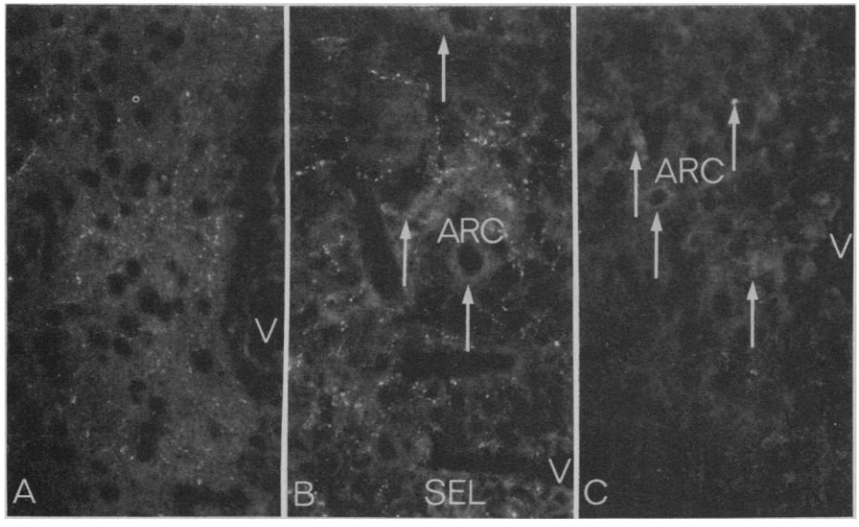

Fig. 1. Immunofluorescence micrographs of the rat dorsomedial hypothalamic nucleus (A) and of the rat arcuate nucleus (*ARC*) with the adjacent parts of the subependymal layer (*SEL*) of the median eminence (B). A plexus of prolactin-positive nerve terminals of medium density is seen in both pictures. Prolactin-positive terminals are present in the subependymal layer. In (C) the section from the arcuate nucleus has been incubated with prolactin antiserum that had first been treated with rat prolactin; no prolactin-positive terminals are present. Arrows point to nonspecific fluorescence; *V*, third ventricle (× 230).

somatotropin (supplied by NIAMDD). None of these antibodies cross-react with the other adenohypophyseal hormones mentioned above.

In the anterior and particularly the posterior periventricular region of the hypothalamus and the preoptic area, networks of varicose, fiber-like structures exhibiting prolactin-like immunoreactivity were observed. The diameter of the varicose enlargements mainly ranged from 0.5 to 1.5 μm and exhibited a strong immunofluorescence. Distinct networks of beaded fibers of medium density were also observed in the dorsomedial hypothalamic nucleus (Fig. 1A), the arcuate nucleus (Fig. 1B), the ventral hypothalamus, the subependymal and inner layers of the median eminence (Fig. 1B), the area ventral to the fornix, the ventral and especially dorsal premammillary nuclei, the preoptic suprachiasmatic nucleus, the area dorsal to the supraoptic nucleus, and the area immediately dorsal to the roof of the third ventricle. The density was low to moderate. Networks of medium density were also observed in the paraventricular rotundocellular thalamic nucleus, the supramammillary commissure, especially medial to the fasciculus mammillotegmentalis, and within Forel's field H_2.

None of the terminals described above were observed after treatment of prolactin antiserum with rat prolactin (Fig. 1C). However, the specific immunofluorescence did not disappear when the prolactin antiserum had first been treated with ovine prolactin or the synthetic corticotropins. Furthermore, the prolactin-like immunoreactive material was still present 1 month after hypophysectomy. No specific immunofluorescence was observed after incubation with antibodies against lutropin, follitropin, thyrotropin, or somatotropin.

The present results give evidence for the existence of hypothalamic nerve terminals containing a prolactin-like protein. The close approximity of this terminal system to the ventricles makes it likely that part of the prolactin in the cerebrospinal fluid (*3*) is derived from this neuronal pool of hypothalamic prolactin-like material. In agreement with the present morphological findings, Clemens *et al.* (*5*) have shown that prolactin-responsive neurons exist in the periventricular rabbit hypothalamus and preoptic area. Therefore, a prolactin-like protein may have a role in synaptic function. An interesting question that now arises is whether different neurons are involved (i) in the inhibitory feedback action of prolactin on its own secretion (*6*) and (ii) in the possible neurotransmitter role of prolactin-like proteins.

These results are evidence for the storage of prolactin-like material in nerve terminals of the mammalian brain and have implications for the role of such proteins in brain function. These initial data must be regarded with caution particularly since it is not known how other peptides, not yet tested, react with the present antiserum.

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Heteromorphic Sex Chromosomes in Male Rainbow Trout

Abstract. *A pair of subtelocentric chromosomes differs in the size of the short arm in male, but not female, rainbow trout (*Salmo gairdneri*). The morphological similarity of the X and Y chromosomes, and the observation of Y chromosomes intermediate between the X and normal Y, suggest that the sex chromosomes are at an early stage of differentiation in this species.*

Heteromorphic sex chromosomes are believed to evolve from morphologically identical homologs (*1*). Although most fish have undifferentiated sex chromosomes, a number of examples of heteromorphic sex chromosomes have been found (*2–4*). In the rainbow trout (*Salmo gairdneri*), I have found what appears to be an early stage in the differentiation of the X and Y chromosomes.

Sea-run rainbow trout (steelhead) returning to the Washington Department of Game hatcheries on the Cowlitz and Washougal rivers were studied. Chromosome preparations were made from white blood cell cultures as previously described (*5*). At least five cells with the modal configuration were counted in all individuals. Previous work (*5, 6*) has shown that a Robertsonian

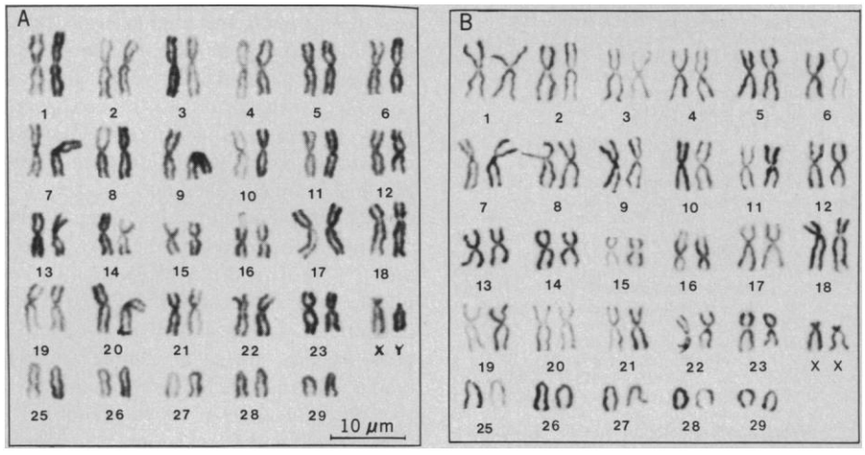

Fig. 1. Karyotypes of rainbow trout with $2n = 58$. (A) Male winter steelhead, fish 191 from the Cowlitz River, May 1976. (B) Female winter steelhead, fish 172 from the Cowlitz River, May 1976.

chromosome polymorphism, resulting in diploid numbers of 58, 59, or 60, is present in some rainbow trout populations. This report describes heteromorphic sex chromosomes in the $2n = 58$ individuals of known sex that I have examined (*7*).

Karyotypes of male and female rainbow trout are shown in Fig. 1. Chromosomes are arranged on the basis of size and centromere position. Three chromosome pairs can be unambiguously identified: Nos. 16, 18, and 24 (*5, 8*). I have noted that the subtelocentric pair (No. 24) is heteromorphic in males; one chromosome (the Y) has a short arm smaller than the short arm on its homolog or on the subtelocentrics in females (the X chromosomes). Figure 2 shows this subtelocentric pair from 46 rainbow trout; the difference in males is consistent although two males (fishes 111 and 116) have Y chromosomes with larger short arms than usual. In a blind study of 23 winter steelhead from the Cowlitz River (May 1976) the correct sex was determined in all fish. The heteromorphic sex chromosomes also appear to be present in males with 59 or 60 chromosomes that I have examined, but the difference is not as clear because these fish have one or two additional subtelocentric chromosomes, respectively (*5*).

The exceptional males (fishes 111 and 116) probably are examples of an intraspecific Y chromosome polymorphism. Other males in Fig. 2 may exhibit the polymorphism to different degrees. The polymorphism may reflect different degrees of morphological differentiation from the X chromosome among Y chromosomes in the species. This would imply that the main step in the differentiation was loss of short arm material on the Y; incipient sex chromosome dimorphism shown by interspecific comparisons has often been associated with pericentric inversions (*1, 2*). Another possibility is that a polymorphism in Y chromosome heterochromatin, analogous to Y chromosome polymorphisms seen in humans (*9*), could result in a fortuitous similarity between some Y chromosomes and the X chromosome. A third explanation of the exceptional males is that they might have been genetic females (XX) that developed as males because of environmental influences (*10*) or because of male influences of autosomal genes (*11*).

It is possible that heteromorphic sex chromosomes are absent in the other rainbow trout populations that have been examined cytologically (*6, 12, 13*). This would be particularly interesting because of the Y chromosome polymorphism seen among the Washougal summer steelhead (fishes 111 and 116); Y chromosomes may have randomly lost more inert genetic material in some populations than in others.

The sex chromosomes of salmonid fish do not appear to be highly differentiated. Meiotic studies in males of several fish species have revealed atypically behaving bivalents, characterized by end-to-end associations or positive heteropycnosis, which are believed to reflect differentiation of the sex chromosomes (*2, 3*). In some cases atypically behaving bivalents were seen in species lacking morphologically differentiated sex chromosomes. Meiotic studies of other rainbow trout populations (*12*) and related species (*6, 8*) have shown no evidence of atypically behaving bivalents. Heteromorphic sex chromosomes may be absent in other species of trout, salmon, and char (*6, 8, 14, 15*). In the rainbow trout, the subtle nature of the difference between the X and Y chromosomes, and

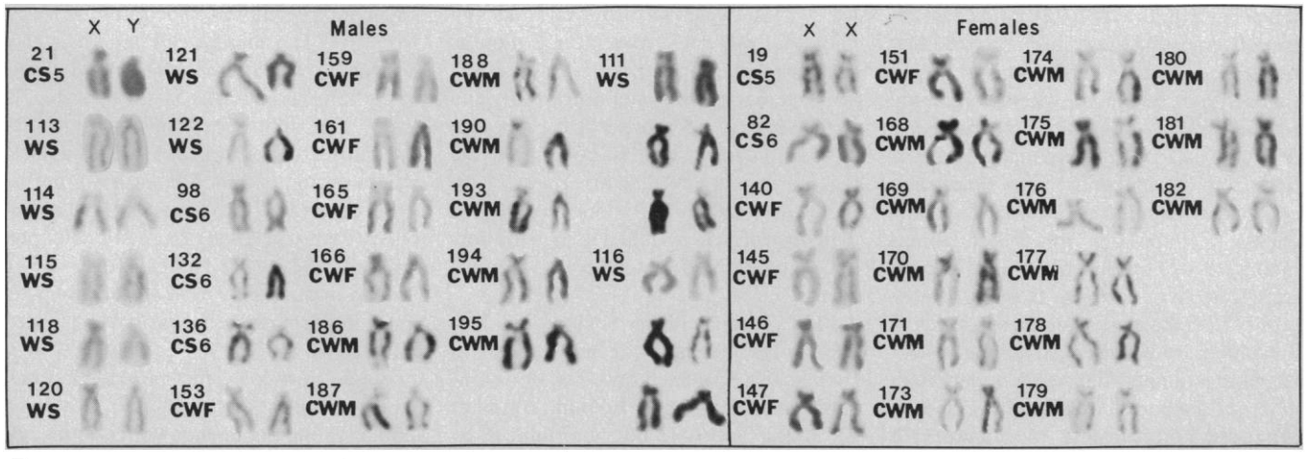

Fig. 2. Subtelocentric chromosomes from 46 rainbow trout with $2n = 58$. A chromosome pair from one cell from each individual except fishes 111 and 116 is shown. In the case of these two exceptional males, chromosome pairs from three cells each are shown. The origins of the fish are abbreviated as follows: Washougal summer steelhead (*WS*), Cowlitz summer steelhead caught in 1975 (*CS5*), Cowlitz summer steelhead caught in 1976 (*CS6*), Cowlitz winter steelhead caught in February 1976 (*CWF*), and Cowlitz winter steelhead caught in May 1976 (*CWM*).

the observation of Y chromosomes intermediate between the X and normal Y, suggest that we may be observing an early stage in the evolution of heteromorphic sex chromosomes.

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Hibernation and Body Weight in Dormice: A New Type of Endogenous Cycle

Abstract. *Under conditions unfavorable to hibernation (22°C), the body-weight cycles of dormice are only a few weeks long, but under conditions in which dormice hibernate (5°C), the cycles can last many months; the more the animals hibernate, the longer are the cycles. Such cycles contrast with the relative independence from torpor of the period of circannual cycles in other hibernating rodents and with the temperature compensation of circannual and circadian cycles in general.*

Circannual cycles in animals maintained in a constant environment for long periods occur in a variety of species, but the underlying mechanisms are poorly understood (*1*). Given the well-developed state of knowledge on circadian rhythms, it has been natural to apply concepts that have been useful in research on circadian rhythms to circannual rhythms (*2, 3*). In this context, one must, when developing theories of circannual rhythms, be aware of cycles that are different in their properties from circadian cycles. One such cycle, with properties different from circadian rhythms and from other circannual cycles so far described, is the body-weight cycle in dormice, *Glis glis*. I now report that the period of these cycles is dependent on whether the animal lowers its body temperature and hibernates, whereas the period of circadian cycles is generally compensated for changes in temperature (*4*).

Body-weight cycles with a period of approximately 6 weeks occur in dormice kept at about 25°C (*5*). Two observations suggested that these cycles might be temperature dependent. (i) A few animals with longer cycles than most tended to be lethargic and cool when weighed, even though they were in a warm room (*6*). (ii) When dormice are kept in a cold room (0° ± 2°C), cycles of torpor in those animals that hibernate average 6.1 months (*7*). However, in the first case there was no systematic assessment of torpor, and in the second, only torpor was recorded; it was thus impossible to compare cycle lengths in animals that hibernated with those that did not. I therefore studied weight cycles in matched groups of dormice maintained at different temperatures.

Fifty dormice were obtained in November 1974 from a dealer (Stacel) in France. They were housed under standard conditions in a holding room at 22° ± 2.5°C with 12 hours of light in 24 hours (LD 12 : 12) (*8*). The first 27 animals showing clear cycles of weight were selected for the experiment. One week after the weight of an individual animal had reached a peak the animal was transferred to either a cold room (5° ± 3°C) or another warm room (22° ± 3°C). Two animals were transferred to the cold room for every one to the warm room. Assignment to the rooms was made on the basis of a prearranged schedule, with the first animal that showed cycles going to the cold room, the second to the warm room, and the third to the cold room; this sequence was repeated for the next three animals as they reached and passed a peak weight, and so on. Three animals died in hibernation, leaving 15 animals in the cold room and 9 in the warm room.

Both cold and warm rooms were on an LD 12 : 12 hour cycle. The animals were weighed to the nearest gram once a week; for hibernating animals, weighing was postponed until they became active. Animals were inspected daily, and hibernation was monitored by the sawdust technique (*9*). After the main experiment was completed, direct measures of rectal temperature with a telethermometer (3-cm immersion) confirmed that animals judged to be hibernating were within 1°C of ambient temperature.

The experiment continued for each dormouse until its weight had reached at least one peak after the transfer to the cold or warm room. To be considered a peak in a cycle, the rise in weight had to be at least 20 g more than the lowest weight since the last peak, and it had to be followed by a drop of at least 20 g. This criterion corresponds well to intuitive judgments made by scanning graphs of body weight (Fig. 1).

The period of the first cycle completed after transfer to the cold room was much greater for those dormice that hibernated extensively than for those that did not (Figs. 1 and 2). Within the cold room the cycle lengths were positively correlated with the percentage of the cycle that was spent in hibernation (Spearman $\rho = .8$, $P < .01$). The mean cycle length in the 5°C room was 162 days (range, 28 to 425); in the 22°C room it was 53 days (range, 22 to 85). The difference stems mainly from the more frequent occurrence of cycles with a period of 3 months or more in the cold room ($P < .01$, Fisher exact probability test). Cycle lengths in animals that did not hibernate extensively, even though in the cold room, were similar to those of dormice in the warm room (Figs. 1 and 2).

When dormice hibernate in a cold room, the periodicity of their cycles is similar to the endogenous circannual cycles of other hibernating mammals (*2, 10*), although they can also be somewhat shorter (*7*). But when dormice are active, whether in a cold or a warm room, their