identical conditions, with rendered lamb fat instead of Intarvin added to it. In each successive generation, thereafter, the maternal rats that received Intarvin were selected from the direct descendants of those that had previously been given the "intarvinated" diet. The control maternal rats in each generation in a series were selected from the direct descendants of those that received a diet containing an addition of lamb fat. The paternal rats in each generation (unrelated to the females) were selected on the same dietary basis as that for the maternal, or from litters on the ordinary diet without addition of fatty matter.

Rats have been carried, in this parallel way, into the fourth generation in one series and into the third generation in another. (The experiments are in progress; and litters in the fifth and fourth generations, respectively, will soon be available.) There has been no discernible effect on the animals thus treated, or on their fecundity. The animals in the third and fourth generations appear to be as completely normal as those with which the tests were started last August.

The tissues of many of the rats have been used in studies of the absorption and distribution of glyceryl tri-margarate. These results, with those of various extensions of the study, will be reported later.

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The effect of light doses of alcohol upon the estrus cycle, and on the number of corpora lutea and prenatal mortality in the mouse.

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Stockard and Papanicolaou, Arlitt, Stockard, Hanson, MacDowell[1] have found that alcohol may cause partial or complete sterility in laboratory mammals.

[1] Stockard, C. R., and Papanicolaou, G. N., *J. Exp. Zoöl.*, 1918, xxvi, 119; Arlitt, A. H., *Psychol. Monog.*, 1919, xxvi, —; Stockard, C. R., *Proc. Amer. Phil. Soc.*, 1923, lxii, 311; Hanson, F. B., and Handy, V., *Amer. Nat.*, 1923, lvii, 532; MacDowell, E. C., *Genetics*, 1922, vii, 117, 427, *J. Exp. Zoöl.*, 1923, xxxvii, 417.

This has been explained both as a result of the elimination of less vigorous germ cells or zygotes, and as an immediate effect of the alcohol upon the activity of the ovary. The present experiments offer a method for analyzing more fully than before the effects of alcohol upon the reproductive processes of a mammal. The following criteria are used here for the first time in a study of this type: (1) the length of the estrus cycle, (2) the number of corpora lutea for given pregnancies, and (3) the proportion of these represented at the subsequent births by living young.

The mice used came from lines that had been inbred for three and four generations by brother to sister mating. Tests and controls from the same litter were kept in the same pen to equalize environmental conditions as far as possible. The alcohol was given by inhalation in pint milk bottles. Before treatment each mouse from one pen was placed in its own bottle; into each test bottle was then inserted a strip of absorbent paper, approximately 37 by 15 cm., wet with 3 cc. commercial (95 per cent) alcohol. The bottle was capped and inverted at once. Into each control bottle was inserted a similar piece of paper without the alcohol; these bottles also were closed and inverted. Each mouse remained in its bottle for 45 minutes. This treatment, starting at weaning (4 to 5 weeks), was given daily. The controls seemed to be unaffected by confinement in the closed bottles. For the first week the alcohol treatment left most mice flat on their sides; later, when most mice could turn over and run off, their behavior still showed an unquestionable effect of the alcohol. A few individuals were left limp, on their sides every time. This treatment is called light in contradistinction to the treatment being used in a subsequent experiment, by which every animal is made dead drunk every time.

Estrus cycle. From weaning till the mice were mated, daily observations were recorded upon the condition of the vagina. After the formation of the vaginal orifice, these observations were based upon the microscopical examination of vaginal smears. The beginning of estrus proper, indicated by the sudden appearance of large cornified epithelial cells (Allen[2]) was chosen as the point by which to measure the length of the cycles. From 41 to 87 smears were taken from each animal; the aver-

[3] Long, J. A., and Evans, H. M., *Mem. Univ. Cal.*, 1922, vi.

age number per mouse was 66.7. The variations in the number of smears taken depends partly upon the variations in the time of the opening of the vagina and partly upon the time of mating.

The age at the opening of the vagina ranged from 35 to 71 days for the tests, with an average of 54.0 days; for the controls, from 37 to 72 days, with an average of 52.2 days. For the rat, Long and Evans[3] give the average age for the opening of the vaginal orifice as 72 days, with a range from 34 to 109. These data may be considered to indicate that the administration of alcohol for an average of three weeks before the formation of the opening has had no effect upon this process.

Long and Evans report that the first estrus in the rat does not occur before the establishment of the vaginal orifice; that the two processes may be synchronous, or the first estrus follow the opening. Of the thirty-nine mice under consideration, only one, a test animal, showed signs of estrus at the time of the opening of the vagina. The age of the tests at the first estrus ranged from 47 to 107 days, with the average of 71.9 days; the age of the controls ranged from 51 to 115 days with the average of 68.2 days. This difference is probably due to random sampling as it is only about twice its probable error (6.6 ± 3.4).

The closest control of living conditions is obtained when the data on the length of the estrus cycle are arranged by pens. The closest control upon genetic differences is obtained when the data are arranged according to strains, for each strain includes closely related litters. Table I gives the data arranged in both the above ways.

This table shows that in four of the six pens, the average length of cycles was somewhat more for the tests; that in three of the four strains the average length of the cycles was less for the tests. The total shows practical identity. As far as these data are concerned, it is apparent that the treatment given did not increase the length of the estrous cycle.

Corpora lutea of pregnancy and prenatal mortality. A surgical technique for counting the corpora lutea of pregnancy has previously been described.[4] The count is made during the last week of pregnancy when the corpora lutea corresponding to the ova that gave rise to the pregnancy are the only large and con-

[3] Long, J. A., and Evans, H. M., *Mem. Univ. Cal.*, 1922, ix.

[4] MacDowell, E. C., *Anat. Rec.*, 1924, xxvii, 59.

TABLE I.
Average length of estrus cycle in treated and control mice.

	Treated.				Controls.			Differences.
	Pen No.	No. of mice	No. of cycles	Av. days in cycles	No. of mice	No. of cycles	Av. days in cycles	+ = test cycles longer
Data	439	4	24	7.9	3	25	7.4	+0.5
	441	3	14	10.2	3	22	8.7	+1.5
arranged	437	4	31	8.3	2	13	8.8	—0.5
	449	3	7	12.2	3	12	11.4	+0.8
by pens	445	2	8	11.6	1	5	9.2	+2.4
	443	6	22	9.8	5	12	14.2	—4.4
Data	Strain							
	C57	9	44	9.1	7	40	9.3	—0.2
arranged	42-30	6	21	10.9	6	34	9.6	+1.3
	86	2	21	7.9	1	6	9.5	—1.6
by strains	101	5	20	9.3	3	8	9.8	—0.5
Totals		22	106	9.3	17	89	9.5	—0.2

spicuous structures in the ovaries. Repetitions of this operation do not interfere with the normal activity of the ovaries. In addition to the evidence previously given upon this point, one case may be recorded in which, as a special test, the operation was repeated at ten successive pregnancies. The tenth litter was born 212 days after the first one, an average of 23.5 days apart. In all 77 young were born. Comparing litter size at the beginning and end of the series, the first three litters averaged 7.33 mice; the last three litters averaged 7.33 mice.

The test and control females in a litter were mated to the same normal male. The number of alcohol treatments given before the first conception averaged 118.8 per mouse, with a range from 76 to 205. No treatment was given between an operation and the birth of the litter. The young were killed on the day of birth and the female returned to her mating box. The daily treatments were resumed on the following day. The results are based upon 107 counts of corpora lutea of pregnancy by operation and 8 counts by autopsy. For the treated mice the average number of corpora lutea per pregnancy was 9.95; for the controls, 9.90.

By subtracting the number of young found at birth from the number of the corresponding corpora lutea a fair measure of the amount of prenatal mortality is obtained. This includes ova that were not fertilized, as well as zygotes that died at any stage before birth. In the previous study it was shown that the operation itself does not play a part in this prenatal mortality. In most cases mice found dead at birth appear to have been alive up to the point of birth but failed to survive the process, although there is no certain exclusion of the possibility that some of those found dead at birth had recently died *in utero*.

The average number of young that were expected at birth and not found, dead or alive, was 3.48 per litter for the tests and 3.44 for the controls. The average number of expected young that were not found alive was 3.82 per litter for the tests and 3.97 for the controls. The number of young found alive gave the average of 5.79 per litter for the tests and 5.79 per litter for the controls.

TABLE II.

Summary of data on the effect of light doses of alcohol upon the number of the corpora lutea of pregnancy and the amount of natal and prenatal mortality.

Strain	C57		42-30		86		101		Total	
	Test	Contr.	Test	Contr.	Test	Contr.	Test	Contr.	Test	Contr.
No. of mice	9	7	6	5	2	1	4	1	21	14
No. of counts										
a, by operation	28	23	16	15	8	9	6	2	58	49
b, by autopsy	4	1	2	0	1	0	0	0	7	1
Av. No. corpora per pregnancy	9.87	10.12	9.16	8.40	11.66	12.00	10.16	9.00	9.95	9.90
No. of litters	28	23	16	15	8	9	6	2	58	49
Av. No. young born alive per litter	5.85	5.43	5.37	5.13	6.12	7.88	6.17	5.50	5.79	5.79
Prenatal mortality per litter	3.28	3.87	3.18	2.60	4.37	3.88	4.00	3.00	3.48	3.44
Natal and prenatal mortality	3.67	4.34	3.75	3.33	4.37	4.22	4.00	3.50	3.82	3.97

Table II presents the above figures with other relevant data, including subdivisions according to strains. The evidence all leads to the conclusion that the alcohol has had no effect upon the number of corpora lutea formed or upon the survival of the zygotes.

Conclusion. It is concluded that the alcohol treatment has had no effect upon the reproduction of the mice studied when judged by the following criteria: (1) age of opening of the vaginal orifice; (2) age at first estrus; (3) length of estrus cycle; (4) number of corpora lutea per ovulation; (5) prenatal and natal mortality; (6) number of living young per litter.

244 (2476)

The food value of intarvin.

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In order to ascertain whether the synthetic fat, intarvin, composed of 17 carbon atom fatty acids, could be used by the body as a food, observations were made on two diabetic patients and four normal individuals. This was done in conjunction with work on dogs reported in another communication.[1] It is the purpose of this paper to present the data obtained from feeding experiments on two of the normal individuals. The other two normals were unable to eat enough intarvin to yield results sufficiently conclusive for interpretation.

Discussion.

The diabetic patients were upon restricted diets for therapeutic reasons. These diets were insufficient in calories to maintain nitrogen equilibrium and weight. They were receiving insulin. Additions of 80 and 100 grams of intarvin showed no increase in ketone bodies and was followed by a lowering in nitrogen excretion. The period during which intarvin was given was for one patient five days, for the other ten days. The results are not decisive enough to make it worth while presenting the protocols. Difficulty was experienced in getting the patients to take the intarvin and it had to be discontinued.

[1] Benedict, E. M., and West, R., Intarvin in phlorhizinized dogs, PROC. SOC. EXP. BIOL. AND MED., 1924, xxi, 225.