Experimental Studies on the Autotransplantation of Thyroid Gland using Micro-vascular Anastomoses

by

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INTRODUCTION

Formidable efforts have been made in order to solve the problems related to organ transplantation, and the recent advances in this field enabled us to obtain successful clinical cases now and then. One of the first considerations in the transplantation of tissues and organs is the reconstruction of the blood supply by vascular anastomosis. Implants, or tissue fragments placed into another tissue, such as muscles, undergo progressive degeneration while the development of penetrating capillaries from the surrounding structures may occur, and their central parts are subjected to necrosis, although a process of regeneration from the surviving cells would follow. The immediate vascularization by vessel anastomosis, however, would offer the most prompt opportunity for the grafts to display their maximal function. It should also be taken into consideration that nervous connection and proper lymphatic vascularization, besides the blood supply, may be of functional importance for the surviving cells. The function of the transplants, however, transplanted by intact blood supply, might be modified by the operative procedures of denervation, severance of lymphatics and changes in hemodynamics, at least for some time or maybe for a long period. The relative simplicity of the function of the thyroid gland makes it easy to evaluate the functional activity of the transplants. Numerous studies on thyroid grafting, experimental and clinical, have been reported[4)61)]. Many of them were chiefly on morphological studies, and the physiological data concerning the graft function are limited. Recently, the development of a radioactive tracer technic gave a fresh force to the investigation of the thyroid function by offering a new experimental approach. In this presentation, the thyroid glands were transplanted into the groin using the microvascular anastomoses. Autologous gland was utilized to avoid the complication of immunologic incompatibility which would have been present if homologous tissue had been used instead. The author investigated the function of the thyroid transplants with special reference to its metabolism of radioactive iodine. Humoral regulation of the thyroid transplants was also studied, because it is important for the thyroid tissue to be well regulated by the anterior pituitary in maintaining its proper function. Histological study was also done.

MATERIALS AND METHODS

1. Materials

One hundred mongrel dogs, weighing from 6 to 22 kg, were used. The animals were anesthetized with intravenous administration of sodium pentobarbital, 25 mg/kg.

The anatomic relationship of the canine thyroid. The gross anatomy of the thyroid gland in the dog is different from that in man.[38] The thyroid gland consists of two separate lobes lying laterally to the trachea, and the isthmus cannot be found. The principal blood supply is provided by a cranial thyroid artery which arises from common carotid artery. The caudal thyroid artery is a small vessel, if present. The venous return passes through cranial and caudal thyroid veins, which join the internal jugular vein. Lymphatics from the thyroid go to the mandibular, retropharyngeal and paratracheal lymph nodes. From there the lymph proceeds to the cervical lymphatic trunk and enters into the venous circulation at the junction of the external and internal jugular veins. The thyroid receives nerve fibers derived from the cranial cervical ganglion of the sympathetic, and from the cranial laryngeal branch of the vagus, arising from the nodosal ganglion or vagosympathetic trunk. The two components of the thyroid nerve usually merge, and pass into the thyroid gland along the course of the cranial thyroid artery and its branches. Fibers from the caudal cervical ganglia may reach the thyroid attending the vessels, but no direct branches from these ganglia are found. As for the distribution of the intrinsic nerves, some consider that the nerves terminate upon the vessels, while others believe that follicles are also innervated[10][26][45].

2. Operative procedures

a. Transplantation into the groin. The thyroid was freed from the surrounding structures, with the blood vessels being left intact. The common carotid artery was dissected at its origin. The internal jugular vein was removed with a patch of the wall of the brachiocephalic vein surrounding the entrance of the internal jugular vein. The left lobe was left intact, though in several cases it was extirpated. Following the preparation of this graft, the right groin was incised. The femoral vessels were exposed, divided and their distal ends were ligated. Not until this stage was the graft removed from the neck.

Vascular anastomosis. The cut end of the common carotid artery which was attached to the graft, was anastomosed to the proximal cut end of the femoral artery, using the INOKUCHI's blood vessel suturing apparatus (Senko Medical Instrument Mfg. Co., Tokyo). The patch of the wall of the brachiocephalic vein, in the center of which the entrance of the internal jugular vein located, was then anastomosed to the proximal cut end of the femoral vein, using the same suturing apparatus. In several cases, the internal jugular vein was directly anastomosed to the proximal cut end of the femoral vein or to the branch of the femoral vein in end-to-end fashion. Physiologic saline solution, with the addition of 0.02% heparin, was used locally to wash out the air in the lumen of the vessels. In one instance, the segment of external jugular vein was interposed between both cut ends of the femoral vein, because the thyroid vein entered into the external jugular vein in this exceptive case.

b. Orthotopic transplantation in the neck. The right lobe of the thyroid was freed from the surrounding structures preserving the continuity with the internal jugular vein.

The common carotid artery was divided at 5 cm caudal to the origin of cranial thyroid artery and re-anastomosed immediately. The distal portion of the common carotid artery was cut and ligated.

c. The denervation of thyroid gland. The cranial cervical ganglion of the sympathetic and the nodosal ganglion of the vagus were resected together with the vagosympathetic trunk unilaterally in the right neck.

d. The ligation of the lymphatics of thyroid gland. Evans-blue (0.15 ml of 0.5 % solution) was injected beneath the thyroid capsule. A few minutes later the lymph vessels, in which the lymph was stained blue, were ligated, and the lymph nodes which were stained blue (mandibular and paratracheal lymph nodes) were resected.

e. Mock operation. An incision was carried out in the right neck, the muscles were separated, the carotid sheath was exposed and the wound was closed.

3. Radioactive tracer study of the thyroid transplants

a. Injection of ^{131}I. A 50 microcurie dose of carrier-free ^{131}I, as sodium iodide, was injected intravenously. Prior to the administration, the animals were fed with bread and cow's milk or boiled rice and vegetables and white fish for 3 days. The tracer dose was drawn into the syringe, and counted with the directional gamma-ray scintillation counter at a distance of 90 cm. The natural background counts were also measured. The net counts per minute at a distance of 90 cm should be corrected, by means of standard gamma-ray source of mock iodine capsule, to the cpm which would be obtained when it was counted at a distance of 30 cm. The net tracer dose administered to the animal was then known in cpm.

b. Measurement of uptake of ^{131}I by thyroid gland. Forty-eight hours later, the animal was sacrificed and the thyroid glands were extirpated, and ^{131}I uptake was determined at a distance of 30 cm. The net thyroid counts, when compared with the corrected net dose counts, gave the percent uptake by the thyroid gland, allowing for the radioactive decay.

c. Chromatographic analysis of ^{131}I compounds in the thyroid.

Preparation of thyroid homogenate and the enzymatic hydrolysis of it. The thyroid gland was weighed, and a small piece of tissue was removed for histology. The thyroid gland was minced with scissors and homogenized in a glass homogenizer with 0.5 ml of physiologic saline solution. The homogenate, added to 2.5 ml of physiologic saline solution, was centrifuged at 9,500 r.p.m. (radius 10 cm, centrifugal force $10{,}000 \times g$) for 10 minutes at 0°C. The pH of the supernatant fluid was adjusted to 8.6 with dilute 0.2 N NaOH, and the thyroglobulin in the fluid was hydrolyzed with pancreatin at 37°C overnight.

Chromatographic procedures. Chromatograms were made on paper strips (47×5 cm, Toyo No. 50) with a solvent consisting of the supernatant fluid from a 17 : 2 : 15 mixture of butanol-acetic acid-water, running in a descending fashion in an airtight box. A beaker containing butanol saturated water was placed in the bottom of the box.

Location of ^{131}I compounds on the chromatograms by radioautography. The resulting chromatograms were dried and placed in an X-ray film holder with a sheet of X-ray film. The film was developed after a suitable interval, usually 2 days later. For the quantitative estimation of the ^{131}I of the compounds on the chromatograms, the papers were divided corresponding to the bands observed on the radioautogram, and each section

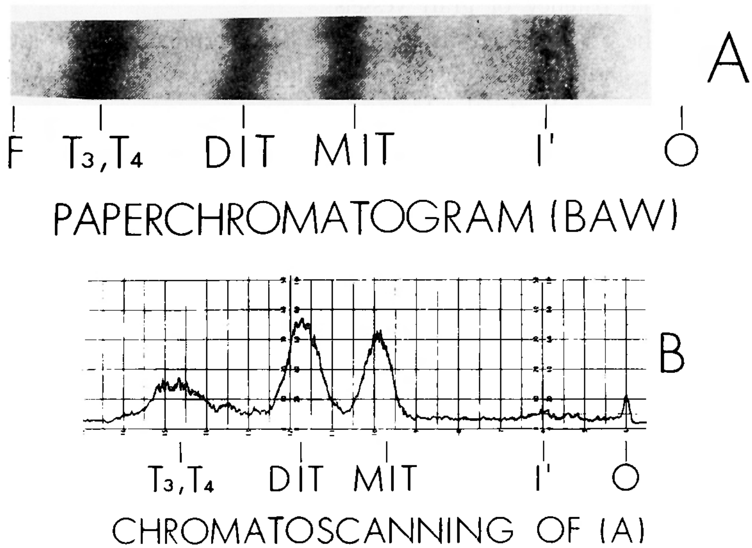

Fig. 1 Top (A), A paperchromatogram stained, demonstrating the four major iodine compounds in the thyroid gland. Bottom (B), Demonstrating the location of radioactivity on (A).
O : Origin component, I′ : Inorganic iodide, MIT : Monoiodotyrosine, DIT : Diiodotyrosine, T_3: Triiodothyronine, T_4: Thyroxine, F : Frontal line of solvent.

was counted in the well-type scintillation counter. All analyses were made in duplication.

4. PBI-TSH test

Prior to the administration of thyroid stimulating hormone (TSH) the left lobe of the thyroid had been extirpated, and a fasting blood sample was taken for basal protein-bound iodine (PBI) determination. TSH (Thytropar, Armour Pharm. Co.) was supplied by Tokyo Tanabe Pharm. Co. This was dissolved in a diluent immediately before use, and a dose of 5 international units of Thytropar (TSH 100 mg) was administered intramuscularly. Blood samples were withdrawn at 3 to 72 hours later.

5. Studies on hemodynamics

a. Measurement of thyroidal blood flow. The thyroid was freed from the surrounding structures, with its vessels being left intact. A polyethylene catheter was introduced into the internal jugular vein and outflow venous blood was collected directly for 5 minutes. The common carotid artery was then occluded at 2 cm cranial to the cranial thyroid artery and the thyroidal blood flow was measured again. These procedures were repeated 3 times and a half. Sodium heparin of 10,000 units was injected, and 10 % dextran solution was infused at a constant rate during the measurement.

b. Measurement of venous pressure in the recipient vein. A saline filled polyethylene catheter was introduced into the femoral vein via its branch, and femoral venous pressure was measured under the following conditions : 1) when the venous return from the leg was kept intact, 2) when the distal portion of the femoral vein was occluded and 3) when the occlusion was released.

6. Ascertainment of the patency of graft vessels

The patency of the anastomosed artery could be ascertained by the pulsation of the grafted carotid artery, and it was confirmed by the angiograms which were made with 76% urografin slowly injected into the femoral artery.

7. Histology

Tissues obtained at autopsy were fixed in 10 % formol, and paraffin sections were prepared and stained with hematoxylin and eosin, or azan stains in the usual manner.

RESULTS

1. Results of micro-vascular surgery

Table 1 summarizes the results of the thyroid transplantation using vascular anastomoses. When the internal jugular vein was directly anastomosed to the recipient vein, most cases were unsuccessful. It succeeded in only two cases, in which the outside diameter of the internal jugular vein was as large as 2.5 mm exceptionally. The arterial anastomosis was performed with a relative ease, because the diameter of the arteries, both carotid and femoral, were larger than 3.0 mm, and bushing size of at least 2.0 mm was available. The period of graft ischemia ranged from 20 to 50 minutes in the transplantation into the groin, and it was within 5 minutes in the transplantation in the neck.

Table 1. Results of thyroid autotransplantation using micro-vascular anastomoses.

Dog No.	Sex	Site of transplant	Size of bushing of suturing apparatus (mm)		Period of follow-up (days)	Result
			artery	vein		
1	M	Groin	3.0	1.5*	23	Unsuccessful
4 (†)	M	Groin	3.0	1.5*	4	Died
5 (†)	M	Groin	3.0	1.5*	2	Died
6 (†)	M	Groin	2.5	3.0	1	Died
7	F	Groin	3.0	3.5	5	Satisfactory
8	F	Groin	3.0	4.0	1	Died
11	M	Groin	3.0	2.5*	30	Satisfactory
12	M	Groin	3.0	4.0	42	Unsuccessful
14	M	Groin	3.0	3.5 & 4.0**	11	Unsuccessful
17	M	Groin	3.0	nylon suture* #	12	Unsuccessful
18	M	Groin	3.0	3.5	13	Satisfactory
19	M	Groin	3.0	4.0	2	Satisfactory
20	M	Groin	3.0	2.0*	60	Satisfactory
21	M	Groin	3.5	4.0	60	Satisfactory
22	M	Groin	3.5	4.0	60	Satisfactory
23	M	Groin	3.5	4.0	60	Unsuccessful
39	F	Groin	2.5	1.5* #	3	Unsuccessful
40	F	Groin	3.0	4.0	3	Died
41	F	Groin	2.5	4.0	9	Satisfactory
43	M	Groin	2.5	3.5	7	Satisfactory
44	M	Groin	3.0	3.5	0	Died
45	M	Groin	2.0	3.0	3	Satisfactory

46	F	Groin	2.5	3.0	5	Satisfactory
47	F	Groin	2.5	3.5	13	Satisfactory
48	F	Groin	3.0	3.5	20	Satisfactory
49	F	Groin	2.0	2.5	13	Satisfactory
50	M	Groin	3.0	3.5	3	Satisfactory
51	F	Groin	2.0	3.5	7	Satisfactory
54	M	Groin	2.0	3.5	11	Satisfactory
55	M	Groin	2.5	3.5	11	Satisfactory
56	M	Groin	2.5	4.0	30	Unsuccessful
61	M	Groin	2.0	3.0	15	Unsuccessful
62	F	Groin	3.0	4.0	13	Satisfactory
63	M	Groin	3.0	4.0	1	Died
65	M	Groin	2.0	3.0	1	Died
66 (†)	M	Groin	2.5	3.5	8	Satisfactory
70	F	Neck	2.5	***	5	Satisfactory
71	M	Neck	3.0	***	20	Satisfactory
72	M	Neck	2.5	***	5	Satisfactory
73	F	Neck	2.5	***	5	Satisfactory
74	M	Groin	2.5	4.0	15	Satisfactory
75 (†)	F	Groin	3.0	4.0	99	Satisfactory
76	F	Neck	2.5	***	20	Satisfactory
78	M	Groin	3.0	4.0	30	Satisfactory
79	F	Groin	2.5	4.0	4	Died
80 (†)	F	Groin	3.0	4.0	26	Satisfactory
81	F	Neck	2.5	***	9	Unsuccessful
82	M	Neck	2.5	***	14	Satisfactory
90 (†)	M	Groin	2.5	3.5	17	Died

(†) The other lobe was extirpated.
* Int. jug. v. was anastomosed to the proximal cut end of fem. v. in end-to-end fashion. (In other cases, the patch of the wall of brachiocephalic v. was used, except for dog No. 14)
** The segment of ext. jug. v. was interposed between both cut ends of fem. v., because the thyroid v. entered into the ext. jug. v.
*** The continuity of the graft with int. jug. v. was preserved.
# The branch of fem. v. was used as a recipient vein in these two cases.

2. Thyroidal blood flow rate

The amount of thyroidal venous return was 0.720 ml/min. (mean value from four measurements) when the carotid circulation to the head was kept intact. It rose to 0.804 ml/min. (from three measurements) when the distal portion of the carotid artery was occluded. The increasing rate was 11% (Table 2).

Table 2 Effect of the occlusion of common carotid artery on the thyroidal blood flow.
(Dog No. 100, 13kg ♂)

	Common carotid artery open	Cranial portion of common carotid artery occluded
Flow rate* (ml/min.)	0.720	0.804

* by means of direct collection of thyroidal outflow venous blood.

3. Venous pressure in the recipient vein

The femoral venous pressure was 58 mmH_2O when the venous return from the leg was kept intact. It reduced to 37 mmH_2O during the occlusion of the distal portion of femoral vein, and returned again to 57 mmH_2O following its release (Table 3).

Table 3 Effect of occlusion of femoral vein on the venous pressure in recipient vein.
(Dog No. 58, 9kg ♀)

	Femoral vein open	Distal portion of femoral vein occluded	Occlusion released
Venous pressure (mmH_2O)	58	39	57

4. Uptake of ^{131}I by the thyroid gland

The ^{131}I uptake of a thyroid lobe ranged from 3,000 to 12,000 cpm above background which was 1,000 cpm. Table 4 summarizes the percent uptake in normal dogs. The uptake of a right lobe was a little higher than that of a left lobe of the respective dog in most cases. Table 5 summarises the ^{131}I uptake in the transplanted dogs. The transplanted lobe picked up the radioiodine as early as on the 3rd postoperative day. Fig. 2 shows that it reached to the normal level on the 13th day, followed by a slight decrease on the 60th day. The ^{131}I uptake of the intact lobe was also suppressed to some extent in early postoperative days. The uptake of the transplanted lobe, when compared with that of the intact lobe of the respective dog, showed a relatively reduced value (Fig. 3). The ratio of transplanted/intact of ^{131}I uptake ranged from 0.176 to 1.05. Table 6 shows the ^{131}I uptake of other groups, and Fig. 4 shows the ratio of operated/intact of ^{131}I uptake of these groups.

Table 4 Amounts of ^{131}I uptake by the thyroid gland of normal dogs.

Dog No.	Sex	Body weight (kg)	Weight of thyroid gland (mg)		Time after injection of ^{131}I (hrs.)	Thyroidal ^{131}I uptake (% dose)		
			r.	l.		r.	l.	mean
24	M	10	326	249	24	6.8	6.2	7.6
28	F	12	271	272	24	7.0	6.8	
31	F	6	318	330	24	9.5	9.6	
25	F	8	208	203	48	13.9	13.5	10.4
29	M	9	833	680	48	9.8	9.0	
32	M	9	235	221	48	8.4	7.8	
26	M	12	512	404	72	15.7	12.8	14.25
30	F	9	597	486	96	16.8	14.5	16.4
33	M	9	250	268	96	16.2	18.0	

Table 5 Amounts of ^{131}I uptake by thyroid gland of transplanted dogs. (Forty-eight hours after injection of ^{131}I)

Site of transplant	Dog No.	Time after transplantation (days)	Weight of thyroid gland (mg) r. #	Weight of thyroid gland (mg) l.	Thyroidal ^{131}I uptake (% dose) r. #	Thyroidal ^{131}I uptake (% dose) l.
Groin	45	3	329	295	2. 2	2. 1
	50	3	1457	1182	5. 2	7. 0
	7	5	—	—	5. 8	8. 8
	46	5	374	342	3. 2	3. 9
	43	7	457	389	6. 7	7. 1
	51	7	429	281	5. 4	5. 3
	66	8	474	***	4. 8	***
	41	9	—	—	3. 8	5. 2
	54	11	547	623	3. 6	9. 5
	55	11	434	401	4. 9	6. 0
	18	13	1470	537	9. 7	12. 1
	47	13	473	426	9. 7	11. 4
	49	13	418	315	13. 3	17. 8
	62	13	467	385	1. 7*	3. 5
	61	15	374	387	1. 0**	22. 0
	74	15	565	510	9. 5	13. 3
	48	20	477	453	15. 2	17. 7
	1	23	302	—	0. 7	9. 5
	11	30	804	573	8. 3	8. 5
	56	30	—	394	0. 0	28. 4
	78	30	783	566	1. 6	9. 1
	20	60	898	802	6. 1	14. 9
	21	60	798	519	9. 6	10. 2
	22	60	839	910	8. 5	14. 6
	23	60	—	703	0. 0	9. 5
Neck	70	5	666	573	3. 7	5. 4
	72	5	555	429	6. 7	9. 4
	73	5	480	394	10. 2	21. 9
	81	9	1659	359	0. 0	9. 5
	82	14	501	672	2. 2	2. 7
	71	20	381	371	7. 4	9. 7
	76	20	462	468	3. 2	5. 1

In all cases, right lobe was transplanted.
* Stored at 4℃ for 24 hours.
** A few follicles containing the colloid were seen in histology.
*** Extirpated.

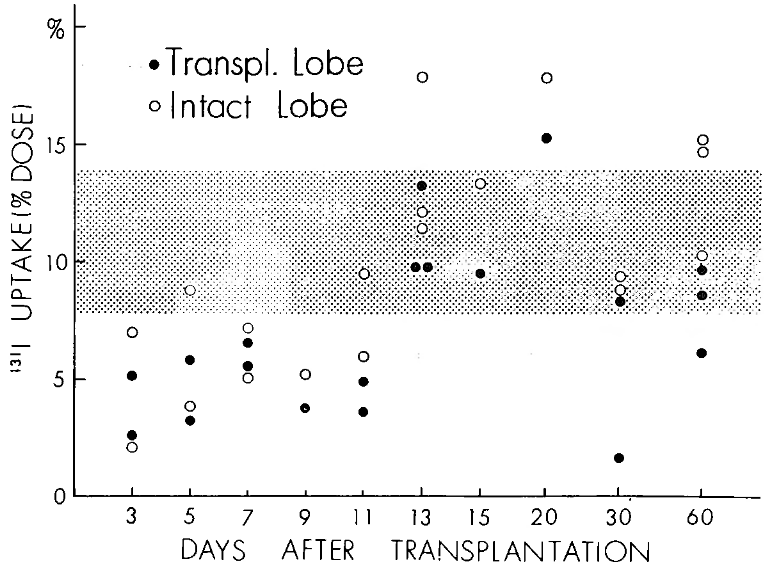

Fig. 2 Percent uptake of ^{131}I by thyroid gland in the transplantation into the groin. Dotted area shows the range of ^{131}I uptake by a thyroid lobe in the normal dogs 48 hours after ^{131}I injection.

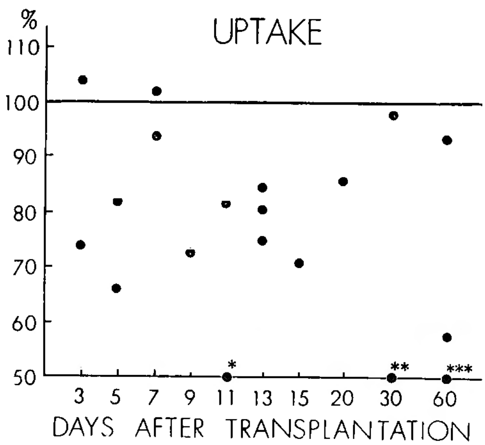

Fig. 3 Comparison of ^{131}I uptake by both thyroid lobes in the transplantation into the groin (transplanted/intact ratio). * : 38%, ** : 17.6%, *** : 41%

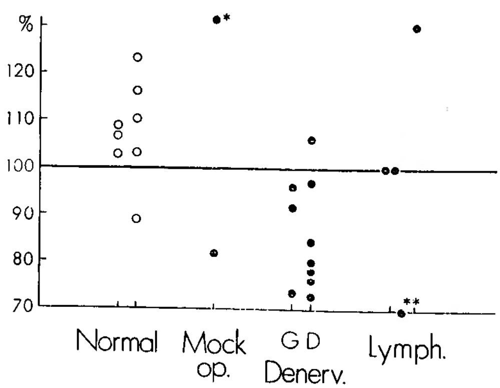

Fig. 4 Comparison of ^{131}I uptake by both thyroid lobes in 8 normal dogs (right/left ratio), 2 dogs subjected to mock operation, 3 dogs (G) with complete denervation of the thyroid, 7 dogs (D) with partial denervation of the thyroid, and 4 dogs with lymphatic ligation of the thyroid (operated/intact ratio). The operation was performed unilaterally on the right side of the neck. * : 144%, ** : 57.5%

Table 6 Amounts of ^{131}I uptake by thyroid gland. (Forty-eight hours after injection of ^{131}I)

Group	Dog No.	Time after operation (days)	Weight of thyroid gland (mg)		Thyroidal ^{131}I uptake (% dose)	
			r. #	l.	r. #	l.
L	83	5	508	448	5.3	5.3
	88	5	404	320	3.3	3.3
	91	14	472	381	2.7	4.7
	92	22	729	601	3.9	3.0
M	84	5	392	437	4.5	5.4
	87	5	418	339	15.4	10.7
G	93	5	437	464	7.4	8.1
	95	5	313	307	8.7	11.8
	96	5	471	468	9.2	9.6
D	85	5	238	268	3.6	4.9
	86	5	418	550	7.0	9.1
	9	28	256	216	12.9	16.1
	10	47	332	402	9.3	11.0
	27	62	642	778	9.1	11.7
	35	68	300	273	24.2	22.7
	36	68	364	387	13.4	13.8

: In all cases, the operation was done on the right side of the neck.
L : Lymphatics of the thyroid were ligated.
M : Mock operation.
G : Thyroid was denervated by means of resection of cranial cervical ganglion and nodosal ganglion.
D : Common carotid artery was divided at about 1cm cranial to cranial thyroid artery. And, in addition, the adventitia of c.c.a. was resected or c.c.a. was cut and re-anastomosed, at about 5 cm caudal to cranial thyroid artery.

5. Chromatographic results

A typical chromatogram is presented in Fig. 1. The four major iodine compounds in the thyroid are seen very clearly. The radioactive counts of each compound ranged from 1,000 to 10,000 per minute. Tables 7, 8 and 9 summarize the results of chromatographic analysis of the thyroid gland of normal dogs, transplanted dogs and other groups, respectively, expressed as % of total ^{131}I taken into the gland. The ^{131}I remaining at the origin of the chromatogram was excluded from the present analysis, as it was considered to be attributed to the unhydrolyzed ^{131}I-thyroglobulin. The content of inorganic iodide (I′) amounted to 15% or thereabout. Monoiodotyrosine (MIT) represented about 30 to 35% and diiodotyrosine (DIT), about 35 to 40%. The content of triiodothyronine (T_3) & thyroxine (T_4) comprised 10 to 20% of total iodine (Fig. 5). When these contents of both lobes were compared with each other, the following tendency could be noted, which was shown in Fig. 6 : 1) The content of I′ was almost always lower in the transplanted lobe, 2) the contents of MIT and DIT were higher in the transplanted lobe, and 3) the content of T_3 & T_4 of the transplanted lobe was lower in general, but it might become higher than that of the intact lobe during a period lasting from the 9th to the 13th day.

Table 7 Summary of chromatographic analysis of thyroid gland pancreatin digest in normal dogs. (% of total ^{131}I in the gland*)

Dog No.	Time after ^{131}I injection (hrs.)	right lobe				left lobe			
		I'	MIT	DIT	T_3, T_4	I'	MIT	DIT	T_3, T_4
24	24	7.3	42.4	41.4	9.1	8.8	42.3	40.3	8.7
28	24	7.1	13.2	64.9	14.8	7.0	13.1	69.1	10.8
25	48	10.0	34.5	40.4	15.1	13.2	33.9	42.1	10.8
29	48	17.1	28.6	40.9	13.4	19.8	28.2	35.3	16.7
32	48	10.0	24.3	39.3	26.4	11.5	28.0	40.1	20.4
26	72	9.1	33.1	36.6	21.2	9.3	30.5	38.6	21.6
30	96	14.1	26.7	42.9	16.3	14.0	16.1	41.6	28.3
33	96	11.5	30.0	40.9	17.6	10.1	30.6	40.1	19.2

* The ^{131}I remaining at the origin of the chromatogram was excluded from the present analysis, as it was considered mostly to be attributed to the unhydrolyzed ^{131}I-thyroglobulin.

Table 8 Summary of chromatographic analysis of thyroid gland pancreatin digest in transplanted dogs. (% of total ^{131}I in the gland)

Site of transplant	Dog No.	Time after transplantation (days)	Transplanted lobe				Intact lobe			
			I'	MIT	DIT	T_3, T_4	I'	MIT	DIT	T_3, T_4
Groin	45	3	13.1	34.8	39.1	13.0	13.6	33.4	34.6	18.4
	46	5	18.6	36.3	32.4	12.7	23.4	32.8	27.3	16.5
	43	7	12.3	30.2	42.0	15.5	13.2	28.8	41.4	16.6
	51	7	17.7	31.0	37.2	14.1	19.3	33.4	33.0	14.3
	66	8	17.4	34.0	33.3	15.3	*	*	*	*
	41	9	13.5	31.4	37.6	17.5	14.0	30.0	39.9	16.1
	54	11	15.6	36.1	31.6	16.7	20.1	37.6	27.7	14.6
	55	11	17.2	31.1	35.1	16.6	20.8	31.0	30.9	17.3
	47	13	11.2	31.2	37.3	20.3	12.7	28.2	39.9	19.2
	49	13	12.6	29.4	38.9	19.1	14.5	27.3	38.9	19.3
	62	13	13.4	24.7	40.6	21.3	14.6	20.1	46.2	19.1
	61	15	15.3	36.8	32.4	15.5	14.1	35.4	32.0	18.5
	74	15	15.2	35.3	37.2	12.3	18.9	35.1	30.1	15.9
	48	20	16.3	32.6	30.2	20.9	18.5	30.1	30.2	21.2
	56	30	**	**	**	**	11.6	35.6	30.2	22.6
	78	30	11.9	31.9	39.7	16.5	15.3	31.6	35.8	17.3
	20	60	13.7	32.7	46.6	7.0	13.7	32.6	43.8	9.9
	21	60	10.1	28.5	41.8	19.6	10.4	27.1	40.5	22.0
	22	60	14.6	36.7	40.1	8.6	14.1	30.8	43.1	12.0
	23	60	**	**	**	**	7.4	29.2	40.5	22.9
Neck	70	5	8.9	39.6	40.7	10.8	14.3	38.3	34.4	13.0
	72	5	13.3	32.6	40.7	13.4	13.5	32.0	41.0	13.5
	73	5	14.7	36.6	40.0	8.7	16.7	37.1	34.4	11.8
	82	14	13.3	34.3	41.3	11.1	16.0	33.9	40.0	10.1
	71	20	27.2	33.8	27.1	11.9	34.3	29.1	23.1	13.5
	76	20	20.0	39.1	31.5	9.4	22.9	37.3	28.9	10.9

* Extirpated ** Uptake 0%.

Table 9 Summary of chromatographic analysis of thyroid gland pancreatin digest. (% of total ^{131}I in the gland)

Group	Dog No.	Time after operation (days)	right lobe#				left lobe			
			I'	MIT	DIT	T3, T4	I'	MIT	DIT	T3, T4
L	83	5	18.4	34.8	31.8	15.0	20.3	35.4	30.4	13.9
	88	5	11.3	35.9	36.6	16.2	14.4	33.0	35.3	17.3
	91	14	14.5	35.7	39.9	9.9	15.4	32.8	37.2	14.6
	92	22	20.0	38.1	28.1	13.8	19.8	38.8	29.5	11.9
M	84	5	16.3	38.7	33.1	11.9	20.7	39.9	28.5	10.9
	87	5	13.2	21.8	43.3	21.7	13.3	21.0	43.3	22.4
G	93	5	13.5	28.3	34.0	24.2	14.5	27.6	33.0	24.9
	95	5	12.3	36.4	35.3	16.0	10.4	37.1	30.8	21.7
	96	5	16.8	36.8	31.9	14.5	21.5	36.2	28.4	13.9
D	85	5	13.9	38.5	32.9	14.7	14.4	36.2	33.3	16.1
	86	5	16.8	27.5	37.8	17.9	15.9	28.4	37.5	18.2
	27	62	18.9	34.9	35.4	10.8	19.0	35.6	35.2	10.2
	35	68	15.8	32.9	32.8	18.5	14.8	33.5	32.3	19.4
	36	68	12.8	31.9	32.6	22.7	14.1	31.5	31.2	23.2

As for the classification of groups, see the footnote of Table 6.
#: Operation was done on the right side of the neck.

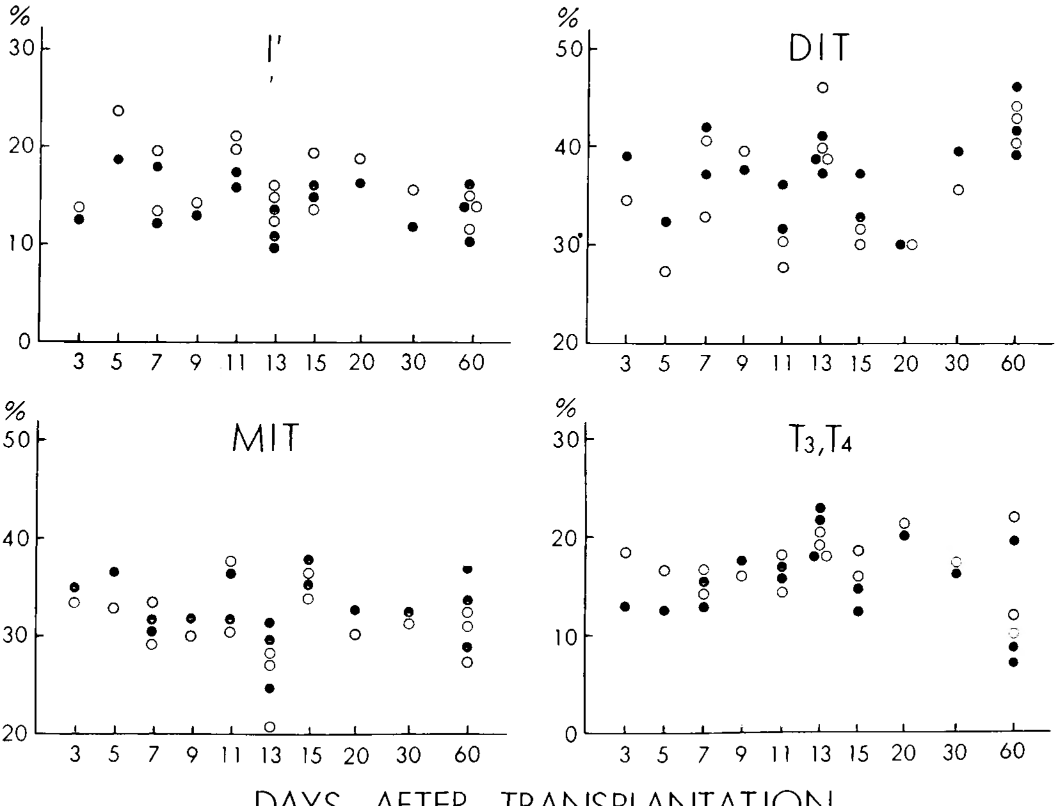

Fig. 5 Contents of ^{131}I-compounds in the thyroid gland in the transplantation into the groin. *Circle*, Intact lobe. *Dot*, Transplanted lobe.

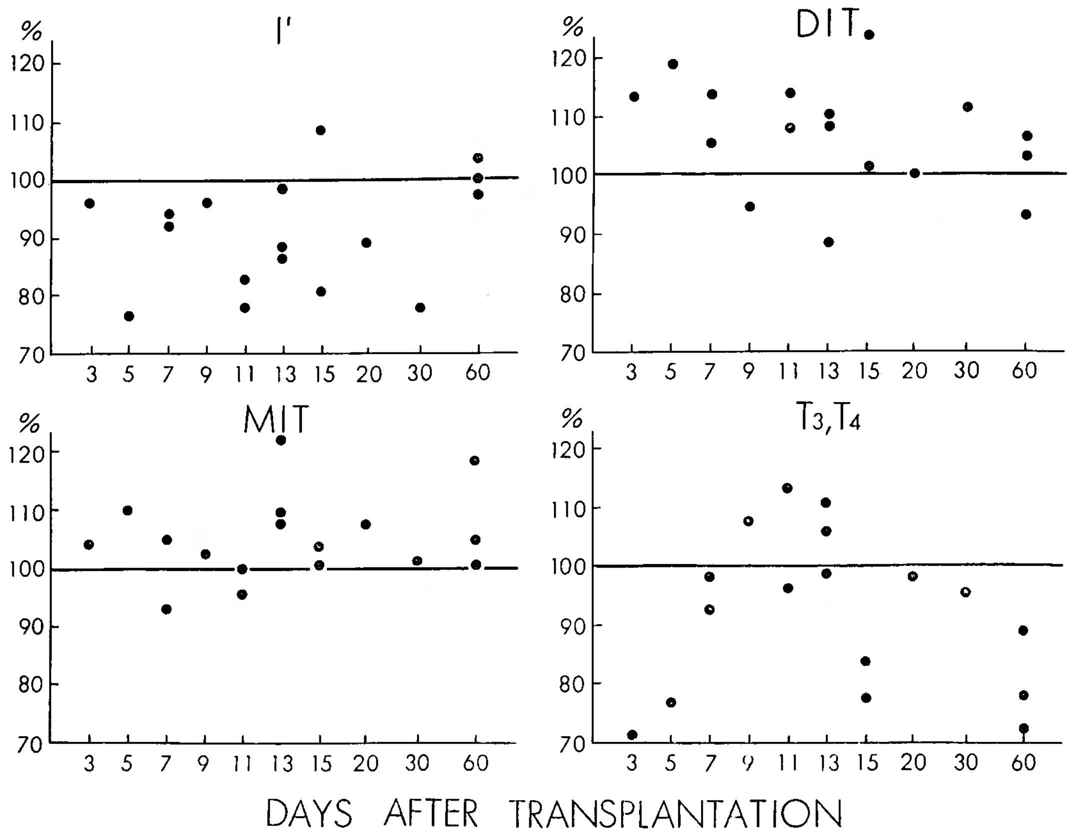

Fig. 6 Comparison of the contents of thyroidal ^{131}I-compounds of both thyroid lobes in the transplantation into the groin (transplanted/intact ratio).

6. Effect of TSH on serum PBI level

After a single injection of 5 units of TSH, the serum PBI level in femoral venous blood increased from a basal level of 3 04 mcg/100 ml to a peak level of 10.1 mcg/100 ml three hours later in a 5-day-old transplant (dog No. 66, Table 10 and Fig. 7). In the control dogs it took 6 to 12 hours to get the peak level. In the 74-day-old transplant, however, the response was neither prompt nor marked.

7. Angiogram

Angiograms of the grafts were taken immediately before sacrifice. The artery and vein of the graft were demonstrated to be well patent. Angiogram showed the collateral circulation already on the 7th day, while the anastomosed vein was patent. The same findings were observed in all angiograms which were made thereafter.

8. Histology

Thyroid tissue composed of follicles varying in size, many of which contained colloid, was seen. In the specimens of 3- to 9-day-old transplants, there were many cells within the colloid. They were not the infiltrating lymphoid cells but the desquamated acinar cells, and it was found that these specimens were from the thyroid lobes which had been subjected to the angiography with 76 % urografin. A few vacuoles appeared at the

Table 10 Effect of thyroid stimulating hormone (TSH) on serum protein-bound iodine (PBI) level. #
(mcg/100 ml)

Dog No.	Sex	Time after transplantation (days)	Serum PBI level Time after TSH injection (hrs.)						
			before	3	6	12	24	48	72
66	M	5~8	3.04	10.1	7.2	4.6	2.56	—	5.5
80	M	14~17	2.5	2.04	4.6	1.2	1.3	2.9	2.36*
90	M	14~17	2.59	1.15	0.2	0.93	—	—	—**
75	F	30~33	2.75	9.12	—	6.02	3.89	4.37	2.5
75	F	74~77	2.25	3.15	3.75	5.66	4.08	1.81	2.32
69	M	control	4.18	4.95	7.9	6.8	—	4.1	2.57
98	F	control	4.83	6.79	8.2	11.07	3.85	1.81	2.32

* Artery and vein were ascertained to be patent in angiogram which was made on the 26th p.o.d.
** Died on the 17th p. o. d.
# In all cases, the left lobe had been extirpated.

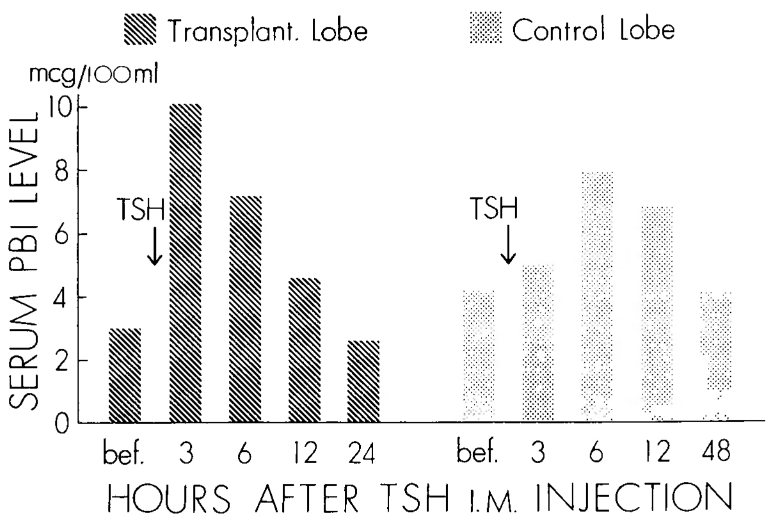

Fig. 7 Graphic representation of the effect of TSH on serum PBI level. Left, 5-day-old transplant (dog No. 66). Right, Control (dog No. 69).

periphery of the colloid in the specimen of 20-day-old transplant. In all instances, there was no lymphoid cell infiltration and no fibrous reaction. In the control, in which the X-ray contrast media was injected and left as it was for 2-3 minutes, the marked desquamation of the acinar cells into the colloid was seen.

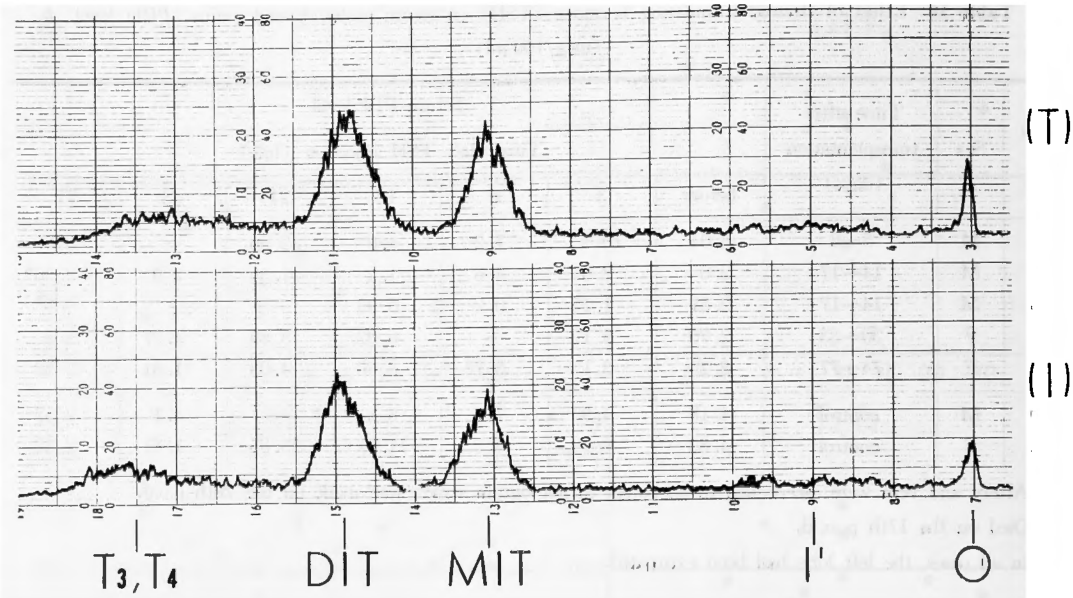

Fig. 8 The scan of chromatograms obtained from dog No. 21 (60 days after transplantation). The transplanted lobe (T) shows the hormone synthesis fairly well. (I) : Intact lobe.

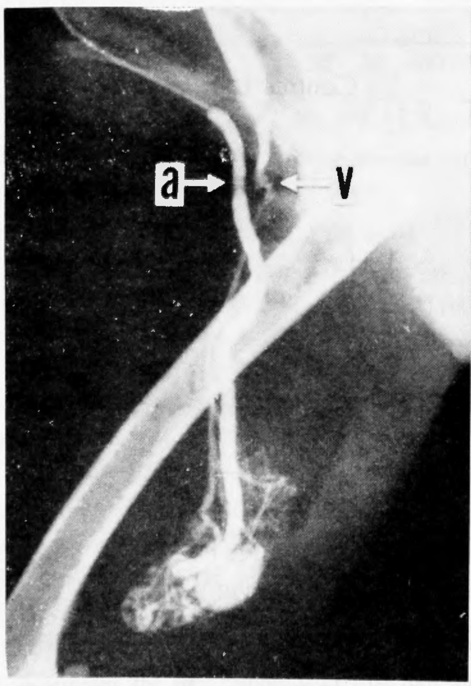

Fig. 9 Angiogram of the graft, 3 days after transplantation (dog No. 50).
a : artery, v : vein. Arrows indicate the sites of vessel anastomoses.

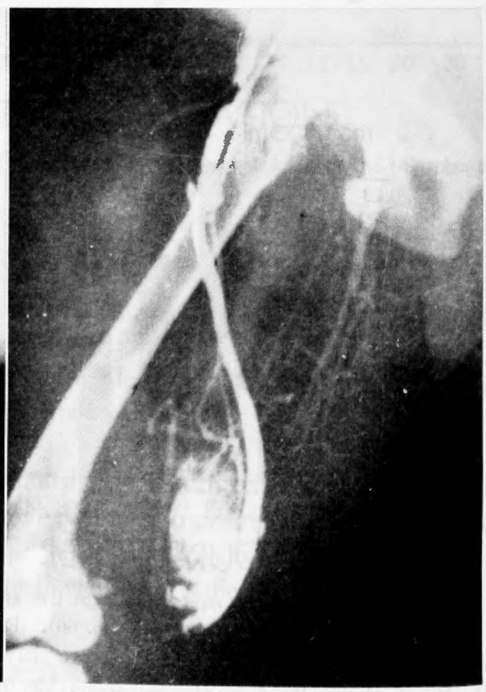

Fig. 10 Angiogram of the graft, 7 days after transplantation showing the collateral circulation in spite of the patency of the graft vessels (dog No. 43).

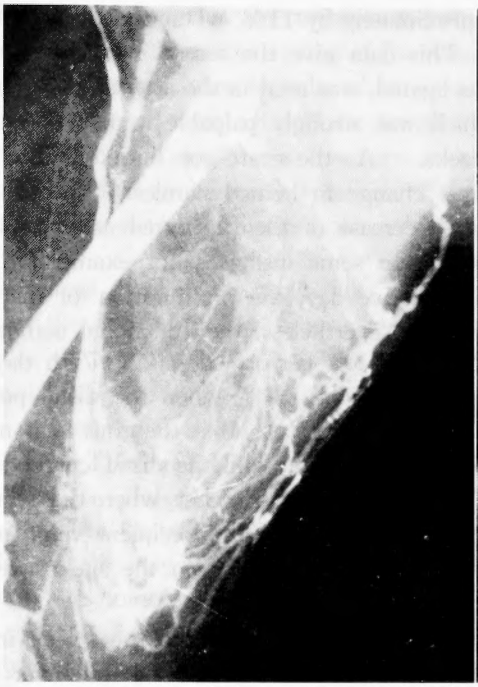

Fig. 11 Angiogram of the graft with collateral circulation, 13 days after transplantation (dog No. 47).

Fig. 12 Angiogram of the graft with collateral circulation, 60 days after transplantation (dog No. 21).

DISCUSSION

Micro-vascular anastomosis and thyroid transplantation. There are two types of free grafting of organs: the implantation of fragments of tissues and the transplantation of a whole organ with immediate reconstruction of blood supply. Implanted tissues are not provided with the immediate blood supply and they must survive by diffusion of body fluid until the spontaneous vascularization occurs. They may undergo progressive degeneration particularly at their cores, though a regeneration would follow. There would be, therefore, a few weeks lag before the surviving grafts might show a hormonal activity. The development of vascular surgery made it practicable to transplant a whole organ by blood vessel anastomosis. GOODMAN[19] and KAWAMURA[32] reported the successful autotransplantations of the thyroid gland with an intact blood supply in dogs as early as in the second decade of this century. However, because the vessels of the thyroid are small in size, it was not easy to obtain the suitable caliber of the vessels, particularly of the vein, which was sufficient for the micro-vascular surgery. In the early stage of the present experiment the internal jugular vein, which was small in dogs and the outside diameter was 1.2 to 2.0 mm in most cases, was directly anastomosed to the recipient vein, but the results were unsuccessful. This obstacle was overcome by using a patch of the wall of the brachiocephalic vein, in the center of which the entrance of the internal jugular vein located. This device made it possible to use the bushing of at least 3.0 mm in diameter in the venous anastomosis, and the outcome of the micro-vascular surgery became satisfactory.

The thyroidal blood flow rate increased approximately by 11% when the distal portion of the common carotid artery was occluded. This data give the reason why the segment of common carotid, the distal end of which was ligated, was used in the arterial anastomosis. However, the pulsation of the graft artery, which was strongly palpable immediately after the operation, became weaker after some weeks. As the graft got buried in the proliferated connective tissue, it gave rise to some change in hemodynamics. This change might play a somewhat significant role in the decrease of the functional activity of the graft, as was seen in the 60-day-old transplants. In some instances, for example in dog No. 75 which was sacrificed on the 99th postoperative day, the proliferation of the connective tissue around the graft was very slight. Nevertheless, the functional activity of this transplant seemed to be suppressed in PBI-TSH test performed on the 74th day.

The venous pressure in the recipient vein was reduced by 33% when the distal portion of the femoral vein was occluded. It seemed desirable, therefore, that the graft vein might be anastomosed to the proximal cut end of the recipient vein, while its distal cut end was ligated. The venous pressure in the recipient vein would become higher when the segment of the graft vein was interposed between the both cut ends of the recipient vein, and it might be unfavourable against the prevention of the thrombosis in the site of venous anastomosis.

HAMOLSKY et al.[20] implanted the autologous thyroid tissues into the spleen in the rats and observed the subsequent low levels of $PB^{131}I$ and concluded that the liver inactivated a fraction of thyroid hormone. The location of the graft in the endocrine tissue transplantation must be carefully chosen.

The ^{131}I uptake and hormone synthesis of the thyroid. The evidence of the functional activity was present in every transplant except for some cases in which the anastomosed vessels were thrombosed. The transplanted thyroid lobe took up the ^{131}I as early as on the 3rd postoperative day, and it recovered the normal activity on the 13th day. It was noted that the iodine accumulation in the intact lobe was also slightly reduced for some period following operation. It is of some interest to analyze the cause why the thyroid activity is apparently suppressed in the early days after operation, as there is a clinical observation that the remnant tissues of the hyperfunctioning thyroid diminish in its size following surgery[29]. The following speculations may be assumed: 1) It is well known that the assaults of anesthesia and operation cause the anterior pituitary to release the adrenocorticotropic hormone, and it is also known that the adrenocortical substances diminish thyroid activity[22]. SHIBUSAWA et al.[47] reported that the thyroid activity was suppressed by the surgical injury inflicted to the body, but its extent was much greater than that which was caused experimentally by the administration of ACTH and cortisone. All the suppression of thyroid activity may not be caused by the increased secretion of adrenocortical substances. 2) The systemic demand for thyroid hormone may be reduced at this time, because the nitrogen balance is shown to become negative following operation[24], and the action of thyroid hormones is to stimulate the nuclear RNA synthesis and sequential cytoplasmic protein synthesis[57][62]. 3) A contrary guess may be possible that the suppression of the thyroid function could be produced by the negative feedback mechanism by acting thyroid hormones which may appear in the circulation following surgical trauma inflicted to the thyroid gland. In any case, further studies will be required.

It was clarified that the transplanted thyroid gland synthesized the hormones fairly well. However, close examination of the data makes it manifest that the functional activity of the transplanted lobes is reduced to some extent, when compared with that of intact lobes of respective dogs. It will be noted that the transplanted lobes show relatively low values in both ^{131}I uptake and hormone content (T_3 & T_4) in general. There seems to exist some defect in hormone synthesis of the transplanted lobes. Some factors caused by the procedures of transplantation may reduce their activities. This problem will be referred to later.

Humoral and neural regulations on the thyroid transplant. A close relationship exists between the thyroid and the anterior pituitary. Administration of TSH to normal animals produces an increase in both uptake of ^{131}I and a secretion of thyroid hormones, resulting in an elevation of the serum PBI level. On the contrary, administration of thyroid hormones causes the decrease in TSH level. In the present study, it was observed that the injection of TSH to the transplanted dog resulted in an elevation of serum PBI level, followed by a probable rebound phenomenon. It is obvious, therefore, that the transplanted thyroid is well controlled by the humoral regulation of TSH.

It is well known that the endocrine and nervous systems correlate to each other to maintain the homeostasis of the body[43]. The nervous stimulation of the anterior region of the hypothalamus causes the liberation of TRF (TSH releasing factor), and TRF is then transported via the hypophysial portal system to excite the anterior pituitary[25].

The relation of the sympathetic nervous system to the thyroid function is by no means clear. Numerous experiments have been done, but the results were conflicting. CANNON et al.[7] made phrenico-sympathetic anastomosis in the cat, and 3 months later, the animals showed the symptoms characteristic of hyperthyroidism. HANEY[21] reported that the faradic stimulation of the cervical sympathetic trunk in rabbits resulted in a marked rise in the basal metabolic rate. HANEY concluded that this increase in metabolism was due to an acceleration of the functional activity of the thyroid gland. However, FRIEDGOOD et al.[16] could not find a consistent effect on the rate of metabolism in a similar experiment. In one of the most recent attempts to establish the autonomic nervous control of thyroid function, IINO[28] and ISHII[30] presented evidences probably in favour of such control. IINO observed that the stimulation of the cranial cervical ganglion resulted in an increased $PB^{131}I$ level in the thyroid venous blood. ISHII reported that the stimulation of the nodosal ganglion also caused an increased $PB^{131}I$ level. IINO and ISHII concluded that the cervical sympathetic and vagus nerves played an accelerating role in the regulation of the thyroid function. In the present study, the transplanted lobes of the thyroid showed the reduced value of ^{131}I uptake when compared with the intact lobes. The content of T_3 & T_4 was also lower, as the coupling of MIT and DIT to form T_3 & T_4 was probably disturbed. And, when the unilateral thyroid was denervated by resecting the autonomic ganglia, the ^{131}I uptake decreased in the ipsilateral thyroid lobe. A hypothesis might be drawn to explain these findings that the autonomic nerves, predominantly the cervical sympathetics maybe, may have some effect on the thyroid activity, by accelerating the action of TSH which increases the iodine uptake and hormone formation of the thyroid gland.

Hypersensitivity of the denervated transplant to the humoral factor. CANNON[6] stated that "when in a series of efferent neurons a unit is destroyed, an increased irritability to chemical agents develops in the isolated structures, the effect being maximal in the denervated part." CANNON gave an example that when stimulatory adrenergic fibers degenerate, the dilator muscle of the iris becomes more than normally responsive to adrenalin, and widely dilated, in certain conditions, on the paralysed side than on the normal side. This was true on the cholinergic nerves, and the glands likewise responded to a greater degree to chemical stimuli after they have been freed from nervous control. In the present study, the content of T_3 & T_4 of the transplanted thyroid, which was usually decreased, became increased from the 9th to 13th postoperative days (Fig. 6, bottom right). Furthermore, in PBI-TSH test, the transplanted thyroid seemed to respond more promptly than the control thyroid (Dogs No. 66 and 69, Table 10 and Fig. 7). These results suggest that the free transplant of the thyroid, the sympathetic and vagus nerves of which, both are considered as stimulatory to the thyroid function, are resected, show an increased sensitivity to chemical stimulation of TSH. In dog No. 75, the response was prompt and greater in amount when the graft was a month old; however, it became somewhat retarded and smaller in amount on the 74th day.

Effect of severance of lymphatics on the graft function. DANIEL et al.[11] observed that the radioactive iodine was released from the thyroid gland of the baboon into the lymphatic vessels which drained the thyroid as well as into the thyroid venous blood. Some 90% or more of the radioiodine was in organic form, and its concentration in the thyroidal lymph was very much higher than that in the thyroid venous blood. It is not yet concluded whether this lymphatics may be certain pathways of hormone secretion or not. In the present study, the thyroidal lymphatics were ligated unilaterally in order to clarify the effect of the severance of the lymphatics on the functional activity of the transplant. The ^{131}I uptake of the ipsilateral lobe did not differ from the contralateral intact lobe on the 5th postoperative day. It was reduced on the 14th day, and became increased on the 22nd day in another dog.

The development of collateral circulation around the graft. The angiograms of the grafts demonstrated the collateral circulations in spite of the patency of the graft vessels. This was noted already on the 7th day after operation and persisted as long as 60 days. These findings might indicate that some portion of the graft could survive even if the graft vein would thrombose, because some venous blood might return through the outflow collaterals, though the thyroid activity would be limited. Dog No. 61 may be an instance of such case (Table 5).

Histologic findings. It must be remembered that the transplants were subjected to the angiography immediately before their extirpation. Control specimens, in which 76% urografin was injected and left as it was for 2-3 minutes, showed a similar histological change of desquamation of acinar cells. The histology of the transplants must be considered with some subtraction. Be that as it may, the damage of the follicles of the transplants seems to be the greatest on the 7th and 9th days, and it may be expected to show the normal histology on the 13th day, which is concordant with the functional recovery. It is noted that no lymphoid cell infiltration was seen in any of the specimens.

Possibility of the occurrence of autoimmune process in the thyroid auto-transplantation. It is well known recently that the pathogenesis of the chronic thyroiditis (Hashimoto's disease) and hyperthyroidism (Graves' disease) is largely due to the auto-immune process[52]. WITEBSKY[60] proved that the extracts of autogenous thyroid gland given to the dog with Freund adjuvants were antigenic for the individual animal itself, and he stated that a true auto-antigen, presumably thyroglobulin, could produce antibodies capable of acting upon the normal thyroid tissue. In the present study, the precipitin reaction was attempted between sera of some transplanted dogs and crude saline extracts of normal canine thyroids. However, it failed to demonstrate the circulating auto-antibodies. There has been also no evidence of thyroiditis in histologic examination of the transplants. The possibility of the occurrence of auto-immune process in the autologous thyroid transplantation would be, therefore, excluded.

The clinical observations of thyroid grafting in the literature. The autotransplantation of the human thyroid tissue has been attempted by SWAN et al.[53], JONES[31], LOW et al.[37] and SKOLNIK et al.[49], following the removal of a lingual ectopia of the thyroid. These slices of the tissues were implanted into the sternocleidomastoid, pectoral and abdominal rectus muscles with a variable degree of success. SZILAGYI et al.[54] made the experimental autotransplants of human thyroid in slices in nine cases, following subtotal and total thyroidectomy. The transplants showed a constant ability to survive, and their functional activity increased with time. The rate and magnitude of increase in function depended on the type of tissues transplanted, the grafts of hyperplastic goiter (Graves' disease) showing the greatest growth potentiality. Homotransplantation of thyro-parathyroid glands were made by BURDETTE,[5] NICKS[41], SCHATTEN et al.[46], STERLING[50] and CONWAY et al.[9] in the patients with hypothyroidism and hypoparathyroidism. STERLING et al.[51] reported a case in which the glands were obtained from a newborn infant and the carotid and jugular vessels were anastomosed to the host vessels in the groin, and the patient's condition was improved. WATKINS et al.[59] removed all the pretracheal tissues from premature infants in continuity with the aorta and superior vena cava, which were anastomosed to suitable vessels in the groin in 3 patients, and in one patient the complete elimination of all medication requirement was obtained. Successful transplantation enabled the patient to dispense with oral substitution therapy, which was always available if the graft should fail.

SUMMARY

1) The canine thyroid glands were transplanted autologously into the groin using micro-vascular anastomoses, and the functional activities of the transplants were investigated by means of radioactive tracer studies and PBI-TSH test.

2) The sufficient size of the vessels which could be expected to succeed in micro-vascular anastomosis was at least 3.0 mm in outside diameter, both with artery and with vein.

3) Free transplants of autologous thyroid gland well took up the radioactive iodine and synthesized the thyroid hormones as early as on the 3rd postoperative day, and it recovered normal activity on the 13th day, though a slight drop in activity followed after a long period of time. The iodine accumulation in the intact lobe was also slightly reduced following operation, and its cause was discussed.

4) The injection of TSH into the transplanted dog resulted in an elevation of serum PBI level promptly and in a great amount; however, it became somewhat retarded and

small in amount after 74 days.

5) A close examination of the data made it manifest that the functional activity of the transplanted lobes was reduced to some extent when compared with that of intact lobes of respective dogs. There seemed to exist some defect in hormone synthesis in the transplanted lobes. The cause was presumed to be due to changes in hemodynamics caused by the procedures of transplantation, and due to the denervation of the sympathetic and vagus nerves. The autonomic nerves were assumed to have some effect on the thyroid activity by accelerating the action of TSH.

6) The content of T_3 & T_4 of the transplanted lobe, which was usually low, became higher than that of the intact lobe from the 9th to 13th postoperative days. Furthermore, in PBI-TSH test, the transplanted thyroid gland seemed to respond more promptly than the control thyroid gland. These results suggested that the free transplant of the thyroid (the sympathetic and vagus nerves both considered as stimulatory to the thyroid function were resected) showed an increased sensitivity to humoral stimulation of TSH. The law of denervation (CANNON) was assumed to exist between the denervated thyroid transplants and TSH.

7) The angiograms of the grafts showed the collateral venous circulations in spite of the patency of the graft vessels.

8) Histologic examination showed the desquamation of the acinar cells in early days following transplantation. The damage of the transplants seemed to be the greatest on the 7th and 9th days, and it was expected it would show the normal histology on the 13th day.

9) It failed to demonstrate the auto-antibodies against the thyroid tissues in the sera of the transplanted dogs. Also, the lymphoid cell infiltration was completely invisible in all specimens. The possibility of the occurrence of auto-immune process in the thyroid auto-transplantation was discussed and it was excluded.

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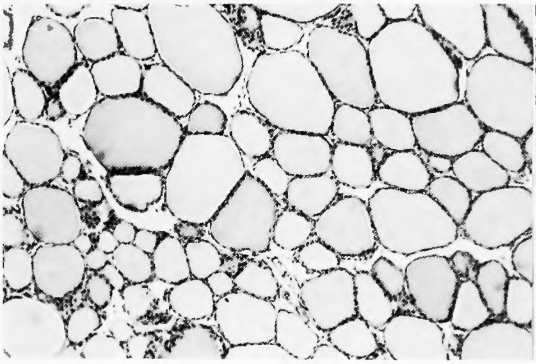

Fig. 13 Photomicrograph of the normal thyroid gland of the dog. (×100, H-E)

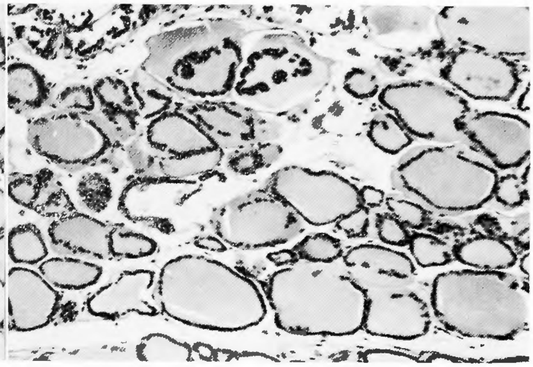

Fig. 14 Photomicrograph of the thyroid gland subjected to angiography with 76 % urografin. The X-ray contrast media was injected and left as it was for 2-3 minutes. (×100, H-E)

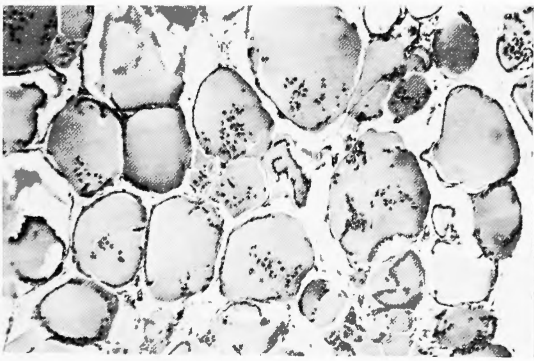

Fig. 15 Photomicrograph of 3-day-old thyroid auto-transplant showing the desquamation of acinar cells into the colloid. This transplant had been subjected to the angiography (Dog No. 50). (×100, H-E)

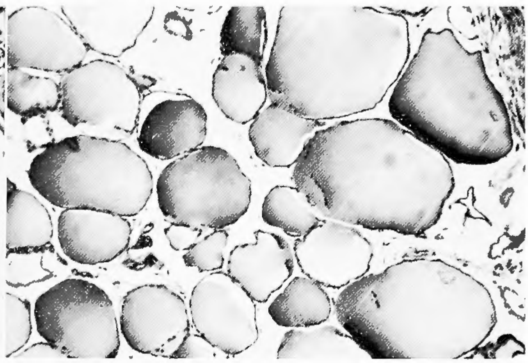

Fig. 16 Photomicrograph of intact thyroid lobe of Dog No. 50. (×100, H-E)

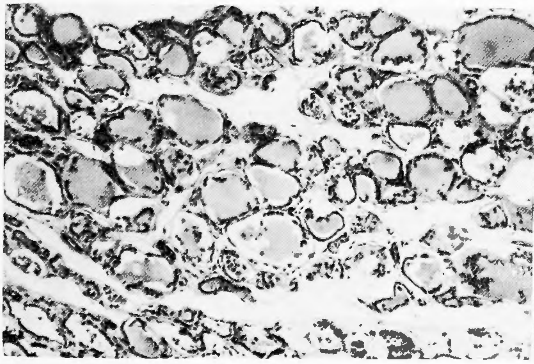

Fig. 17 Photomicrograph of 7-day-old thyroid auto-transplant, subjected to the angiography (Dog No. 51). (×100, H-E)

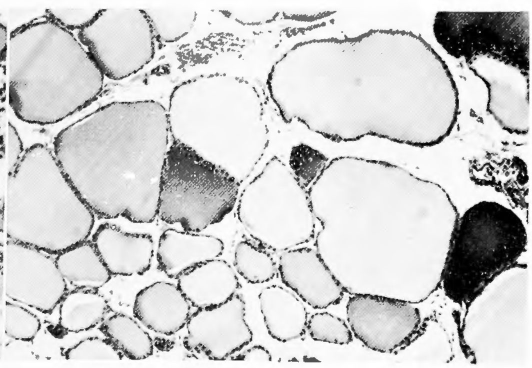

Fig. 18 Photomicrograph of intact thyroid lobe of Dog No. 51. (×100, H-E)

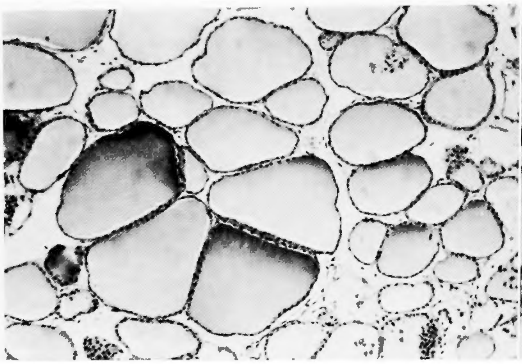

Fig. 19 Photomicrograph of 13-day-old thyroid auto-transplant, subjected to the angiography (Dog No. 47). (×100, H-E)

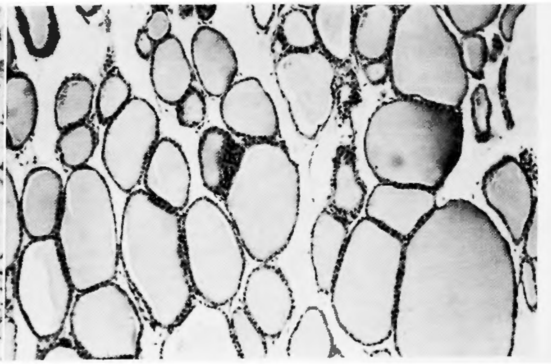

Fig. 20 Photomicrograph of intact thyroid lobe of Dog No. 47. (×100, H-E)

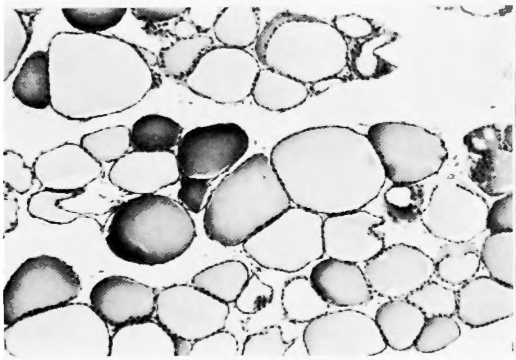

Fig. 21 Photomicrograph of 20-day-old thyroid auto-transplant, subjected to the angiography (Dog No. 48). (×100, H-E)

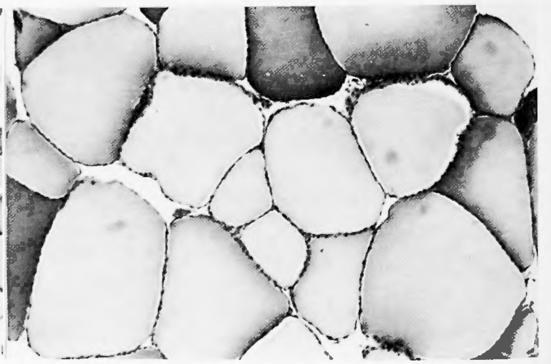

Fig. 22 Photomicrograph of intact thyroid lobe of Dog No. 48. (×100, H-E)

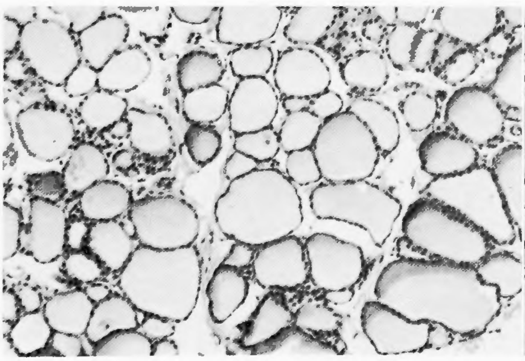

Fig. 23 Photomicrograph of 99-day-old thyroid auto-transplant (Dog No. 75). (×100, H-E)

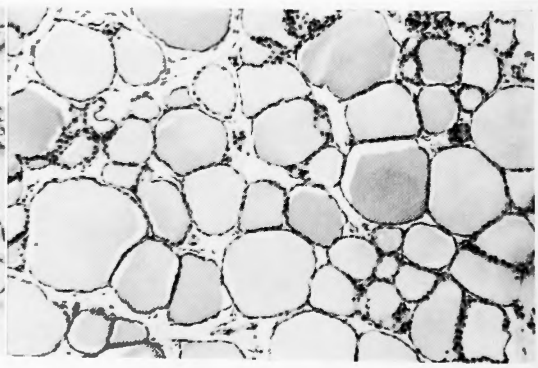

Fig. 24 Photomicrograph of intact thyroid lobe of Dog No. 75, extirpated on 28th p. o. d. (×100, H-E)

和文抄録

細小血管吻合を使用した甲状腺自家移植に関する実験的研究：とくに移植甲状腺のヨード代謝ならびに体液性調節について

京都大学医学部外科学教室第2講座（指導：木村忠司教授）

長　嶺　慎　一

近年，細小血管吻合手技の発達にともない，内分泌腺を臓器全体として移植することが可能となつた．

著者は，犬甲状腺を総頸動脈および腕頭静脈をつけて取り出し，それぞれ大腿動脈および大腿静脈へ血管吻合をおこなつて自家移植し，移植腺機能を検討する目的で，

1）追跡量の放射性ヨード約50 μCi を投与して甲状腺ヨード摂取率を測定した．

2）甲状腺組織中の無機ヨードならびに各ヨードアミノ酸（モノヨードチロジン，ジヨードチロジン，トリヨードサイロニンおよびサイロキシン）の含有比率を濾紙クロマトグラフィーで分析した．

3）移植甲状腺にTSH（5単位筋注）を負荷して血清PBI値の変動を測定した．

4）移植にともなう自律神経ならびにリンパ管切断が，移植腺機能におよぼす影響を検討した．

5）自家甲状腺移植犬の血中自己抗体を Ouchterlony 法で検討し，また組織学的検査をもあわせておこなつて，次の結論を得た．

1）吻合に使用する血管の大きさは，動脈および静脈とも外径が3.0mm以上を有することが必要である．

2）自家移植甲状腺は良好に生着し，移植13日目には正常水準の機能を回復したが，移植2ヵ月目にはやや機能が減退した．正常側甲状腺も術後しばらくはヨード摂取率低下を示した．

3）移植腺にTSHを負荷すると，血清PBI値が迅速に上昇したので，移植甲状腺は体液性調節を良好に受けることが明らかとなつた．しかし，移植74日目には，反応はやや遅延しその頂値も低かつた．

4）実験成績を詳細に検討すると，移植腺機能は同一犬の正常側腺と比較してやや減退を示し，移植腺のホルモン合成能が障害されていることが認められた．機能減退の原因は，移植にともなう血行動態の変化ならびに移植腺の交感および迷走神経切断の影響が関与していると考えられた．甲状腺支配自律神経は，甲状腺に対するTSHの作用を微妙に促進していると考えられた．

5）移植甲状腺組織中のホルモン（サイロキシンおよびトリヨードサイロニン）の含有比率は，一般に正常側腺よりも低いが，移植後9日目から13日目にかけては，移植腺の方が高い値を示した．またPBI-TSH試験で，移植腺の方が対照よりもTSH負荷に対してより迅速に反応する時期が認められた．これらの成績は，甲状腺機能に対して促進的に働くといわれる交感・副交感の両自律神経を切断された移植甲状腺が，液性支配因子であるTSHに対して過敏な反応を呈する時期があることを暗示し，Cannon の神経切断の法則が移植甲状腺とTSHの間にも成立することを示唆するものと思われた．

6）移植腺の血管撮影では，移植7日目以後の全例に，吻合静脈本幹がよく開通しているにもかかわらず，側副血行の発生が認められた．

7）移植腺の組織学的検査では，コロイド含有濾胞が一般に認められたが，移植後早期のものでは濾胞上皮の剝離が強かった．組織像の対照として76%ウログラフィンを注入して2，3分間放置した甲状腺に，同様の上皮剝離を認めたので，造影剤による影響を考慮しなければならないが，7日目から9日目にかけて組織障害の程度が最も強く，13日目にはほぼ正常の組織像を示した．

8）リンパ球浸潤はどの標本にも全く認められなかつた．血中自己抗体も証明されなかつたので，甲状腺自家移植にともなう自己免疫現象の発現は一応否定された．