(spheroidal, ovoidal, piriform etc...) and cell size (from 530 μm^3 to 56,000 μm^3).

In the ganglia the largest cells are generally located in the periphery, the smaller in a more internal layer; nerve fibers run through the center of the ganglion. The small internal cells have 1 or 2 nucleoli, where the peripheral largest cells have a nucleolar number ranging from 1 to 20. In this last case the total nucleolar volume varies greatly and the nucleolar volume of a large cell can be less than in a small one; it was long ago described that the state of functional activity is a factor of importance in regulating the size of the nucleolus in neurons[12,13].

Some of the nucleoli are small (less than 1 μm) but 1 or 2 nucleoli are large (4 μm diameter). In Toluidine blue or Feulgen-light green preparation, these larger nucleoli appear like a ring (Figure 3) thus suggesting that the nucleolus has the shape of a cup. The internal and external surfaces of the cup are lined with Feulgen material, i.e. the nucleolus associated chromatin.

In the concavity of the cup there are one (Figures 1 and 2) or more (Figure 4) refringent bodies. A quite similar morphology of the nucleolus can also be clearly seen in the living cells (Figure 5).

In the Feulgen preparations the chromatin externally associated with the nucleolus is encircled, at a distance of 1.5–2 μm, by a ring of chromatin granules. The clear circular area between this ring and the chromatin externally associated with the nucleolus is crossed by tiny heterochromatic filaments. The same structure is present in the smaller more internal ganglion cells, but the number of granules in the ring is scanty.

Discussion. The pattern of the chromatin around the nucleolus recalls the DNA body of the oocytes of insects[1–5], because in this case also the nucleolus lies inside the body but we should stress that: 1. tiny filaments cross the clear area between the chromatin externally associated with the nucleolus and the DNA body which could indicate that each one of the granules of the DNA body belongs to the same part of the genonema bearing the nucleolus associated chromatin; 2. in the nervous cells, i.e. in highly specialized somatic cells, the largest peripheral cells of the ganglion have more granules of chromatin in the DNA body than the smaller, more internal ones; thus the increase in the cell size is accompanied by an increase of the DNA body. Then the behaviour of the DNA body of the nervous cells of *Biomphalaria* is different from that of the oogonia where the DNA body after being synthesized in the interphase is lost in the late diplotene.

It is possible to suppose that in the oocytes the influence of the cytoplasm on the genome takes a different pattern with the biochemical changes at the diplotene stage, whereas the constant pattern of the nucleocytoplasmic interrelationships in the differentiated cells induces as a consequence the permanence of the DNA body. The quantitative increase of activity could account for the enlargement of the DNA body: the paranucleolar irregular heterochromatic structure, associated with the *Y* chromosome in mammalian neurons, enlarges its area after stimulation and in other experimental conditions[6].

It is the goal of further experiments to see if the DNA body is under the control of endogenous or exogenous factors and if there is some type of correlation between the DNA body and the nucleolus associated chromatin for the production of the nucleolar material.

Résumé. Dans les cellules nerveuses de *B. glabrata* on observe un anneau de chromatine, «DNA body», qui entoure le nucléole; entre le «DNA body» et la chromatine attenante à la paroi externe du nucléole existe un anneau clair parcouru par de très fins filaments. Le «DNA body» des cellules nerveuses rapelle une formation similaire, mais transitoire, présente dans les oocytes d'autres Invertébrés.

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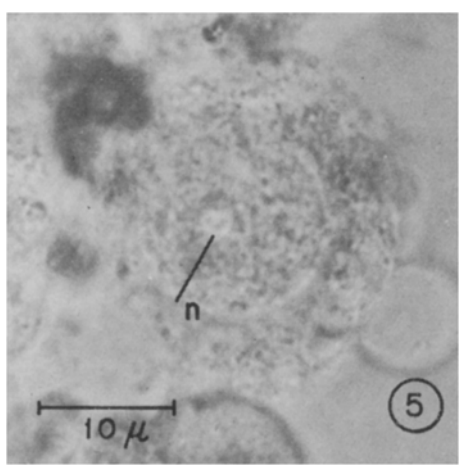

Fig. 5. A cup shaped nucleolus (n) in a living cervical ganglion cell. Phase contrast. UV Filter III. ×2,000.

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Interferon Production by Sendai Virus-Treated Hamster and Monkey Cells

By using SV40-transformed cells of Cl_2TSV_5 and RHaT lines[1], several generations of tumors produced in Syrian hamsters were studied[2]. Tumor cells were then fused with SV40-permissive monkey cells to study the rescuability of SV40; UV-light-inactivated Sendai virus (UV-SeV) was used as fusing agent. Since UV-SeV may induce interferon which may inhibit or reduce the rescuability of SV40 from fused cell cultures[3,4], a study was undertaken to test tumor and non-transformed cells for interferon production.

Experimental. Cl_2TSV_5 and RHaT cells, the SV40-permissive monkey cells (AGMK and BSC), the hamster primary kidney (HK) cell cultures, and the PK-15

(CCL22) pig kidney cells, were all grown as described previously[1]. Details on the production of tumor generations and their cell cultures are given elsewhere[2]. Because of its simplicity, reliability and reproducibility[5], the procedure chosen for interferon assay was that of inhibition of viral cytopathic effect (CPE) of SELLERS and FITZPATRICK[6]; the 2 viruses used for this test were SV40 (strain rescued from RHaT) and vesicular stomatitis (VSV, a stock strain from this laboratory).

The UV-SeV-treated cell cultures and the different combinations tested for interferon production are given in the Table. Cl_2TSV_5 and RHaT cultures used for these studies were derived from the tumors at the 3rd and 4th in vivo passage levels, respectively. 2 identical cultures of each cell mixture or cell type were prepared and incubated at 37 °C for 48 h without changing the medium which contained 0.5% of heat-inactivated rabbit anti-SV40 serum. Medium harvested from these cultures after 24 and 48 h was treated as if it were interferon[4, 6] and applied to cultures of AGMK, BSC and HK cells. 20 h later, these cultures were rinsed twice with Eagle's medium (MEM) and challenged with VSV or SV40.

Observations. For interferon assay, AGMK cells proved to be about 25% more sensitive than the BSC cells to the interferon preparation from the same source (either AGMK or BSC). Both cell cultures (AGMK and BSC) appeared to show the same sensitivity to SV40 and VSV, and both these viruses appeared to be equally sensitive to the interferon. Also these was no significant difference in the sensitivity to VSV between hamster normal and tumor cells.

As shown in the Table, all fluids from cultures containing AGMK or BSC cells treated with UV-SeV showed virus inhibitory activity. Culture fluids, from UV-SeV-treated monkey cell cultures, harvested at 24 and 48 h, did not show any significant difference in interferon titer when tested on AGMK cells. Culture fluids from UV-SeV-treated BSC cultures did exhibit interferon activity which was about 25% less than that of AGMK cultures under the same conditions.

The Table also indicates that UV-SeV-treated Cl_2TSV_5 and RHaT cultures did not produce interferon or, if produced, it was not present in the culture fluid at a detectable concentration. Another possible explanation might be that the HK cells used in the experiment did not constitute the best system for the detection of small amounts of interferon. However, interferon was detected in hamster kidney cell cultures treated with UV-SeV. This interferon protected HK cells against VSV (Table). It did not, however, protect AGMK cells against the same virus (VSV). Treatment of HK cells with an undiluted interferon preparation from AGMK or BSC cultures did not protect HK cells against VSV infection; in contrast, AGMK and BSC cells treated with this same preparation we protected against VSV or SV40 infection.

Thus, the species specificity, stability at pH 2.0, lack of toxicity and the lack of virus specificity of the anti-viral substance present in the tested media indicated that this substance has the characteristics of interferon[7].

Discussion. The absence of detectable interferon in hamster tumor cell cultures suggests that the SV40-genome present in those cells might interfere with the mechanism responsible for interferon synthesis. It has been suggested that decreased interferon production is an early consequence of infection by oncogenic viruses, and a property which persists in many lines of transformed cells[8]. It would be worth while to see whether a correlation does or does not exist between decreased interferon production and the degree of oncogenicity of a transformed cell population. It is interesting to note that, although the above hamster tumor cells do not produce interferon, they are as sensitive to VSV as the normal (HK) cells.

Interferon might make more difficult the rescue of virus from fused cultures possibly by 2 mechanisms: first, by its production and anti-viral action in fused cells and, second, by its action on the next generation(s) of non-fused permissive cells present in the culture of Sendai virus-treated cell mixture. The present study suggests that no

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Interferon production by Sendai virus-treated hamster and monkey cells

Cell types and combinations	Sendai Virus[a]	Types of interferon detected		Interferon titers[b]
		Hamster	Monkey	
1 Cl_2TSV_5 + AGMK	+	0	+	1:32
2 CL_2TSV_5 + BSC	+	0	+	1:24
3 RHaT + AGMK	+	0	+	1:32
4 RHaT + BSC	+	0	+	1:24
5 Cl_2TSV_5	+	0	0	0
6 RHaT	+	0	0	0
7 AGMK	+	0	+	1:64
8 BSC	+	0	+	1:48
9 HK	+	+	0	1:16
10 Co-cultivated and other controls[c]	0	0	0	0

[a] 4,000 hemagglutinating units of UV-SeV were (+) per 1×10^7 cells[1]. [b] The titers given here were obtained by graphical interpolation, and represent the final dilutions of the interferon preparations at which there would have been 50% protection of the cell sheets. [c] Cells were co-cultivated according to the combinations given in this Table in 1–4. Other controls referred to here represent cultures as indicated above in 5–9, but without Sendai virus.

interferon is produced in UV-SeV-treated monkey cell cultures after 24 h. Therefore, it seems very important to rinse these cultures 24 h after cell fusion to discard the interferon present in the culture fluid. Thus, the recovery of virus from UV-SeV-treated cultures may not be significantly affected, because it appears that, in most cases, the cellular resistance to the virus declines within a relatively short period after the removal of the interferon or its inducer[9]. The present study also suggests that in systems such as ours, the lack of detectable interferon in tumor cell cultures perfectly agrees with the successful rescue of SV40-genome from all these cultures[2].

Finally, from the results described above with the virus-cell systems used in the present work, it becomes clear that no correlation exists between the ability of a culture to produce interferon and the sensitivity of its cells to the virus infection.

Résumé. On présente une étude comparée sur la production d'interféron par les cellules non-transformées et tumorales de hamster ainsi que par les cellules de singe, après leur traîtement par le virus Sendai inactivé. Les cellules tumorales ne produisent pas l'interféron sous l'action du virus Sendai. Les cellules non-transformées cessent de produire ou de libérer l'interféron, dans leur milieu de culture, en moins de 24 h après l'enlèvement du virus Sendai de ce milieu.

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Über das Vorkommen von Hämagglutininen (Anti-H und Anti-B) beim Flussbarsch (*Perca fluviatilis*)

Die vor allem im Genitalapparat mehrerer Schnecken und in den Eiern verschiedener Fische aufzufindenden Agglutinine wurden von Prokop, Uhlenbruck und Köhler[1] als Protektine zusammengefasst, weil sie eine immunbiologische Schutzfunktion dieser antikörperähnlichen Verbindungen für die Eier angenommen haben.

Aus den weiblichen Gonaden des Barsches *Perca fluviatilis* wurde 1967 ein Hämagglutinin, Anti-H (Titer gegen 0-Erythrocyten etwa 1:256) beschrieben[2]. So weit uns bekannt, wurden nur Gonaden untersucht. Wir sezierten 5 Barsche (Anfang April gekauft, 3 Männchen und 2 Weibchen, Länge 18 bis 21 cm, alle hatten vollreife Gonaden) und untersuchten die einzelnen Bestandteile auf Agglutininaktivität gegen menschliche A_1, B und O-Erythrocyten (Methode nach[3]). Die Ergebnisse sind in der Tabelle wiedergegeben.

Unerwartet stark war die Anti-H-Aktivität der weiblichen Gonaden – sie hängt sichtlich von der Jahreszeit und dem Reifezustand der Gonaden ab (die in [2] untersuchten Tiere wurden Anfang Februar, unsere Anfang Aprilgefangen). Das Ausbleiben einer Reaktion bei den männlichen Gonaden entspricht den Befunden bei Forellen[4]. Das Anti-B aus dem Blut steht offensichtlich in keinem Zusammenhang mit dem Anti-H aus der weiblichen Gonade – es handelt sich hier sicherlich um zwei verschiedene Systeme.

Die an Hand der Gonaden gewonnenen Ergebnisse sprechen zwar für die bereits angenommene Eischutzfunktion des Anti-H aus der weiblichen Gonade[1] – bewiesen ist sie damit aber noch nicht. Alle in Schnecken zu findenden Agglutinine sind z.B. sicher nicht nur als Eischutzstoffe anzusehen[3]. Falls diese Hämagglutinine überhaupt eine biologische Bedeutung für die Tiere haben (Agglutininaktivität «rein zufällig»?), könnte sie auch auf einem anderen Gebiet liegen. Vor kurzem wurde diskutiert, ob es sich bei dem Agglutinin aus der Weinbergschnecke *Helix pomatia* nicht auch um einen Teil eines «Transportmechanismus» handeln könnte[5].

Summary. Perches (*Perca fluviatilis*) were dissected and the parts tested for agglutinin activity. Only in the female gonads a very strong anti-H could be found. Its strength is certainly subjected to seasonal variations. Anti-B was found in the blood. There is obviously no connection between the anti-H in the female gonads and the anti-B out of the blood.

Vorkommen von Hämagglutininen bei *Perca fluviatilis*

Untersuchte Bestandteile	Agglutininaktivität
Gonaden: weiblich	sehr stark Anti-H (Titer gegen 0-Erythrocyten 1:16000 bis 32000, B-Erythrocyten 1:2000 und A_1-Erythrocyten 1:16 bis 32)
männlich	keine
Laterale Körperwandung	keine
Blut	deutlich Anti-B (Titer gegen B-Erythrocyten 1:4 bis 32)
Eingeweidetrakt Niere Kopfniere	überwiegend schwach Anti-B

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