with strain, sex, season or age, characteristics which often vary from one experiment to another[14].

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Résumé. Chez des rats mâles anésthésiés à l'éther avant la décapitation, on a observé une augmentation du niveau sérique de LH et de FSH, sans effet sur le niveau de testostérone. L'intervention chirurgicale subie 2 jours avant le sacrifice a fait diminuer seulement le niveau de LH. Les niveaux de LH, de FSH et de testostérone ont également diminué après 2 jours d'absence de nourriture.

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Absence of Sulphation Factor (Somatomedin) Activity in Preparations of Colony Stimulating Factor and Nerve Growth Factor

The term somatomedin (SM) has been proposed for a factor or group of factors in serum which are growth hormone dependent and which stimulate the in vitro incorporation of ^{35}S into cartilage proteoglycan[1]. The in vitro effects of SM, which are not species specific[2], do not appear to be limited to cartilage as partially purified preparations have also been shown to have insulin-like activity[3]. In addition, recent reports indicate that SM stimulates the growth of several cell-lines in culture[4–6].

As it was of interest to learn whether other growth factors present in serum shared some of the biological effects of SM, we have studied the effects of mouse nerve growth factor (NGF) (Wellcome Laboratories; K5740) and colony stimulating factor (CSF) on the in vitro incorporation of ^{35}S into embryonic chick catilage.

Colony stimulating factor was prepared from pregnant mouse uterus by the method of Bradley[7]. Colony stimulating activity was assayed by a modification of the method of Bradley and Metcalf[8] using mouse bone marrow culture in semi-solid agar. Equal volumes of 0.6% agar and double-strength modified Eagle's medium were mixed with mouse bone marrow cells to give a final cell-count of 2×10^5/ml. 1 ml aliquots of this mixture were added to plastic petri dishes containing test material. The number of colonies were counted after 7 days incubation at 37 °C in an atmosphere of 5% CO_2-95% air. Observations were made in duplicate. Approximately 200 colonies were formed when 0.05 ml of the CSF extract was used.

Proteoglycan synthesis was studied by incubating pelvic cartilages from 11-day chick embryos with ^{35}S for 18 h at 37 °C. Six cartilages, incubated in groups of three in 1 ml medium, were used for each observation. The medium contained dilute human serum in some experiments. Further details of the method are published elsewhere[9].

We found that addition of 0.1 ml of the preparation of mouse CSF had no significant effect on the incorporation of ^{35}S into chick pelvis, either in the absence of serum or in the presence of 2.5% or 40% serum (Table I). Similarly, NGF at concentrations of 10 U/ml–1000 U/ml had no effect on the incorporation of ^{35}S into cartilage (Table II). In addition, NGF at concentrations of up to 1000 U/ml did not stimulate colony formation by mouse bone-marrow cells in agar culture or inhibit the activity of CSF (Table III).

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Table I. Effect of colony stimulating factor prepared from mouse uterus on ^{35}S incorporation into chick pelvic cartilage

Experiment	Test sample	^{35}S incorporation ± SEM (cpm × 10^{-3}/mg cartilage)		
		0 [a]	10% [a]	40% [a]
1	0.1 ml CSF	5.7 ± 0.5	—	—
	Control	9.0 ± 1.4	—	—
2	0.1 ml CSF	—	23.7 ± 0.9	37.1 ± 1.8
	Control	—	23.9 ± 2.1	35.9 ± 3.8

Six cartilages were used for each observation. Incubation volume = 1 ml. [a] Serum concentration in medium.

Table II. Effect of mouse nerve growth factor on ^{35}S incorporation into chick pelvic cartilage

Experiment	Test sample	^{35}S incorporation ± SEM (cpm × 10^{-3}/mg cartilage) 0 [a]	2.5% [a]	20% [a]
1	0.1 ml NGF (100 U/ml)	8.7 ± 0.8	—	—
	Control	9.0 ± 1.4	—	—
3	0.1 ml NGF (100 U/ml)	—	58.8 ± 3.7	84.9 ± 3.5
	0.1 ml NGF (500 U/ml)	—	47.9 ± 3.7	81.6 ± 4.3
	Control	—	50.8 ± 1.1	79.7 ± 3.4
4	0.1 ml NGF (10,000 U/ml)	—	33.8 ± 2.2	57.3 ± 2.2
	Control	—	32.6 ± 2.9	59.0 ± 1.1

Six cartilages were used for each observation. Incubation volume = 1 ml.
[a] Serum concentration in medium.

These results indicate that neither CSF obtained from mouse uterus nor NGF prepared from mouse salivary glands stimulate proteoglycan synthesis in chick embryo cartilage, even when used at concentrations well in excess of those which are effective in promoting colony formation or neural growth in vitro. In addition, they indicate that mouse NGF has no appreciable colony stimulating activity. The findings are in agreement with the observation by METCALF et al.[10] that CSF had no effect on the growth of fibroblasts and several other cell-lines in culture. It therefore appears that neither NGF nor CSF have the more general biological effects which have been attributed to SM.

Table III. Effect of mouse nerve growth factor on colony formation by mouse bone-marrow cells

Test sample	No. of colonies
0.1 ml NGF (625 U/ml)	0,4
0.1 ml NGF (2500 U/ml)	0,0
0.1 ml NGF (10,000 U/ml)	0,4
0.1 ml H_2O	0,2
0.05 ml CSF	221,202
0.05 ml CSF + 0.05 ml NGF (5000 U/ml)	201,208

Results obtained with mouse colony stimulating factor are also shown.

Résumé. Le facteur nerveux de croissance des souris (mouse nerve growth factor: NGF) et le «colony stimulating factor» (CSF) furent testés pour contrôler l'activité de la somatomédine, en mesurant l'incorporation du S^{35} dans du cartilage d'embryon de poulet. Aucune de ces deux préparations ne stimulait cette incorporation. De plus, le NGF n'avait pas d'effet sur la formation des colonies de moelle osseuse. Ces découvertes indiquent que ni le NGF ni le CSF ne participent aux effets biologiques généraux attribués à la somatomédine.

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Plasma Thyrotrophin Concentration in the Foetal Calf

Previous observations on thyroxine levels in foetal calves with indwelling vascular catheters have shown that total plasma thyroxine falls in the last 3 days of intrauterine life, while the concentration of total free thyroxine does not change significantly[1]. These findings suggest that the pituitary-thyroid axis adjusts the level of secretion of thyroxine to maintain a constant circulating concentration of free thyroxine. The present paper reports the foetal plasma thyrotrophin (TSH) concentrations in the same 7 animals, and the changes in TSH, cortisol and thyroxine which occurred when parturition was induced with Synacthen.

Materials and methods. Plasma samples were obtained from chronically catheterized foetuses during the last 15 days of gestation. The calves were born between 261 and 283 days gestation (term 280 days)[2]. Premature parturition was induced in 2 cows by the administration of 0.06–0.125 mg Synacthen β 1–24 Corticotrophin (CIBA) 4 times daily i.m. to the foetus. In a 3rd cow the foetus was given cortisol (Efcortelan, Glaxo, i.v., 4 times daily) to initiate delivery. Gestational age was between 250 and 260 days in these induced pregnancies.

Plasma TSH was measured in a double antibody radioimmunoassay. The coefficient of variation of the assay was 7.4% and the recovery of added TSH from calf plasma was 96.1% over the range 1.25–25 μU/ml. Cross

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