

heated Berkefeld filtrate from closed intestinal loops was more toxic for guinea pigs than fluid heated for one hour at 65° C. The possibility that the diminished toxicity of the heated fluid might have been due to mechanical loss of toxins rather than to thermal inactivation has not been excluded.

4445

**The Free-martin Effect in Experimental Parabiotic Twins of
Triturus torosus.***

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It is known since the work of Keller and Tandler¹ and Lillie² that in heterosexual twins of cattle with fused chorionic membranes the male part exerts a very characteristic influence on the differentiation and the development of the gonads of its female cotwin. This action results in the following distinctive features of the free-martin gonad:

- (1) Sterility in consequence of the absence of germ cells.
- (2) Reduced or completely suppressed ovarian cortex.
- (3) Hypertrophied medulla with more or less pronounced male type of differentiation.

Experiments with amphibians joined in parabiosis at an early embryonic stage were expected to provide possibilities of a detailed analysis of the mechanism of this effect. This hope was rather disappointed, however, when it appeared from the results of Burns³ and Witschi's⁴ investigations that *Amblystoma* and *Rana* behave otherwise than cattle twins and represent two different and new types of reactions. In heterosexual combinations of *Amblystoma* either one of the sexes may already in the primary sex differentiation assume the absolute leadership, so that pairs of males and pairs of females are obtained exclusively. In frogs, on the other hand,

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¹ Keller, K., and Tandler, J., *Wiener Tierärztl. Wochenschrift*, III. Jahrg., 1916, 513.

² Lillie, F. R., *Science*, 1916, lviii, 611; *J. Exp. Zool.*, 1917, xxiii, 371; *Biol. Bull.*, 1923, xlv, 47.

³ Burns, R. K., *J. Exp. Zool.*, 1925, xlii, 31.

⁴ Witschi, E., *Biol. Bull.*, 1927, lii, 137; *J. Exp. Zool.*, 1927, xlvii, 269.

the two sexes first develop independently. Later on the male becomes predominant and causes a regular sex transformation in the female cotwin. The case of *Rana* comes closer to the cattle type than the case of *Amblystoma*, because of the finally established dominance of the male sex, however, it differs from it by the accomplishment of perfect sex inversion in the female cotwin.

A new group of experiments with the Californian salamander, *Triturus torosus*, yielded results more nearly comparable with the conditions in cattle. Of twenty-two pairs of twins, joined in parabiosis at the stage immediately preceding the beginning of muscular movements and raised until close to metamorphosis, one half proved to be unisexual (4 ♀ ♀ and 7 ♂ ♂), while the other half was heterosexual (6 ♀ ♂ and 5 ♂ ♀). The second group shows a very interesting range of variation which will be described in detail in a paper under preparation. It comes nearer the cattle type than any other case of experimental parabiosis so far recorded. (1) The male, without exception, is the leading part. Its gonads are always larger, often many times larger than those of the cotwin. (2) The gonads of the female contain only a very few scattered germ cells or are completely sterile. In no case do they contain ovocytes or young egg cells, which at this time are abundant in single controls and in the double female twins. (3) The gonads of the female show a tendency to assume male characters. The medullary cords are compact and only in a few places may show some rudiments of ovarian sacs. In four out of the eleven heterosexual twin females a very few spermatogonia (from 1 to 6 per animal) are found. A fifth one possesses a testicular segment of considerable size in the right gonad, while the left side is perfectly sterile. These occurrences indicate that at least in some cases a complete sex inversion might have been the final result.

While thus, in a general way, parabiosis in *Triturus* produces an effect which closely resembles the observed consequences of chorionic fusion in cattle, there are some particularities to be mentioned, never recorded so far for any other material. (4) The heterosexual twins influence each other *mutually*. Not only the gonads of the female but also the testes of the male contain a much reduced number of germ cells. It is true that in no case has resulted a completely sterile condition in the male, though the histological appearance of the testis is often thoroughly changed by the scarcity of spermatogonia. This is mainly true in those cases which are conspicuous for a luxuriant growth of the medullary cords.

(5) The most interesting feature displayed by the heterosexual

twins is the dislocation of the germ cells. Especially in the females one finds practically all the germ cells outside of the gonads in the mesovarium in the direction of the kidney as well as towards the fat body. In the males the extra regional germ cells concentrate rather in the hilum region of the gonad. The germ cells outside the gonads are more or less reduced in size but show the characteristic multilobate nucleus. The failure of the germ cells to reach their final position is not due to the general conditions of parabiosis as evidenced by the unisexual twins in which extragonadal germ cells may only exceptionally be found. It evidently is caused by an antagonistic reaction between the sexes, resulting in a mutual inhibition of the processes leading to the differentiation of the primordial gonads. Therefore in this experiment we deal with a so far unique case of manifestation of sex-antagonism prior to the morphological formation and differentiation of the gonads.

4446

Effect of Oestrus Upon Achilles Reflex Time.*

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Several authors have remarked upon the relationship, in mammals, between the functional sex glands and behavior—general¹ and specific.² No work has been done on the relationship between the normal functional sex glands and the central nervous system. This report gives the influence of oestrus on the electromyographically determined Achilles reflex time. Eight normal female rats with known sex histories were followed through 18 consecutive cycles, varying from 1 to 4 cycles per individual, and reflex time determinations were taken during the active phase of the sex cycle—oestrus, and during the quiescent phase—dioestrus. Vaginal smears were made daily according to the technique of Long and Evans (1922) to determine the phase of the cycle. Our determination of

* The expenses for this investigation were defrayed in part by a grant from the National Research Council Committee on Research in Problems of Sex.

¹ Richter, C. P., *Quart. Rev. Biol.*, 1927, ii, 307.

² Macht, D. I., and Seago, D. W., *J. Comp. Psychol.*, 1924, iv, 151.