tions in grasshoppers[17], beetles[18], *Drosophila ananasse*[19]. However, some authors in the past[20] have concluded that there is no direct correlation between translocations and speciation.

We believe that the present study illustrates very well the karyotype variation between the 2 species of *Microgale*, *M. dobsoni* and *M. talazaci* and that the mechanism by which such a variation might have occurred in all probability is reciprocal interchange of chromosome segments[21, 22].

Résumé. Deux espèces d'insectivores, *Microgale dobsoni* et *M. talazaci*, ont le même nombre de chromosomes, $2n = 30$. Leurs caryotypes respectifs présentent des différences portant sur 2 pairs d'autosomes. Une translocation réciproque, devenue homocygote, permet d'expliquer ces différences. On peut concevoir que la spéciation est alors intervenue à la suite d'effets de position, de la stérilité des hybrides et de la préférence des porteurs de formules chromosomiques différentes pour telle ou telle «niche» écologique.

D. S. Borgaonkar and E. Gould

Division of Medical Genetics, Department of Medicine, The Johns Hopkins University School of Medicine and Laboratory of Comparative Behavior, Department of Mental Hygiene, The Johns Hopkins University School of Hygiene and Public Health Baltimore (Maryland 21205, USA), 11 December 1967.

[17] M. J. D. White, J. Hered. *31*, 137 (1940).
[18] S. G. Smith, Can. Ent. *94*, 941 (1962).
[19] Th. Dobzhansky and A. Dreyfus, Proc. natn. Acad. Sci., U.S. *29*, 301 (1943).
[20] S. Satina, Am. J. Bot. *40*, 638 (1953).
[21] Acknowledgements. The authors gratefully acknowledge the generous hospitality and help given by Drs. Brygoo, Dodin, Mayeaux of the Institut Pasteur, Tananarive, Madagascar, and thank Dr. J. Eisenberg for some useful suggestions and discussion. Expert technical assistance of H. Herr, E. Mules, and K. Johnson in the chromosome laboratory is appreciated.
[22] Part of this work is supported by National Science Foundation Grants Nos. GB 4978 (D. S. B.), GB 4436 (E. G.), U.S. Air Force AFOSR 586-67 (E. G.), American Philosophical Society Penrose Fund Grant No. 4314 (D. S. B.).

On the Genetics of a Circadian Rhythm in *Drosophila*

From all we know so far certain DNA-controlled processes seem to play an important part in the control of cellular circadian rhythms[1]. It is, however, not clear whether the entire genome, chromosome sections, or single genes determine the parameters of the cellular rhythm and maintain the oscillations. Probing experiments had led us to the idea that sexual differences in the circadian rhythm may be correlated to the ratio of X chromosomes to autosomes. In order to analyze this question in more detail we tested the pattern of the circadian rhythm of oxygen consumption in 26 mutants of *Drosophila melanogaster* Meigen.

Each experiment was started when the flies were 3–6 days old. O_2-consumption was measured for groups of 5 animals and for single controls in 12 ml and 6 ml Warburg flasks, respectively. The water bath around the flasks was kept at 25° ± 0.01 °C. The flies were exposed to light (90 lx) from 09.00 to 21.00 h. Readings from 3 flasks (with 5 animals each) were averaged over 2 days and the standard error calculated. In our method the absolute values may depend on the diffusion- and absorption-velocities of the CO_2 emitted. For the present argument, however, only relative changes are considered.

The results of the various mutant stocks indicate that the circadian pattern of oxygen consumption of females (Figure 1, b, c, d) and males (Figure 1f, g, h) differ. As shown previously[2], wild type females and males show a morning and an evening peak of oxygen consumption (Figure 1d, f), a pattern that also appears in the hormonal system[3]. In females, however, the morning maximum is smaller than the evening maximum, and in males both maxima are almost equal with respect to the total oxygen consumed between 2 minima. In about half of the mutants tested, the females show only a single peak in the evening (Figure 1b), or a second peak in the morning is just faintly indicated, e.g. in a triploid stock (Figure 1c). The males never have only one peak in the evening. They show, varying in extent, higher morning and lower evening maxima (Figure 1f, g, h), as compared to the females. The closed-X mutants, in particular, yield a very small evening maximum or, in some experiments, none at all.

If these differences in the circadian pattern were due to a different ratio of X chromosomes to autosomes it should be possible to support this idea by testing other ratios, as represented e.g. by intersexes (0.67) or superfemales (1.5). Intersexes of the triploid stock (Figure 1e) show, in fact, a morning maximum that is about the intermediate of the females (Figure 1c) and the males of this stock. (The additional broadening of this peak into the dark period is probably connected with the gene w^a, since w-males also show this change.) Four females, out of approximately 900 females of an attached-X stock (cs^{53}/*y w bb*) which showed wild phenotype and turned out to be sterile, we assumed to be superfemales. In one case we observed a detachment in this stock, where the appropriate number of males with *y w bb* turned up in the progeny. The attached-X and the detached-X females and the males show a two-peak pattern of O_2-consumption, whereas the 4 superfemales tested exhibit a pronounced, single maximum in the evening (Figure 1a).

If one arranges the curves according to a relative decrease in the evening maximum and to a relative increase in the morning maximum, it becomes evident that in this sequence also the ratio of X chromosomes to autosomes

[1] M. W. Karakashian and J. W. Hastings, J. gen. Physiol. *47*, 1 (1963). – E. Schweiger, H. G. Wallraff and H. G. Schweiger, Science *146*, 658 (1964). – C. F. Ehret and E. Trucco, J. theor. Biol. *15*, 240 (1967). – H. D. Frey and B. Schwemmle, in *Biologische Rhythmen* (Ed. G. Birukow and L. Rensing; Nachr. Akad. Wiss. Göttingen 1967), vol. 10. – D. Neumann, Helgoländer wiss. Meeresunters. *15*, 163 (1967).
[2] L. Rensing, Z. vergl. Physiol. *53*, 62 (1966).
[3] L. Rensing, Science *144*, 1586 (1964); Z. Zellforsch. mikrosk. Anat. *74*, 539 (1966).

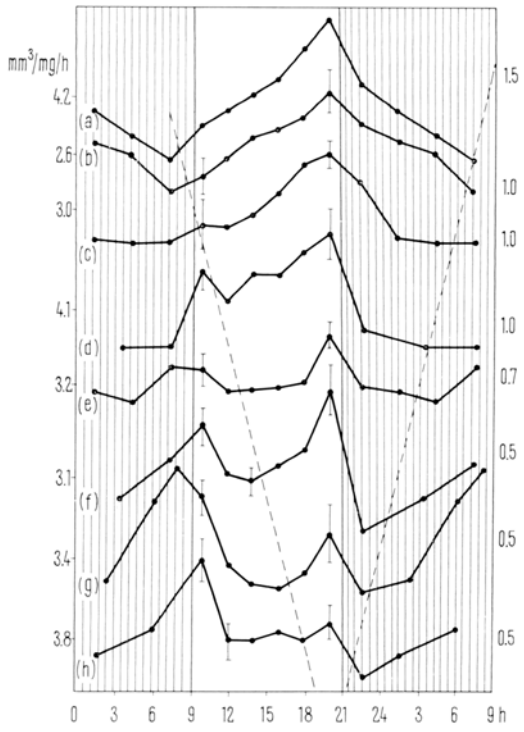

Fig. 1a–h. Circadian pattern of oxygen consumption. (a) Super-females from an attached-*X* stock (*cs*^{53}/*y w bb*; Pasadena). (b) Females (*sn*; Göttingen). (c) Triploid females (*y*^{2}*sc w*^{a} *ec*/FM4, *y*^{31d} *sc*^{8} *dm B*/*sc*^{8}. *Y*; *cn*/*cn*/*cn*; Pasadena). (d) Wild females (Princeton). (e) Intersexes (cf. *c*). (f) Wild males (Princeton). (g) Males (I(2)*gl cn bs*/*SM5*, *al*^{2} *Cy*, *lt*^{v}*sp*^{2}; Philadelphia). (h) Males (*X*^{c2}, *w*^{vc}/*sc*^{8}. *Y*; Tübingen). Left ordinate: mean O_2-consumption during 24 h in mm³/mg fresh weight/h. Right ordinate: *X* chromosome/autosome ratio. Abscissa: time in hours. Points of the dark phase are plotted twice on the same curve; vertical lines indicate the standard error, dotted line the approximate shift of the minima.

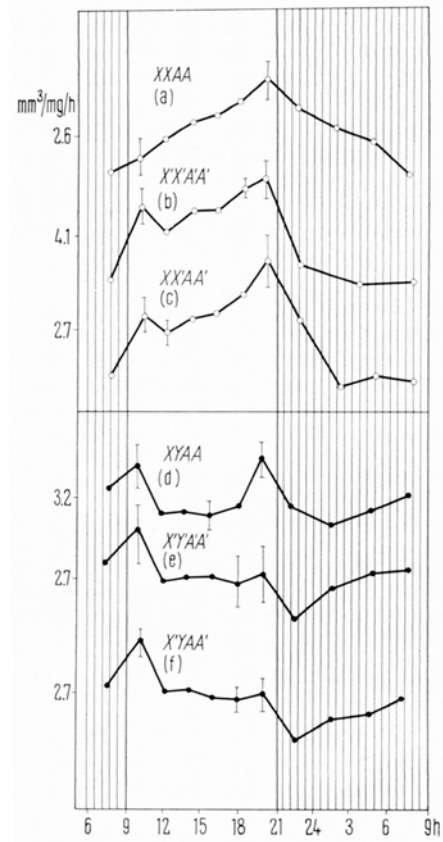

Fig. 2a–f. Crossings between stocks with different circadian pattern of O_2-consumption. (a) *sn* females. (b) Wild type females. (c) F_1-females from *sn* ♀♀ *X* wild type ♂♂. (d) Males of an attached-*X* stock (*cs*^{53}/*y w bb*). (e) Males (*X*^{c2}, *w*^{vc}/*sc*^{8}. *Y*). (f) F_1-males from attached-*X* ♀♀ *X* closed-*X* ♂♂. Ordinate, abscissa and standard error as in Figure 1.

is decreasing (Figure 1a to h). If this correlation is relevant, the differences among various genotypes of the same ratio (*X* chromosomes to autosomes) might be explained by disturbances in the effects of *X* chromosomes and/or autosomes on the circadian pattern.

In crossing experiments we aimed at a further clarification of the role the *X* chromosomes and autosomes play in determining the circadian pattern. In a first series, we crossed females without a morning maximum (Figure 2a) with wild type males. The F_1-females (Figure 2c) show a small morning maximum that is about equal to that of wild females (Figure 2b). From this experiment, however, no decision can be made whether this effect is brought about by more autosome action or less *X* chromosome action of the wild genome. In order to determine the extent to which the *X* chromosome forms the pattern, we crossed in several series of experiments, following the attached-*X* method, closed-*X* males (Figure 2e) with attached-*X* females. The pattern of the F_1-males (Figure 2f) corresponds well with the pattern of the closed-*X* fathers, but not with the pattern of the wild type males of the attached-*X* stock (Figure 2d). Consequently, since the autosomes are derived from both parents, the *X* chromosomes can be considered as the determining factor for the small evening maximum in this case. A clear dominance of the father autosomes would offer an alternative explanation but in view of the above described quantitative character this does not seem very likely.

Interpreting these results one might speculate that there are 2 circadian oscillations which have a phase difference of 12 h and which depend quantitatively on the *X* chromosome and autosomes, respectively. Various kinds of interactions between these oscillations may occur, but this remains to be analyzed in further experiments[4].

Zusammenfassung. Das tagesperiodische Muster des Sauerstoffverbrauches von Drosophilamutanten weist mehr oder weniger grosse Unterschiede zwischen den Geschlechtern auf: von zwei Maxima ist das abendliche bei den Weibchen stärker ausgeprägt als bei den Männchen, bei denen das Morgenmaximum relativ höher ist. Aus Versuchen mit Superweibchen und Intersexen sowie aus Kreuzungsexperimenten scheint hervorzugehen, dass das Verhältnis von *X*-Chromosomen zu Autosomen bei der Ausprägung der beiden Maxima eine Rolle spielt.

L. RENSING, W. BRUNKEN and R. HARDELAND

1. Zoologisches Institut der Universität, 34 Göttingen (Germany), 24 November 1967.

[4] This work was supported by the Deutsche Forschungsgemeinschaft.