the ovum and sperm separately, then the zygote, wandering up-and-down in the lumen of the female genital tract before embedding in the endometrium. Period III is seen as the most hopeful for a control by immunological means.

Little has been discovered about the immune response on the mucosal surface, although recently our knowledge on this subject has increased. The thorough reviews by Franklin (1974), Waldman & Ganguly (1974) and Vaereman & Férin (1974) make the point that the local response is not a mere reflection of the systemic immune status but consists of immunoglobulins synthesized by local plasma cells and secreted actively through the epithelium. There is also a local cell-bound immunity, probably due to a local stimulation of lymphocytes and macrophages. Facts in this area are still vague, for example, it is not established whether cellular immunity is endowed with real memory.

While these studies are in progress, two considerations can be suggested:

a) The correlation between serum antibodies and the efficiency of local immune responses cannot be taken for granted. This could account for the overall poor statistical significance of the correlation between the circulating antisperm antibodies and infertility reported by Jones (1974).

b) There is need for a better understanding of what regulates the reactivity – local and general – of the prospective mother towards the seminal antigens, in particular, the components of the spermatozoa which have been demonstrated to be antigenic by Goldberg (1974) and Jones (1974). There are obviously many factors involved in this kind of sensitization – dose, timing and repetition of contact are just some examined in the studies by Waldman & Ganguly (1974) and Vaereman & Férin (1974). Overall one tends to be surprised that the incidence and the clinical impact of sensitization is not more striking. Before hoping to activate an immunological control, it is urgent to find out whether tolerance, blocking or any suppression of reactivity normally takes place.

The ovum as a target

Shivers (1974) reports that heteroantibodies directed against extracts of ovaries and eggs react selectively with the zona pellucida. The zona is an interesting target because of the limited time of its existence around the ovum. These experiments yield information about the physiology of fertilization, but are less likely to lend themselves to applications in women. It would be out of the question either to try to autoimmunize or to passively transfer heteroantibodies. Passive immunization with antiserum raised in males of the same species (if the zona turns out to be iso-immunogenic) may be envisaged.
The sperm cell as the intruder

The male gamete is so highly specialized that many of its components are recognized as foreign by the female (and some by the male) immune system. The frequency of occurrence of circulating antibodies in women is accordingly high – whatever their significance to infertility – and they can react against different parts of the sperm cells, an observation recorded by Jones (1974). Edidin et al. (1974) have reviewed the evidence for mouse sperm cells possessing antigens controlled by minor H genes. Amos (1974) has reported that the product of the major histocompatibility locus (H-2 in mice, HL-A in man) is present, although in questionable amounts, on sperm and in the later stages of embryonic development.

Bennett et al. (1972) and Yamagisawa et al. (1974) have found antigens specified by alleles at the T locus. They are not present in other adult cells, and they may afford the basis for a distorted transmission in certain combinations; alternative ways to explain the distortion are as a function of T antigens in sperm-ovum recognition or as an affected viability of the sperm.

On the whole, allo-antigens (both histocompatibility and T) do not seem to be ideal tools in a planned aggression on sperm cells, for two reasons. Firstly, they would be useful only in certain combinations and immunization would then have to be carried out on a case by case basis, which is certainly inconvenient. Secondly, sensitization with H specificities would possibly become a barrier to transplantation therapy (e.g., kidney transplant) in later life, a limitation which has to be seriously considered.

Antigens present in all sperm cells and limited to them are a far more interesting subject for study. Goldberg (1974) has reported on a series of isoenzymes that are specific for the spermatozoa. Some of them have a known function in connection with the penetration of the sperm head into the ovum (hyaluronidase, acrosomal proteinase). There is still no definitive information on the effect of the antibody-enzyme interaction in vivo. If the inhibitory effects of the antibodies are confirmed, the models illustrated here by Ruth Arnon (1974) may be applicable, i.e., identification of the relevant antigenic determinants may lead to partial synthesis of the immunogen, instead of painstaking purification from material that may be hard to obtain in large quantities.

Another brilliant way to bypass the source problem is suggested by the experiments of Siddharta Sarkar of the Salk Institute (oral communication by Donato Cioli) who finds cross-reactions between bacterial glycoproteins and human gamete antigens. By far the best defined and most studied sperm-specific antigen is the LDH-X enzyme, about which Goldberg (1974) has reported. This seems to show great promise. Immunization of prospective mother rabbits with crystalline enzyme will cause a significant decrease in fertility.
However, several points should be elucidated before experiments in humans are started. Can the fertility rate be lowered further by applying the immunizing antigen locally, thus taking advantage of the mucosal immune response? What is the extent of cross-reaction between LDH-X from different species? Could one use a cross-reacting LDH-X to immune humans? Why should there be post-implantation death in blastocysts fertilized in an immune environment?

The last question raises a disturbing point: Goldberg (1974) also observed a harmful effect of anti-LDH-X on embryos, while LDH-X is limited to sperm cells. Does the antiserum contain antibodies directed or cross-reacting against other antigens, or does the embryo at early stages still contain LDH-X?

A recent study by Erickson & Coll (to be published) failed to repeat the damage on embryos. These authors used an antiserum raised in rabbits (injected with LDH-X from mice, purified by affinity chromatography) that had a far lower inhibition titer than that used in Goldberg's studies but was active in preventing fertilization in vitro.

CONCLUSIONS

We must consider that to improve on existing pharmacological and mechanical methods a new type of fertility control should meet a number of requirements:

(a) to be safe for the woman (low side effects)
(b) to be efficient (approaching full-proof)
(c) to be safe for the children in case of failure
(d) to be easy to apply and inexpensive
(e) to be reversible
(f) to be active on the fertilization rather than on the implantation step.

This would facilitate (without granting it!) moral acceptance by religious authorities.

It is clear from this meeting that an immunological method is not ready yet. But it is also clear from the many points which have been discussed that it may be feasible in a relatively short time. We may therefore guess which points will be easy and which will be more difficult to satisfy. a is fundamental and would certainly rule out any kind of autoimmunity, and probably any disruption of the delicate maternal-foetal relationship. Smaller obstacles seem to exist with the use of "local" reactions, although complete predictability still seems far away. The use of antigens specific to the male gamete would more easily satisfy b, c, d and probably f, whatever emphasis one wants to put on the latter. e is a problem with roots in the nature of immunology itself. In
theory, by creating local antigen excess, it might be possible to block an immune reaction also in a primed organism. However, it seems that any efficient method of fertility control based on an immune reaction, if achieved, will almost certainly constitute an irreversible decision.

REFERENCES

DISCUSSION

Nossal: Thank you for bringing to our attention these experiments of Billingham’s group. Could you enlighten me about your prediction with respect to animals which possess T cells but not B cells? If the effect of runting early after the skin graft transplantation is due to B cell function not yet having been built up, we should have a situation in which an animal possessing a competent T cell system but not the capacity to form antibody should never achieve multiparity. Of course, the experiment is not so easy because we don’t have “anti-nude” mice and because completely agamma-globulinaemic humans are usually males.

Celada: I think it would be fascinating to try to modify the mother-foetus relationship by changing the balance between humoral and cellular response (or B/T cells). One possible experiment in the mouse would consist in introducing insolubilized antigens (e.g., placental extract) into millipore chambers placed in the peritoneal cavity of pregnant females. While circulating antibodies would be absorbed, sensitized cells would not be able to penetrate the millipore membrane and would remain in circulation. According to the hypothesis we are discussing, under these circumstances a rejection of the “foetal grafts” may take place.

Nossal: Dr. Fudenberg might answer for the human.

Fudenberg: There have been a couple of patients reported, females with almost a complete absence of immunoglobulins; as you might expect, this is a very rare condition, presumably due to a double recessive gene; they have actually given birth to children. I know of one child, at least, but I do not know if they have given birth a second time.

Waldman: A couple of times during the Conference mention has been made about the short period of time for immunologic mechanisms to affect sperm, and you implied that you were pessimistic about the possibilities of immunologic intervention because of the short time period. Turning to another area, that is respiratory viral infections, certainly the time period is shorter than this time period and I think there is good evidence that antibody can act fast enough to interfere with the implantation, if you will, and the penetration of the virus into the cells. I am not sure that one should be pessimistic about the short time period.
Celada: I am not pessimistic: what I am saying is that the time factor may be a limitation. Also, antibody-mediated virus inhibition may have totally different kinetics than cell cytotoxicity directed against gametes wandering in the genital tract.

Jones: There have been two verbal reports (Materno-fetal Workshop, 2nd International Congress of Immunology, Brighton 1974), one in mice and the other in rats, although not the same rat strain used by Beer & Billington, which failed to confirm their results.

Mitchell: At a Brighton Workshop, several people reported that they have not been able to repeat this work. Now we know that Billington is very rarely wrong. What has accounted for the Beer & Billingham runting syndrome? Can Dr. Edidin talk about this?

Edidin: One wonders if there are not serious strain differences in the reactions of cells and placenta. This sort of thing has come up before. For example, Tuffry et al. (1969) claimed that there was easy cell passage across the mouse placenta in one strain and most other workers in the field could not find this with other strains (Billington et al. 1969). This makes one feel that it is important to make an exact repeat of the experiments with the same strains of animals.

Mitchell: Has no one really done the precise repeat of Beer & Billingham's experiment?

Edidin: I can recall no one who has done it with the precise strains and combinations.

Stevens: Dr. Southam mentioned earlier, and Dr. Celada mentioned again in his presentation the possibilities of using passive immunization to various hormones or other substances as a means of fertility control. We certainly know ways in which we can direct antibodies to certain substances that will disrupt reproduction. However, all immunologists I have talked with tell me that there is no safe way to immunize, on a repetitive basis, with animal sera or gammaglobulins. When one considers worldwide application to millions and millions, there is no practical way that one could obtain adequate numbers of human volunteers to contribute human globulin or serum to such a programme. So, I think that if someone could tell us how we could use animal antisera safely, there is a very real likelihood that we could use such an approach.

Amos: One of the states of Nature that has not been discussed is the one child sterility. I don't know whether this is because it is no longer a point of controversy, but it would seem to me that possible immunological involvement in this condition should be discussed. I would like to register concern about the advocacy of grandchildless and think that the possible consequences of this are difficult to foresee. Alternatively, I wonder whether the possibility of encouraging, by immunological methods, single child families could be explored. On another point, the possibility of immunoselection, I have been very interested during the Workshop in the possible exclusion by the mother of genetically deformed foetuses. The high rate of something like 12% of pregnancies terminating in abortion has to be borne in mind. If genetically abnormal foetuses are rejected, can this process be exploited to include other conditions? For example, could the wife of a man carrying the HL-A antigen W27 who develops ankylosing spondylitis (or one of those few other conditions in which there is a very strong association between a given HL-A type and a disease) be stimulated to abort W27-carrying foetuses? In another example, we are studying multiple polyposis in one large family with something like 50% of the children being affected. Assuming linkage with HL-A,
I would have thought that the families would certainly have welcomed some form of immunogenetic counselling if this was available.

Edidin: I am grateful to Dr. Celada for raising the point of placentae elsewhere in Nature than in mammals. One could say, really, you should not take these other examples seriously, they are clear examples of convergence, of the same mechanism evolving independently multiple times. And yet, why wasn't that apparently more efficient way to raise a foetus fixed in evolution, why do you see only scattered groups of invertebrates with placentae of some sort? Your suggestion that perhaps one needs further immune response to have the placenta as an efficiently working, interacting, organ in pregnancy is fascinating.

Another comment is closer both to the topic of the Meeting and to our own work. I was very pleased to see the inclusion of Dr. Parmiani's data on immunization with tumours and an apparent effect of decrease in pregnancy incidence. Clearly there are grave ethical problems implied by extending this sort of model. I nevertheless would suggest that it may be worth exploring, if our colleagues who can define peptides and synthesize them are prepared to come to the aid of those who work with tumour derived materials in the initial stages of immunization and synthesize immunogenic portions of tumour antigens.

Nossal: On that point, Dr. Celada, before you answer it, would you define a bit more what you meant by pregnancy? Was it viable offspring?

Celada: Parmiani's data are expressed as per cent successful matings. Whenever a mating was successful, there was no significant difference in litter size between immunized and control mothers.

I agree wholeheartedly with Dr. Edidin's invitation not to overlook the possible connections with tumour immunology (and also, may I add, with the immunology of parasites).

Another point I wish to underline is the deposition of immune complexes in the organs of pregnant animals described by Kenneth Tung. It seems interesting to me that this phenomenon exists, on a smaller scale, also in females that are pregnant as a result of syngeneic mating. The antigens involved here may be related to those studied by Parmiani.

References: