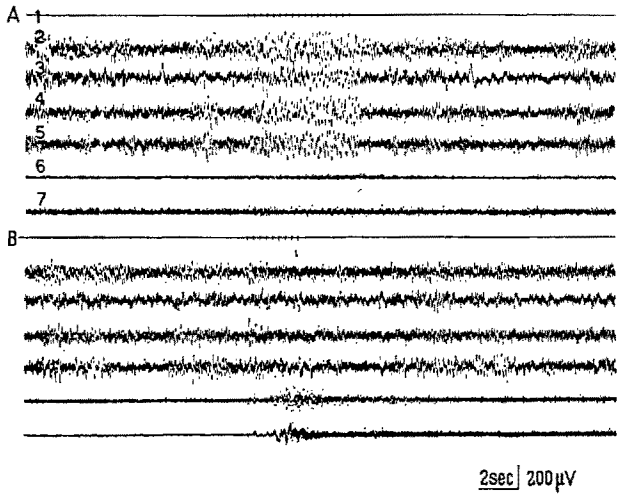

Stimulation of the left superficial radial nerve at 3/sec, 0.5 msec and 395 μA produced synchronization (A). The eyes promptly closed at the onset of the stimulus and remained so. The stimulus intensity used was just below arousal threshold, as shown by the slight increase of EMG activity near the end of the stimulus train. Slight increase of the stimulus intensity to 430 μA caused arousal (B). The animal's eyes reopened and EMG activity increased markedly. 1: stimulus artefact; 2: right parietal-temporal; 3: right temporo-occipital; 4: left parietal-temporal; 5: left temporo-occipital; 6: EMG left neck extensor; 7: EMG right neck extensor.

bursts. Between 3–8/sec the EEG synchronization was usually uninterrupted and often tended to follow the frequency of the stimulus providing the stimulus intensity was sufficiently high. Increasing the frequency above 8/sec brought about a gradual reduction of the amplitude of the synchronized EEG pattern until all evidence of an induced synchronization disappeared altogether. The appearance of the true arousal occurred at frequencies higher than 12–16/sec. In such cases the effect was stronger when stimulus intensity was high enough to excite a small number of fibers of higher threshold capable of producing arousal in addition to those driving synchronizing mechanisms.

The conclusion is drawn that, under appropriate conditions, peripheral nerve stimulation may activate mechanisms responsible for EEG synchronization.

Riassunto. La stimolazione a 3–8/sec di nervi cutanei e muscolari, eseguita in gatti integri non anestetizzati, produce una sincronizzazione dell'attività elettrica cerebrale. La soglia per questa risposta corrisponde in media a 0,4–0,9 volte l'intensità minima di corrente che applicata allo stesso nervo e in analoghe condizioni sperimentali produce la reazione di risveglio.

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The Effect of Litter Rank on the Secondary Sex Ratio

The secondary sex ratio (the number of males per 100 females at birth) has been reported to decrease in succeeding litters of rats[1] and with increasing birth order in humans[2]. In mice PARKES[3] found a decrease in later litters while HOWARD et al.[4] failed to observe any change. Recently, during the course of investigations employing mice from a variety of inbred strains data were obtained pertaining to the secondary sex ratio in succeeding litters. Even though the sample size was small, it was felt that

[1] H. D. KING and J. M. STOTSENBURG, Anat. Rec. *9*, 403 (1915).
[2] E. NOVITSKI and L. SANDER, Ann. Human Genet. *21*, 123 (1956–57).
[3] A. S. PARKES, Biol. Rev. *2*, 1 (1926–27).
[4] A. HOWARD, A. MCLAREN, D. MICHIE, and G. SANDER, J. Genet. *53*, 200 (1955).

The secondary sex ratio of mice of successive litters

Strain	Litter rank	Litters	Total mice	Males	Males per litter	Females	Females per litter	Ratio
C3H/Sp	1	17	82	55	3.2 (0.4)[a]	27	1.6 (0.3)[a]	204
	2	17	101	55	3.2 (0.4)	46	2.7 (0.3)	120
	3	12	75	37	3.1 (0.3)	38	3.2 (0.5)	97
C3Hf/Sp	1	13	64	40	3.1 (0.5)	24	1.8 (0.3)	167
	2	17	91	56	3.3 (0.3)	35	2.1 (0.2)	160
	3	10	52	22	2.2 (0.4)	30	3.0 (0.5)	73
PL/Sp	1	18	77	57	3.2 (0.5)	20	1.1 (0.3)	285
	2	18	83	48	2.7 (0.4)	35	1.9 (0.4)	137
	3	9	42	25	2.8 (0.7)	17	1.9 (0.3)	147
AKR/Sp	1	16	90	62	3.9 (0.5)	28	1.7 (0.4)	221
	2	14	76	42	3.0 (0.4)	34	2.4 (0.4)	124
	3	6	34	22	3.7 (0.6)	12	2.0 (0.5)	183
MA/Sp	1	10	59	39	3.9 (0.8)	20	2.0 (0.4)	195
	2	9	57	31	3.4 (0.5)	26	2.9 (0.3)	119
	3	8	53	38	4.7 (1.1)	15	1.9 (0.4)	253
C57BL/Sp	1	10	42	23	2.3 (0.4)	19	1.9 (0.3)	121
	2	9	44	21	2.3 (0.6)	23	2.6 (0.3)	91
	3	7	36	18	2.6 (0.7)	18	2.6 (0.7)	100
Pooled F_1 hybrids C57BL × PL/Sp	1	13	82	46	3.5 (0.4)	36	2.8 (0.4)	128

[a] Figures are averages and standard errors.

some of the differences were of enough significance to warrant presentation at this time.

Female mice of the strains listed in the Table, obtained from the inbred stocks of the Detroit Institute of Cancer Research, were mated with their brothers and the progeny of the first, second, and third litters were sexed within 16 h after birth. Sexes were rechecked at least twice thereafter. Mating pens consisted of 1–4 females and 1 male. All mice were kept in wooden boxes, which were changed weekly, on racks in an air-conditioned room. Water and a commercial diet (Purina Laboratory Chow) were given *ad libitum*. When a female became detectably pregnant, she was isolated. She was returned to her original mate after her litter was weaned at 5 weeks or earlier if the litter died prior to weaning. As far as could be judged from carcasses, the number of stillbirths was estimated at less than 1%; and since it was difficult to sex the remains, they have been excluded from the data.

The results are presented in the Table. All data were analyzed statistically. Those involving the number of mice per litter were tested by Student's t-test; all others were analyzed by the Chi square method with Yates's correction for sample size.

The secondary sex ratio was elevated in all first litters but was significantly different from the theoretical value of 100 only in those born to C3H/Sp ($P < 0.05$), PL/Sp ($P < 0.01$), and AKR/Sp ($P < 0.02$) mothers. The ratio in second and third litters was invariably lower and not significantly different from the theoretical except in the case of third litters of MA/Sp mice ($P < 0.05$). The ratio was significantly decreased in second and third litters of C3H/Sp, $C3H_f$/Sp and PL/Sp mice ($P < 0.05$) from that found in first litters.

The decrease in the secondary sex ratio in succeeding litters could have been due to a decrease in the number of male births and/or an increase in female births. To elucidate this point the number of male and female births per litter were compared in those strains (C3H/Sp, $C3H_f$/Sp, and PL/Sp) that showed a significant decrease in the ratio. Significant increases in female births were found in second and third litters (P was 0.01) of C3H/Sp mice. The increase in female births in third litters of $C3H_f$/Sp and PL/Sp mice was at the borderline of significance (P was 0.05). No significant change in the number of male births was observed. These findings suggest that the increases in female live births played a role in the decrease of the secondary sex ratio in later litters of these strains. The data are contrary to the generally held idea that the decrease in the sex ratio at birth is due to a diminution in male births[5,6].

The deviation of the observed ratio from the theoretical value of 100 in first litters of C3H/Sp, PL/Sp, and AKR/Sp mice could be due to genetic and/or nongenetic factors. Mating of PL/Sp females with C57BL/Sp males (from a strain in which no deviation of the observed from the theoretical ratio was found) yielded first litter F_1 hybrids in which the ratio was 138. The latter did not significantly deviate from the theoretical. The reciprocal crosses yielded first litter progeny in which the ratio was 122. The latter did not significantly deviate either from the theoretical or from the actual ratio found in the first cross. The data for the first litter reciprocal F_1 hybrids were, therefore, pooled. The ratio of these pooled hybrids differed significantly from that of PL/Sp first litter mice ($P < 0.05$) but not from that of C57BL/Sp first litter progeny. These figures suggest that the apparent increase in the secondary sex ratio of first litter PL/Sp mice may be partially attributed to genetic factors. The decrease of the ratio in succeeding litters then implies a compensating nongenetic mechanism.

Zusammenfassung. Es wird gezeigt, dass sich verschiedene Mäusestämme im sekundären Geschlechterverhältnis unterscheiden. Die Wurffolge beeinflusst zwar die relative Zahl der Geschlechter bei der Geburt, aber dieser Einfluss wechselt mit dem Stamm selber. Der Ausgleich des Unterschieds in späteren Würfen erfolgt durch Weibchenüberschuss. Sowohl erbliche wie nichterbliche Faktoren spielen eine Rolle in der Beeinflussung des Geschlechterverhältnisses.

S. Albert, Kittie Tomson, and R. R. Small

Detroit Institute of Cancer Research and Wayne State University College of Medicine, Detroit (Michigan), February 14, 1961.

[5] J. A. Weir, J. Heredity *46*, 277 (1955).

[6] A. S. Parkes, Proc. Roy. Soc. London *95*, 551 (1923–24).

[7] *Acknowledgment.* This investigation was supported in part by research grant No. C-2151 from the National Cancer Institute, Public Health Service and in part by an institutional grant to the Detroit Institute of Cancer Research from the United Foundation of Greater Detroit through the Michigan Cancer Foundation.

The Content of Acid Fraction in Cannabis Resin of Various Age and Provenance

Considerable interest has recently been focused on the alkali soluble part of cannabis resin. Although this fraction does not exhibit any marked hashish activity[1,2], it has been observed that such constituents readily become converted by heat or by long standing into biologically active cannabinolic compounds which are insoluble in aqueous alkali[1,3]. After the isolation of cannabidiolic acid from the alkali fraction of the resin[4,5], this constituent was believed to be precursor of other cannabinolic substances[6]. By decarboxylation it yields cannabidiol[6] which can be converted by intramolecular condensation into physiologically active tetrahydrocannabinols[7], the latter yielding by spontaneous dehydrogenation almost inactive cannabinol[8,9].

Although the process of formation of various constituents of cannabis resin seems to be a very complex one, cannabidiolic acid may be considered as the main initial constituent in the gradual conversion of cannabinolic compounds. Consequently, the stage of the conversion process in a given sample of cannabis may be expected to be roughly indicated by the approximate content of cannabidiolic acid in hemp resin. Such data might be of interest in approaching the problem of determining physiological potency, geographical origin and the age of the drug.

In order to obtain a picture of the approximate content of cannabidiolic acid in various types of hemp resin, the acid fraction of the resin extracted from 31 available

[1] A. Madinaveitia, P. B. Russel, and A. R. Todd, J. chem. Soc. *1942*, 628.

[2] B. C. Bose and B. Mukerji, Nature *152*, 109 (1943).

[3] C. C. Fulton, Industr. eng. Chem. Analyt. Ed. *14*, 407 (1942).

[4] Z. Krejči and F. Santavy, Acta Univ. Olomuc. *6*, 59 (1955).

[5] O. E. Schultz and G. Haffner, Arch. Pharm. *291/63*, 391 (1958).

[6] O. E. Schultz and G. Haffner, Arch. Pharm. *293/65*, 1 (1960).

[7] R. Adams, D. C. Pease, C. K. Cain, and J. H. Clark, J. Amer. chem. Soc. *62*, 1868 (1944).

[8] J. Levine, J. Amer. chem. Soc. *66*, 1868 (1944).

[9] S. Loewe, Science *102*, 615 (1945).