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Hormonal Control of Ossification of the Caudal Vertebrae in the Rat

III. Skeletal reponse of hypophysectomized and normal female rats to hormonal therapy

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The objective of the present study is to determine the pattern of reaction of the caudal vertebrae of hypophysectomized and normal female rats to hormonal therapy and to evaluate their usefulness for the biologic assay of hormonal preparations. Standards of tail growth and development of normal and hypophysectomized female rats have been established previously (3, 4, 5).

Earlier studies (1, 2, 9) showed that growth hormone induced epiphyseal cartilage growth and stimulated endochondral and periosteal ossification in hypophysectomized rats. Chondrogenic and osteogenic processes were reestablished after postoperative intervals exceeding one year. When combined with thyroxin, growth hormone proved more effective than when administered alone. Similar results were obtained in the medial epicondyle of the humerus (6). In the same experiment the predominant effect of thyroxin was one of cartilage erosion without proliferation. This response prevailed in the epiphysis of the third metacarpal even when concurrently injected with pure growth hormone (7). A similar antagonism was described (8) in the condyle of the mandible. Corollary studies had established the additional fact that the skeleton of normal mature rats continues to grow as long as pure growth hormone is injected (10).

For the purpose of comparison, the consequences of substitution therapy as well as chronic treatment with pure growth hormone upon the

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tail of normal and hypophysectomized rats will be dealt with in the present report.

Material and method

The experimental arrangement, shown in Table 1, was as follows: A total of 41 female rats of the Long-Evans strain were hypophysectomized at 26 to 28 days of age and then divided into 4 groups:

Group 1 served as untreated hypophysectomized controls, and they were autopsied at 306/455 days after operation.

Some members of Group 2 were treated with growth hormone over extended periods; in others, again, growth hormone injections were initiated after a delayed postoperative interval.

In Subgroup 2A, injections were started 12 days following hypophysectomy and continued for 437 consecutive days[1].

Subgroup 2B received 39 injections after postoperative intervals of 257/406 days during a 7 week period.

Group 3 was injected with thyroxin subcutaneously after a postoperative period of 257/475 days during a 7 week period. After one week the initial dose of 7.5 γ was reduced to 5 γ.

Group 4 was given 39 combined doses of growth hormone and thyroxin during a 7 week period after a postoperative interval of 259/405 days.[2]

Group 5 is composed of 8 intact rats which from their 192nd day of life were given an average dose of 1.7 mg growth hormone during 437 consecutive days.[3]

The anus-tail tip length of all experimental animals was measured at the onset of the injection period and at intervals thereafter. The length of the ninth caudal vertebra was determined from roentgenograms taken at autopsy in the manner described in the previous papers (3, 4). The histologic analysis also was made according to this routine procedure.

Results

A. Metric findings

The growth response of the caudal vertebrae to hormonal therapy is shown in Table 1. The anus-tail tip length of the hypophysectomized rats (Groups 1, 2, 3, 4) at the onset of therapy averages 12.2 cm. Chronic administration of growth hormone elicited a growth of 5.7 cm to reach a total length of 18.2 cm, nearly equaling that of intact rats. Terminal administration of growth hormone to old hypophysectomized rats re-

[1] For further details of experimental conditions see *Simpson*, *Evans* and *Li* (12).

[2] The exact experimental conditions of groups 2A, 3 and 4 have been described by *Becks*, *Simpson*, *Evans*, *Ray*, *Li* and *Asling* (9).

[3] For more details of the experimental condition see *Evans*, *Simpson* and *Li* (11).

Table 1
Experimental Arrangement and Measurements of the Lengths of Tail and Ninth Caudal Vertebra*

	Treatment		Number of rats	Onset of treatment Postop. days	Total of inject.	Average measurements			
						Anus-tip length			9th c. v. Autopsy mm
	Group	Daly dose				Onset cm	Autopsy cm	Increase cm	
1	Uninjected control	–	10	332/444	–	11.9 ±0.6	12.1 ±2.4	0.2	5.82 ±0.16
2A	Chronic Growth h.	** 0.2 mg	5	40	437	12.5 ±2.4	18.2 ±3.8	5.7	9.32 ±0.19
2B	Terminal Growth h.	0.2 mg	10	257/406	39	11.9 ±0.6	13.9 ±0.94	2.0	6.82 ±0.32
3	Terminal Thyroxin	*** 5 μg	7	257/475	39	12.5 ±0.67	12.7 ±0.67	0.2	5.8 ±0.22
4	Terminal Combination	both above	9	259/405	39	12.5 ±0.7	16.0 ±0.97	3.5	8.08 ±0.13
5	Normal + Growth h.	**** 1.2 mg	8	192/221	437	18.7 ±0.2	21.1 ±0.28	2.4	10.35 ±0.25

* Averages and standard errors

** Average dose: 0.1 mg 111 days; 0.2 mg 270 days; 0.4 mg 22 days

*** Average dose: 7 μg 8 days; omit 2 days; 5 μg 11–38 days

**** Average dose: 0.4 mg 22 days; 0.6 mg 57 days; 1.0 mg 29 days; 1.5 mg 99 days; 2.0 mg 167 days

sulted in an increase of 2 cm. Thyroxin therapy under similar conditions virtually had no effect upon the vertebral dimension. Old hypophysectomized rats responded to terminal combination therapy with an average increase of 3.5 cm, disclosing the synergic effect of both hormones. The lengthening of 2.4 cm recorded in group 5 reveals that growth hormone effectively stimulate growth of the caudal vertebrae even in normal animals.

Measurements of length of the ninth caudal vertebra for each group also are found in Table 1. The individual data have been computed in the graph of Fig. 1. Dimensional changes of the ninth caudal vertebra of hypophysectomized rats following hormonal treatment reflect the same principles as those of the entire tail skeleton. The drastic stunting of growth following hypophysectomy is not checked by thyroxin administration (Group 3). A significant response of 1 mm average is elicited in the old hypophysectomized animals after terminal growth hormone therapy (Group 2B). The increase in length is almost doubled

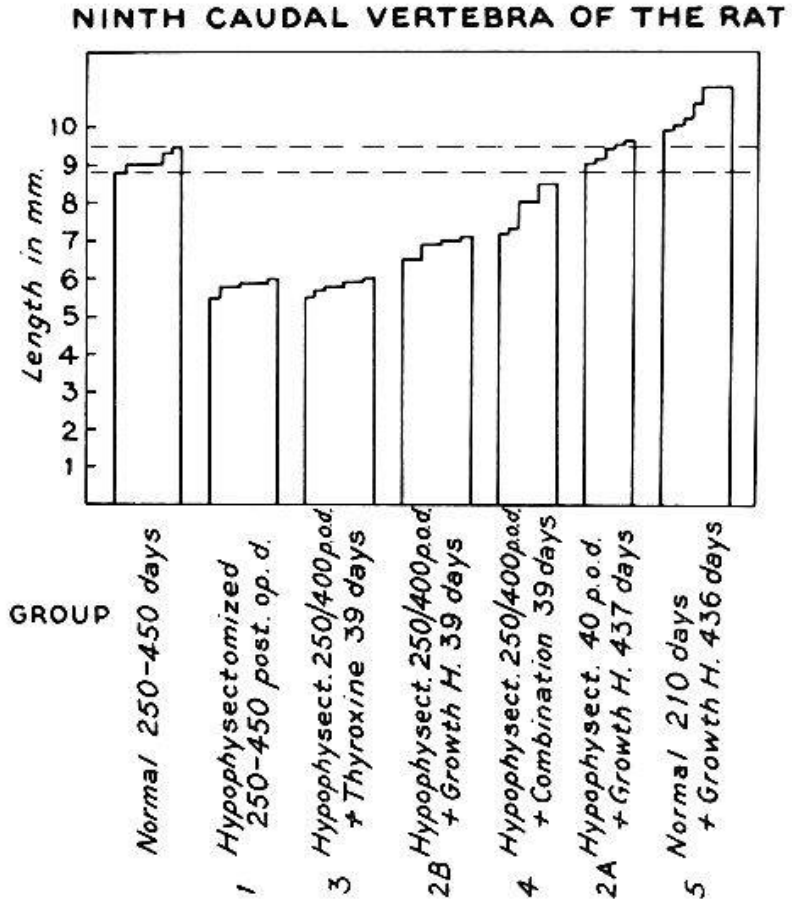

Fig. 1.

if thyroxin is given in combination with growth hormone (Group 4); it can be noticed that during the short therapeutical period of 7 weeks, some animals with 8.5 mm reached the lower margin of the normal littermates. A complete restoration to normal length of the ninth segment is achieved by chronic administration of growth hormone to young hypophysectomized animals (Group 2A). In view of the prolonged time of treatment, this response, however, does not match the spurt of the group which received terminally growth hormone and thyroxin concurrently. This is also true for the effect of growth hormone injected chronically to normal plateaued rats (Group 5). Based on the average of the normal controls (9.3 mm), the injected animals show an increase of 1.0 mm.

B. Histologic findings

1. For the purpose of comparison, the status of the ninth caudal vertebra of a rat 334 days after hypophysectomy is shown in Fig. 2. It will be noted that the diminutiveness in the longitudinal dimension is proportionate to a deficient transverse development.

2. Chronic administration of growth hormone to young hypophysectomized rats (Group 2A) produced increased size and density of bone.

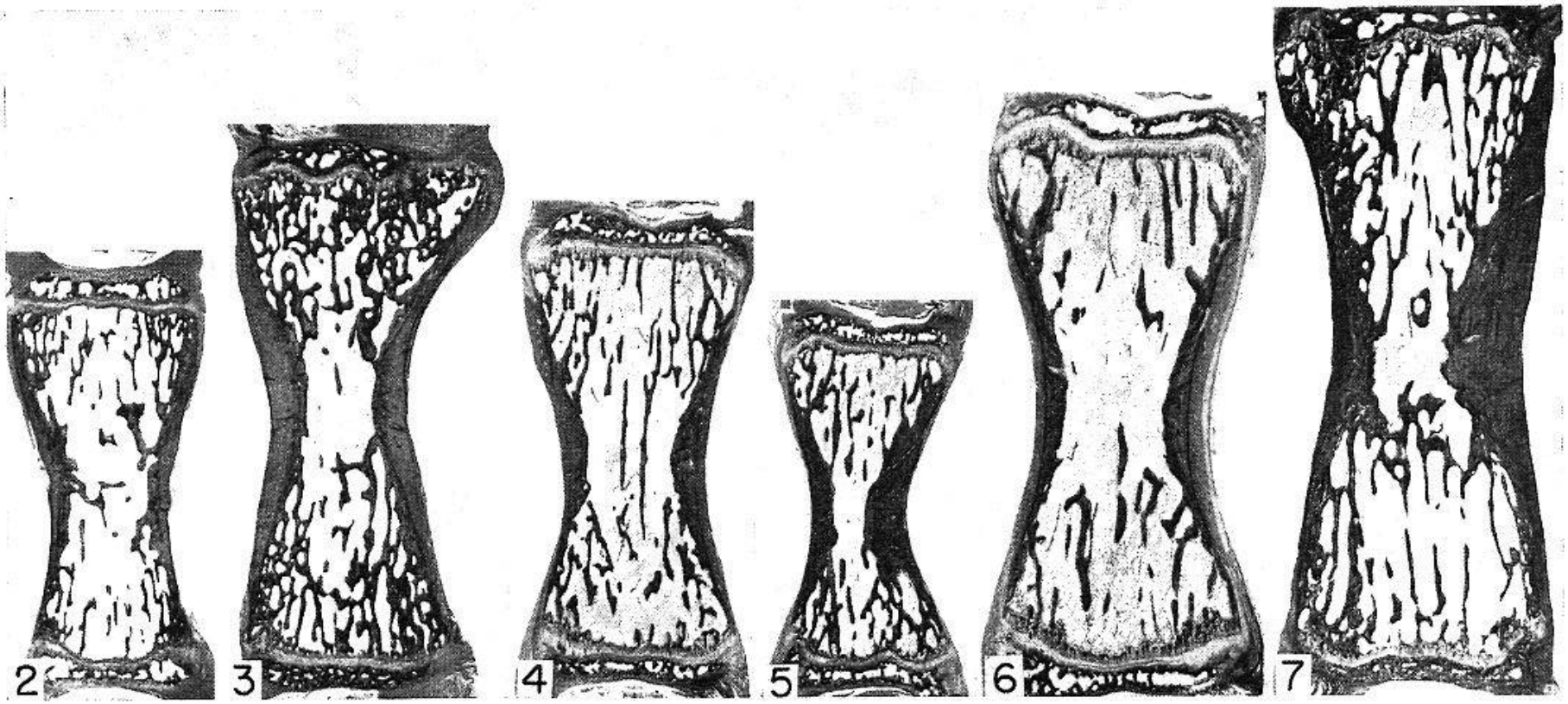

Plate 1 (Fig. 2–7)

Photomicrographs of the ninth caudal vertebra of rats hypophysectomized 26–28 days of age. Median sagittal sections, H. and E. stain, × 15.

Fig. 2. Uninjected control, autopsied 334 days after hypophysectomy. Sp. 8767 Pl. 9261

Fig. 3. Growth hormone, 0.2 mg injected daily for 437 postoperative days beginning at 40 days of age. Sp. 9023 Pl. 2309

Fig. 4. Growth hormone, 0.2 mg injected daily in 39 doses before autopsy at 331st day after operation. Sp. 8777 Pl. 2305

Fig. 5. Thyroxin, 5 μg injected daily in 39 doses before autopsy at 324th day after operation. Sp. 8779 Pl. 2306

Fig. 6. Combination, 0.2 mg growth hormone and 5 μg thyroxin injected in 39 daily doses before autopsy at 324th day after operation. Sp. 8787 Pl. 9367

Fig. 7. Growth hormone, 1.7 mg average injected daily for 436 days to a 196 day old normal rat. Sp. 2302 Pl. 2310

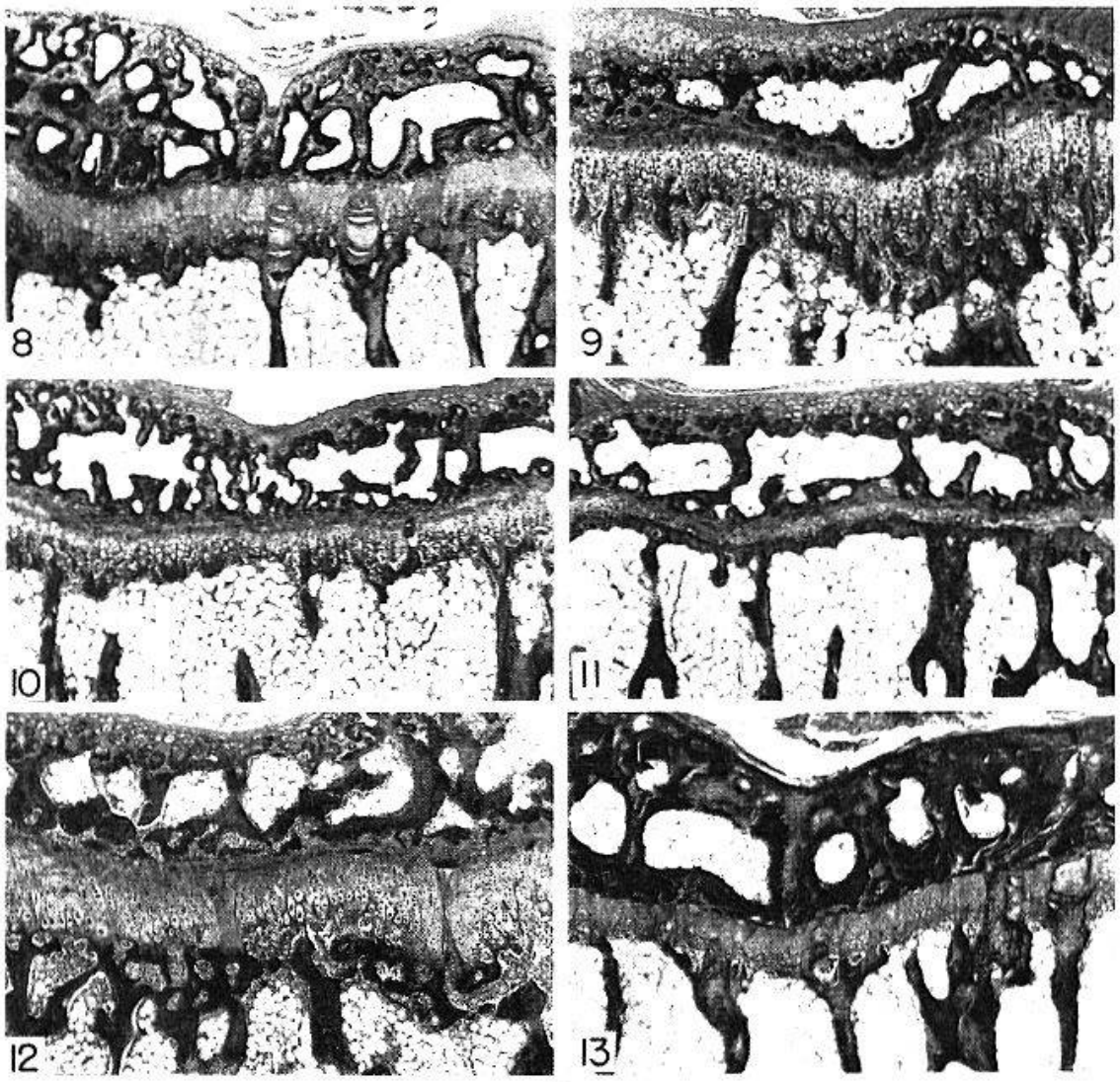

Plate 2 (Fig. 8-13)

Photomicrographs of proximal epiphysis of the ninth caudal vertebra of rats hypophysectomized at 26 to 28 days of age. Median sagittal section, H. and E. stain, × 90.

Fig. 8. Chronic injections of growth hormone (average 0.2 mg) for 437 consecutive days beginning 12 days after hypophysectomy. Sp. 9024 Pl. 2304

Fig. 9. Terminal injections of 39 doses of 0.2 mg growth hormone beginning 344 days after hypophysectomy. Sp. 8776 Pl. 8773

Fig. 10. Terminal injections of 39 doses of 5 μg thyroxin beginning 218 days after hypophysectomy. Optimal reactions are shown. Sp. 8779 Pl. 8776

Fig. 11. Terminal injections of 39 doses of 5 μg thyroxin beginning 278 days after hypophysectomy showing minimal response. Sp. 8780 Pl. 8775

Fig. 12. Terminal injections of 39 combined doses of growth hormone and thyroxin beginning 353 days after hypophysectomy. Sp. 8795 Pl. 8778

Fig. 13. Chronic injections of 436 daily doses of 1.7 mg (average) growth hormone to a normal rat beginning at 196 days of age. Sp. 8989 Pl. 2303

Although normal length is attained, width remains inadequate at the diaphysis in spite of massive deposition of lamellated bone. The trabeculae also are extremely thick and copious fillings in most parts of the shaft (Fig. 3). The epiphyseal cartilage is much wider than in the normal controls. At high magnification (Fig. 8) abundance of cartilage

matrix is striking; the zone of erosion has an atypical appearance. A continuous sealing-off lamina contains irregular lacunae of vascular mesenchyme which attacks the cartilage at variable levels. On the surface of the trabeculae some modeling resorption is brought about by rather small cell elements. The main activity, however, consists of endosteal bone apposition. The marrow displays a greater amount of vascular and myelogenous constituents than in hypophysectomized controls.

The articular cartilage consists of a wide layer of matrix enclosing many chondroblasts without any zonal arrangement. Most of the epiphyseal marrow spaces are replenished by hypercalcified osteochondroid tissue.

The effect of terminal administration of growth hormone to old hypophysectomized rats (Group 2B) differs from that of the previous group. Due to proliferation of the epiphyseal discs in a peripheral direction, the vertebra has gained in width (Fig. 4). The trabecular framework of the shaft, except for the juxta-epiphyseal portion, is similar to that of the hypophysectomized controls. On the surface of the shaft, periosteal activation is evidenced by a series of new incremental lamellae.

The status of the proximal epiphysis, shown at high magnification in Fig. 9, is comparable with that of a young normal animal. While the zone of proliferation is as wide as in the previous group, the zone of vesicular cells shows definite differences. Three to five layers of greatly enlarged chondrocytes are interspersed by slim spicules of matrix. Nearly every terminal cell is vigorously eroded by a capillary tuft. A wide zone of newly-formed, delicate trabecular bone containing cartilage remainders adjoins the epiphyses. Osteogenesis, judged from the abundance of osteoblasts, is in a highly active stage. Osteoclasia seems to be detained allowing the trabeculae to extend for a considerable distance into the diaphysis. The fat content of the marrow is decreased while the myelogenous components are increased correspondingly. All animals examined behave quite uniformly in this response; differences merely are restricted to the length of the newly formed trabeculae.

The articular cartilage, though greater in width and more numerous in cell content respecting the hypophysectomized controls, does not participate in the reactivation of endochondral ossification. Only few scattered cartilage cells are eroded from the epiphyseal marrow.

3. The histogenetic changes following thyroxin therapy at long postoperative intervals (Group 3) show considerable variability. One half the animals retained a status similar to the hypophysectomized controls (Fig. 5). The other half responded with wider epiphyseal discs and decreased density of trabecular bone. At high magnification reactivation

of the erosion process constitutes the prominent feature (Fig. 10). Vascular mesenchyme has broken up the sealing off bone and is eroding almost each cartilage column. An increased number of cells in the zone of proliferation evidences resumption of chondrogenesis. Osteogenesis, however, shows little stimulation; the new trabeculae are very sturdy and the old ones show many Howship's lacunae. Next to chondroclastic and osteoclastic reaction, reorganization of the hemopoietic marrow is conspicuous.

The articular cartilage responds incomparably less; some chondroblastic cell division has occurred as numerous cells are seen in the crypts of the calcified cartilage matrix. Erosion of the articular cartilage, however, does not proceed at an increased rate.

This rather optimal response to thyroxin is contrasted by pictures which are characterized by an aplastic epiphyseal growth apparatus, regeneration of the myelogenous tissue being the sole changes (Fig. 11).

4. Simultaneous injections of thyroxin and growth hormone after long postoperative periods result in a significantly greater growth in length and width of the vertebra (Fig. 6). The histologic composition resembles that seen in the growth hormone treated group (2B). Marked proliferation of the epiphyseal cartilage at the lateral peripheries is instrumental in enlarging the segment. Between the columns of chondrocytes an increased amount of hyaline matrix is observed (Fig. 12). There is great irregularity in the zonal arrangement as some chondroblastic rows retain an embryonic cell character and do not undergo cytomorphic changes. The high vitality of the cartilage is also evident from the zone of erosion where exceedingly large chondroclasts are operative in breaking up the crypts of the enlarged but not vacuolated cartilage cells. Bone formation proceeds at a very fast rate so that the trabeculae fuse immediately to huge masses of spongy bone extending deep into the shaft. Lacunae containing large osteoclasts indicate vigorous processes of modeling resorption. The marrow consists for the main part of vascular and hemopoietic elements.

The articular cartilage is equally rich in matrix but endochondral ossification occurs only sporadically. Endosteal bone formation is very active so that compact bone has replaced the chondrosteoid tissue at the epiphyseal head. There is evidence of continuous reorganization of the spongy bone as well as the compact bone of the shaft resulting from increased activity of Haversian blood vessels.

5. Chronic administration of growth hormone to normal adult rats (Group 5) results in a proportionately oversized vertebra with correspondingly denser trabecular and cortical bone (Fig. 7). At both epi-

physeal heads cancellous bone has reduced the marrow spaces. The epiphyseal cartilage plate shows signs of continued growth activity (Fig. 13). Though not wider in size than normal, it contains an increased amount of flattened cells aligned in rows. The basophilia of the abundant matrix in the zone of enlarged cells is not as marked as in normal controls but degenerate areas are equally frequent. The zone of erosion looks similar to that of the chronically treated growth hormone group (2A). The newly formed trabeculae fuse laterally so that a honey-combed lamina adjoins the cartilage. Its exact boundary cannot be determined as processes extend deep into the trabecular bone. The marrow contains more myelogenous components than the normal controls.

The articular cartilage is reduced to a narrow rim of calcified matrix lined by a cellular perichondrium.

Conclusions and summary

In order to determine the mode of reaction of the caudal vertebrae of the rat of the Long-Evans strain to hormonal therapy, 41 hypophysectomized animals were injected with 39 doses of either pure pituitary growth hormone or thyroxin or the combination of both after long postoperative intervals. Another group of hypophysectomized rats received shortly after operation 437 injections of pure growth hormone. The response of normal adults to chronic administration of growth hormone also was analyzed metrically and histologically. It was found that the caudal vertebrae grossly follow the pattern established for the tibia.

Growth hormone reactivated or stimulated chondrogenesis and osteogenesis while it had little effects upon cartilage erosion and bone resorption. The epiphyseal cartilage responded more intensively than the articular cartilage. Growth hormone injected chronically either to hypophysectomized animals or normal adults, however, was unable to prevent fibrotic degeneration of cartilage matrix.

Thyroxin did not check the stunting effect of hypophysectomy; also, it engendered some reactivation of cartilage erosion through marrow restoration and vascularization. Little cartilage proliferation took place.

Concurrent administration of both thyroxin and growth hormone had optimal effects on the rate of growth as well as repair of the postoperative bone pathology. Thirty-nine combined doses administered to old hypophysectomized animals almost paralleled the growth spurt elicited by 437 injections of growth hormone alone to young hypophysectomized animals. Histologically it restored the equilibrium between chondral and osteal proliferation, and resorption respectively. A status comparable to

that of very young normal controls was brought about. Again it was noticed that only the epiphyseal cartilage profited from this hormonal synergism. The articular cartilage showed antagonism as its width was decreased relative to that of the growth hormone treated groups according to the pattern already described in the tibia and condyle of the mandible.

The above results suggest that the genetic pattern of cartilage activity determines the mode of reaction to hormonal stimulation. Normally persisting cartilages respond to combination therapy with equally increased rates of proliferation and erosion in the sense of a synergism. In genetically exhausted or overdue cartilages, erosion is more activated by thyroxin than chondrogenesis is stimulated by growth hormone, thus effecting an antagonism of both hormones.

Easy accessability, simplicity and stability in the structural arrangement, absence of functional interference, possibility of a complete morphogenic survey, sensitive reactivity to endocrine conditions make the caudal vertebrae useful test bones in endocrine experiments.

Zusammenfassung

Zwecks experimenteller Erforschung der hormonalen Beeinflussung der Skelettmorphogenese am Beispiel des Rattenschwanzes wurden in dieser dritten Serie einer Untersuchungsreihe 41 Tiere, welche die Hypophysektomie am 28. Lebenstage während 300 bis 400 Tagen überlebt hatten, mit 39 täglichen Dosen entweder von reinem hypophysärem Wachstumshormon oder von Thyroxin oder der Kombination von beiden Hormonen behandelt. Eine andere Gruppe von weiblichen Ratten erhielt einige Tage nach der Hypophysektomie während 437 Tagen Injektionen von reinem Wachstumshormon. In gleicher Weise wurde die Reaktion normaler erwachsener Tiere auf die chronische Abgabe von 437 Injektionen von Wachstumshormon metrisch und histologisch geprüft (siehe Versuchsanordnung Tabelle 1).

Es zeigte sich, daß sich die Schwanzwirbel im allgemeinen ähnlich verhielten, wie das früher bereits für die Tibia festgelegt worden ist, wobei immerhin Unterschiede in der Knochentransformation wohl infolge Fehlens einer namhaften Funktion in Erscheinung traten.

Das Wachstumshormon kontrolliert das Längenwachstum über die Mechanismen der Chondrogenese und Osteogenese, während es auf die Knochentransformation, also Knorpel- und Knochenresorption, wenig Einfluß hat. Die Epiphysenscheiben reagieren dabei intensiver als die Gelenkknorpel. Fibrotische Altersdegeneration des Knorpels wird durch Wachstumshormon eher beschleunigt.

Thyroxin ist nicht imstande die wachstumshemmende Wirkung der Hypophysektomie zu beheben. Dagegen vermag es Knochenreifung, also Knorpelerosion und modellierende Knochenresorption wieder in Gang zu bringen, während es die Chondrogenese nur wenig aktiviert.

Kombinierte Therapie mit Wachstumshormon plus Thyroxin zeitigt optimale Wirkung sowohl bezüglich der Wiederherstellung der Morphogenese wie der Maturation. Mit 39 terminalen Kombinationsdosen an alte hypophysektomierte Tiere wurde fast die gleiche Skelettlänge erreicht wie mit chronischen Injektionen von 437 Dosen von Wachstumshormon allein an junge hypophysektomierte Tiere. Histologisch konnte eine Wiederherstellung des Gleichgewichtes zwischen Knorpel- und Knochenproliferation einerseits und ihrer Transformation anderseits festgestellt werden.

Die histologischen Zustandsbilder waren vergleichbar mit denen von jungen normalen Tieren. Wiederum zeigte es sich, daß hauptsächlich die Knorpelscheiben auf Kombinationstherapie im Sinne eines Synergismus reagieren, während die Gelenkknorpel eher antagonistisch antworten, indem deren Durchmesser nach Kombinationstherapie viel kleiner war als nach alleiniger Wachstumshormoninjektion. Eine gleiche antagonistische Wirkung der Kombinationstherapie wurde auch in den Gelenkknorpeln der Tibia und besonders in den Kondylen des Unterkiefers beobachtet.

Die Summe der metrischen und histologischen Ergebnisse scheint dahin zu deuten, daß letzten Endes die Erbanlage über die Reaktion einzelner Knochen auf die hormonale Beeinflussung entscheidet. Sämtliche junge wie auch alte, physiologischerweise persistierende Epiphysenscheiben reagieren auf die Kombinationstherapie mit gleichzeitig verstärkter Proliferation und Transformation im Sinne eines Synergismus von Wachstumshormon und Thyroxin. In den Gelenkknorpeln und genetisch überfälligen Wuchsknorpeln dagegen hat die Maturationswirkung des Thyroxins ein Übergewicht über den wachstumsfördernden Einfluß des Wachstumshormons, was einem Antagonismus beider Hormone gleichkommt.

Vom experimentellen Standpunkte aus erweisen sich die Schwanzwirbel der Ratte infolge ihrer leichten Zugänglichkeit für Biopsie wie Nekropsie, dank ihrer strukturellen Einfachheit und Stabilität infolge Fehlens wesentlicher Funktionseinflüsse und dank der Abwicklung sämtlicher morphogenetischer Vorgänge in der postnatalen Phase sowie infolge ihrer großen Ansprechbarkeit auf hormonale Stimuli als ausgezeichnete Testknochen für endokrine Experimente.

Résumé

Dans cette 3e série d'investigations concernant le contrôle endocrinien de la morphogenèse osseuse, 41 rats femelles hypophysectomisés à l'âge de 28 jours ont été injectés avec 39 doses (journalières), soit d'hormone de croissance, de thyroxine ou la combinaison des deux hormones après de longs intervalles post-opératoires, afin de pouvoir étudier le comportement de leurs vertèbres caudales. Un autre groupe de rats hypophysectomisés ont reçu, quelques jours après l'opération, 437 doses d'hormone de croissance pure. La réaction d'adultes normaux à l'administration chronique de l'hormone de croissance fut également analysée métriquement et histologiquement.

Il a été trouvé que la réaction des vertèbres caudales aux traitements hormonaux ressemble à celle qui a été établie pour le tibia; cependant, la transformation et la maturation osseuses furent modifiées.

L'hormone de croissance stimule les processus morphogénétiques, soit par la chondrogenèse, soit par l'osteogenèse; elle n'a que peu d'effet sur la maturation, c'est-à-dire sur l'érosion du cartilage ou la résorption de l'os. Le cartilage épiphysaire répond plus intensément que le cartilage articulaire. L'hormone de croissance injectée chroniquement, soit à des animaux hypophysectomisés, soit à des adultes normaux, n'empêche cependant pas la dégénérescence fibrotique de la matrice du cartilage.

La thyroxine n'est pas capable d'influencer la restauration de la croissance déficiente de l'os provoquée par l'hypophysectomie; elle n'a qu'un faible effet stimulant sur la chondrogenèse, mais rétablit l'érosion du cartilage et de l'os par une vascularisation de la moelle.

L'administration concurrente de thyroxine et d'hormone de croissance a des effets optima sur la rapidité de croissance aussi bien que sur la réparation des dégâts causés par l'hypophysectomie. 39 doses de thyroxine combinée à l'hormone de croissance ont été administrées à des animaux survivants, à de longs intervalles post-hypophysaires; on a pu constater que l'action de ces 2 hormones sur la croissance est approximativement la même que celle qui est conditionnée par 437 injections d'hormone de croissance seule chez des animaux récemment hypophysectomisés. Du point de vue histologique, elle rétablit l'équilibre entre la prolifération chondrale et ostéale, d'une part, et la résorption modelante, d'autre part. L'aspect histologique ressemble à celui de jeunes contrôles normaux. Il fut observé, comme par le passé, que le cartilage épiphysaire seul profite de cette synergie hormonale.

Les résultats obtenus montrent donc que des facteurs génétiques sont ultérieurement responsables du mode de réaction des différents cartilages à une stimulation hormonale. Les jeunes cartilages aussi bien que

les cartilages physiologiquement persistants répondent aux traitements hormonaux de thyroxine combinée à l'hormone de croissance, avec une égale augmentation de la prolifération et de l'érosion dans le sens de synergie.

Les vieux cartilages épiphysaires qui génétiquement devraient disparaître et les cartilages articulaires réagissent plus intensément à la thyroxine provoquant leur érosion qu'à l'hormone de croissance stimulant leur chondrogenèse. Il en résulte un antagonisme des deux hormones.

Du point de vue expérimental, la commodité d'accès, la simplicité ainsi que la stabilité dans l'arrangement structural, l'absence d'interférence fonctionnelle, la possibilité d'un aperçu morphogénique complet, ainsi que la réaction sensible à des conditions endocriniennes rendent les vertèbres caudales des os favorables à l'examen biologique des fractions hormonales.

Riassunto

Allo scopo di studiare sperimentalmente l'influsso degli ormoni sulla morfogenesi dello scheletro, prendendo come esempio la coda del ratto, vennero trattati 41 animali, facenti parte della terza serie di un seguito di esperienze, e che avevano sopravvissuto per 300–400 giorni all'ipofisectomia eseguita 28 giorni dopo la nascita, con 39 dosi giornaliere di ormone ipofisario di crescita puro, o di tirossina, o dei due insieme. Un altro gruppo di ratti femmina ricevette alcuni giorni dopo l'ipofisectomia e per 437 giorni iniezioni di ormone di crescita puro. Similmente venne studiata la reazione di animali adulti normali alla somministrazione prolungata di 437 iniezioni di ormone di crescita (vedere Tabella 1).

Risultò che le vertebre della coda si comportarono in modo analogo a quanto fu in precedenza osservato a proposito della tibia, ma con differenze nella trasformazione ossea certamente in seguito alla mancanza di un importante elemento funzionale. L'ormone di crescita controlla, mediante i meccanismi di condrogenesi ed osteogenesi, la crescita in lunghezza, mentre non ha alcun influsso sulla trasformazione dell'osso, cioé sul riassorbimento della cartilagine e dell'osso. I dischi epifisari reagiscono in questo processo più intensamente che le cartilagini articolari. La degenerazione fibrosa senile della cartilagine viene piuttosto accellerata dall'ormone della crescita.

La tirossina non è in grado di sopprimere l'inibizione della crescita dovuta alla ipofisectomia. Al contrario favorisce la maturazione dell'osso, cioè l'erosione della cartilagine ed il riassorbimento plastico dell'osso, mentre non attiva che in misura ridotta la condrogenesi.

La terapia combinata con ormone di crescita e tirossina dimostrò di

avere un'azione ottimale tanto in rapporto alla restituzione della morfogenesi, quanto in rapporto alla maturazione. Mediante 39 dosi terminali combinate, somministrate ad animali anziani ipofisectomizzati, si ottenne quasi la stessa lunghezza scheletrica che si ebbe mediante terapia prolungata con 437 dosi di ormone di crescita in animali giovanni ipofisectomizzati.

I quadri istologici erano paragonabili e quelli presentati da animali giovani normali. Si constatò di nuovo che l'azione sinergica della terapia associata si manifesta principalmente sul disco epifisario cartilagineo, mentre nell'azione sulle cartilagini articolari é evidente un antagonismo, in quanto i diametri delle stesse dopo terapia combinata erano molto più piccoli di quelli in cui l'ormone di crescita era stato impiegato solo. Un'uguale azione antagonista della terapia combinata fu osservata anche nelle cartilagini articolari della tibia e specialmente nei condili della mandibola.

L'insieme dei risultati delle ricerche metriche ed istologiche sembra deporre per il fatto che, in definitiva, é la situazione ereditaria che é determinante nella risposta delle singole ossa all'azione ormonica. Tutti i dischi epifisari giovani, come pure quelli vecchi fisiologicamente persistenti reagiscono alla terapia combinata contemporaneamente con accentuata proliferazione e trasformazione nel senso di un sinergismo tra ormone di crescita e tirossina. Invece nelle cartilagini articolari e nelle cartilagini di crescita che da un punto di vista genetico sono superflue, predomina l'azione maturante della tirossina sullo stimolo alla crescita dovuto all'ormone ipofisario, ciò che corrisponde ad un antagonismo tra i due ormoni.

Dal punto di vista sperimentale, le vertebre della coda del ratto si rivelano come adeguato materiale di studio per ricerche ormoniche in quanto facilmente accessibili al controllo necroscopico e bioptico, sia ancora per la semplicità e stabilità della loro struttura dovute all'assenza di fattori modificanti funzionali e grazie allo svolgimento di tutti i processi morfogenetici nella fase post-natale, sia infine in conseguenza della loro grande sensibilità a stimoli ormonali.

1. *Asling, C. W.*, and *Evans, H. M.:* Anterior Pituitary Regulation of Skeletal Development, in Bourne, G. H.: Biochemistry and Physiology of Bone. Academic Press, N. Y., 1956. – 2. *Asling, C. W., Simpson, M. E.*, and *Evans, H. M.:* Effects of Pituitary Factors and of Thyroxin on Skeletal Morphogenesis in the Rat, in *Reifenstein, E. C.:* Metabolic Interrelations, Josiah Macy, jr., Fondation, N. Y., 1951. – 3. *Baume, L. J., Becks, H.*, and *Evans, H. M.:* Helv. odont. Acta **1**, 9 (1957). – 4. *Baume, L. J., Becks, H.*, and *Evans, H. M.:* Helv. odont. Acta **1**, 28 (1957). – 5. *Baume, L. J., Becks, H.*, and *Evans, H. M.:* Helv. odont. Acta (in press). – 6. *Becks, H., Asling, C. W., Simpson, M. E., Evans, H. M.*, and *Li, C. H.:* Amer. J. Anat. **82**, 203 (1948). – 7. *Becks, H., Asling, C. W.*

Collins, D. A., Simpson, M. E., Li, C. H., and *Evans, H. M.:* Anat. Rec. **101,** 17 (1948). – 8. *Becks, H., Collins, D. A., Simpson, M. E.,* and *Evans, H. M.:* Amer. J. Orthodont. and Oral Surg. **32,** 446 (1946). – 9. *Becks, H., Simpson, M. E., Evans, H. M., Ray, R. D., Li, C. H.,* and *Asling, C. W.:* Anat. Rec. **94,** 631 (1946). – 10. *Evans, H. M., Becks, H., Asling, C. W., Simpson, M. E.,* and *Li, C. H.:* Growth **12,** 43 (1948). – 11. *Evans, H. M., Simpson, M. E.,* and *Li., C. H.:* Growth **12,** 15 (1948). – 12. *Simpson, M. E., Evans, H. M.,* and *Li, C. H.:* Growth **13,** 131 (1949).