Zusammenfassung. Röntgenbestrahlung von Embryonen und Keimzellen bestimmter Zahnkarpfen-Genotypen, deren Makromelanophorenproduktion durch Kontrollgene auf die Bildung kleiner Flecken begrenzt ist, verursacht bei heranwachsenden Tieren und deren Nachkommen Prämelanome. Demgegenüber hat die gleiche Behandlung dieser Entwicklungsstadien bei Genotypen, deren Makromelanophorenproduktion infolge Fehlens der Kontrollgene im Verlaufe des weiteren Lebens zur Melanombildung führt, keinen Effekt. Bestrahlung der melanomtragenden Adulten dieses Genotyps verursacht eine vorübergehende Unterbrechung des Tumorwachstums.

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Delay in Skin Allograft Rejection in Rats Grafted with Fetal Adrenal Glands

The adrenal gland is large in the mammalian fetus, decreasing later at a different rate according to the species. In mice[1] and rats[2] this involution is considerably retarded with respect to other species and the immunological system of these animals is not completely developed at birth[3–5]. In men[6] and guinea-pigs[7] the involution of the fetal adrenal gland takes place early in gestation and this is correlated with a better development of immunological functions at birth[8–10]. These facts, added to the well known functional antagonism between adrenal gland and thymus and the immunosuppressive action of adrenal glucocorticosteroids, suggest that the fetal adrenal gland might be involved in the control of the development of the immunological system. With the aim of exploring this possibility, the following experiments were performed.

Material and methods. 125 inbred male albino rats were grafted on the 10th day after birth with black skin allografts obtained from inbred male hooded rats. The hosts were treated as follows: In 22 animals, fetal adrenal glands were implanted under the skin on the 6th and 8th day after birth. The number of adrenal glands implanted was 10 in 18 cases and 5 in 4 cases. The adrenal glands were obtained from last week of gestation isogeneic fetuses. In 26 rats isogeneic adult adrenal glands were implanted under the skin on the 6th and 8th day after birth (2–4 glands per animal). 33 animals were daily injected with different doses of a suspension of 16β-methyl prednisone (from 10 to 40 µg) from the 6th day after birth until the rejection of the graft. 44 rats received the skin allograft only.

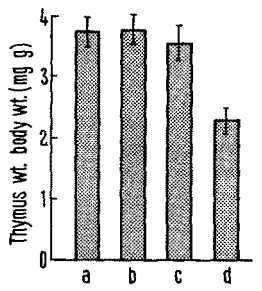
Evaluation of the success or failure of the skin grafts was based on gross inspection. Rejection of grafts was characterized by an initial take followed by inflammation, induration, hardening of the graft, and ultimate complete slough. Prolonged survival of the graft was considered to be present when the graft showed a delayed onset of the early evidence of rejection.

Histological study was performed in order to determine the fate of the grafted adrenal glands and the effect of the different treatments over the thymus. The material was composed of 7 rats implanted with 10 fetal adrenals on the 6th day of life. 7 rats daily injected by s.c. route with 30 µg of 16β-methyl prednisone. 8 rats implanted with 2 adult adrenals on the 6th day of life. 5 normal rats.

The animals were sacrificed on the 16th day of life. In the animals in which adrenals were grafted, a wide zone of skin and subcutaneous tissue corresponding to the site of the adrenal grafts and the thymus in all animals were fixed in 10% Formol, embedded in paraffin, serial sectioned at 6 µm thick and stained with Hema-

Treatment	Skin allograft survival (days) No. of animals	Less than 15	15 to 50	More than 50
Skin allograft only	44	44	–	–
Implanted with 5 fetal adrenal glands	4	3	1	–
Implanted with 10 fetal adrenal glands	18	10	1	7
Implanted with 2–4 adult adrenal glands	26	25	–	1
Daily injected with 10–25 µg of 16 MP	21	23	–	–
Daily injected with 30 µg of 16 MP	5	4	–	1
Daily injected with 40 µg of 16 MP	7	3	2[a]	2

16 MP, 16β-methyl prednisone. [a] Death before day 50th.



Thymus relative weight (thymus wt./body wt., mean S.E.) in: a) controls; b) animals grafted with 10 fetal adrenal glands; c) animals grafted with 2 adult adrenal glands; d) animals injected daily from the 6th day after birth with 30 µg of 16β-methyl prednisone.

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toxilin Eosin. Body and thymus weight were obtained in every animal.

Results. The result of skin grafts survival are summarized in the Table. A delay in skin allograft rejection with respect to a group of 44 untreated controls was only obtained in the following conditions: 1 rat out of 4 implanted with 5 fetal adrenal glands (rejection time: 38 days). 8 rats out of 18 implanted with 10 fetal adrenal glands (mean rejection time 67.1 ± S.E. 3.2 days). 1 rat out of 26 implanted with 2–4 adult adrenal glands (rejection time: 58 days). 1 rat out of 5 daily injected with 30 μg of 16β-methyl prednisone (rejection time 63 days). 2 rats out of 7 daily injected with 40 μg of 16β-methyl prednisone (rejection time: 57–69 days, other 2 died during treatment).

The rejection time was not affected by lower doses of 16β-methyl prednisone. The animals treated with the higher doses of 16β-methyl prednisone were very wasted and showed growth impairment.

In the histological studies of the fate of the implanted adrenal glands it was possible to recognize only 2 glands per area examined in 3 animals out of 7 that had received 10 fetal adrenal glands. The possibility that the glands could have slide under the skin away from the site of implantation cannot be excluded. It was not possible to recognize any viable gland in 8 animals that had received 2 adult glands each.

The thymus of rats grafted with fetal or adult adrenal glands did not show histological changes. In the rats treated with 30 μg of 16β-methyl prednisone it was found a great limphoid depopulation and numerous images of picnotic nuclei. There were no differences in thymus relative weight (thymus wt./body wt.) between controls and rats grafted with fetal or adult adrenal glands. On the contrary, the thymus weight of the animals treated with 30 μg of 16β-methyl prednisone, showed a significant diminution (Student's t-test, $p < 0.001$; Figure).

Discussion. From these data it could be concluded that the implantation of fetal adrenal glands can delay the rejection of skin allografts in the rat, and that the corticosteroid doses necessary to obtain an effect similar to that determined by the fetal adrenal glands are high. The animals treated with these doses were very wasted (2 animals given 40 μg of 16β-methyl prednisone died during treatment). It was observed that the administration of the corticosteroids induced a marked reduction of thymus weight, while in the animals grafted with fetal adrenals there was not such reduction. If the thymolitic effect is taken as an indirect estimation of the glucocorticosteroids level in blood, these facts suggest that the glucocorticosteroids might not be responsible for the delay in the immunological rejection induced by fetal adrenal glands. Furthermore it is rather unlikely that in hosts receiving a gland, which have their own adrenal glands, the corticosteroid level would rise so much, being submitted to regulation by a feed-back mechanism[11].

The ability of fetal adrenal gland to delay the rejection of skin grafts might be related to its histological appearance, different from that of adult adrenals[12,13].

Resumen. Se ha observado en este trabajo que el injerto de suprarrenales de fetos en ratas de 6 días de edad determina aumento del tiempo de supervivencia de aloinjertos de piel realizados al 10° día de vida en relación a los testigos y a ratas injertadas con suprarrenales de adulto. Un efecto similar se observó cuando se administró diariamente a partir del 6° día de vida 16β-metil prednisona en dosis altas que producían gran trastorno del crecimiento, evidente disminución de peso del timo y en algunos casos muerte del animal.

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Étude de la compétition antigénique chez des souris thymectomisées à la naissance

Depuis une dizaine d'années, le thymus s'est révélé jouer un rôle important dans différents phénomènes immunitaires: hypersensibilité retardée, immunité humorale vis-à-vis de certains antigènes, tolérance immunitaire. Nous avons recherché, dans ce travail, l'intervention éventuelle de cet organe dans la compétition antigénique chez la souris. Nous avons choisi comme modèle de cette compétition, celle qui apparait lorsque des globules rouges de mouton (GRM) sont injectés quelques jours après une immunisation par des globules rouges de cheval (GRC). Ces derniers étant pratiquement sans relation antigénique avec les GRM[1].

Matériel et technique. Dans un premier temps nous avons déterminé quel était l'intervalle de temps entre l'injection du premier antigène (GRC) et du deuxième antigène (GRM) qui permettait d'obtenir l'inhibition maximum de la réponse primaire vis-à-vis du deuxième antigène. Dans un deuxième temps nous avons comparé l'importance de cette compétition antigénique chez des souris faussement thymectomisées ou thymectomisées à la naissance.

Dans la première série d'expériences nous avons utilisé 30 souris CF 1 âgées de 1 à 2 mois, réparties en 6 groupes de 5 animaux que nous avons immunisés par voie i.p. Un groupe témoin a reçu uniquement 10^8 GRM, les 5 autres lots ont été injectés d'abord avec 10^9 GRC puis avec 10^8 GRM 2, 3, 4, 5 ou 8 jours après, suivant les groupes. Ces animaux ont été sacrifiés 4 jours après l'injection des GRM et le nombre de cellules formatrices d'anticorps anti GRM de type IgM dans la rate a été déterminé par une technique des plages d'hémolyse dérivée[2] de celle de CUNNINGHAM et SZENBERG[3].

Résultats. Le Tableau I indique les résultats que nous avons obtenus. Il permet de constater comme TALMAGE[1] l'avait signalé que l'inhibition de la réponse primaire aux GRM est maximum lorsque 4 jours séparent l'injec-

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