Brief Note

Exceptional Desoxyribose Nucleic Acid (DNA) Findings in a Sterile Dwarf Bull.*

By CECILIE LEUCHTENBERGER AND FRANZ SCHRADER. (*From the Institute of Pathology, Western Reserve University, Cleveland, and the Department of Zoology, Columbia University, New York.*)‡

In an extensive investigation of the cytological, cytochemical, and clinical features that are involved in the sterility of mammals (1–3), we have also made a study of so called dwarfism in cattle. It is almost certain that in cattle, as in most other mammals, several different types of dwarfs occur (4) and it is unfortunate that relatively little is yet known concerning their genetic background. It is clear however that, in some of its manifestations, dwarfism is linked with varying degrees of sterility (5), and this will explain our interest in the present case. The material in question stems from a dwarf bull that originated in a herd of pure bred Hereford cattle and constituted one of the individuals that is mentioned in a paper by Johnson, Harshfield, and McCone (5).

The tissues were fixed in 10 per cent formalin and embedded in paraffin. We obtained several of the paraffin blocks through the great kindness of Dr. L. E. Johnson and Dr. J. S. Harshfield. To serve as controls, tissues were also obtained from several normal bulls and were fixed in 10 per cent formalin, Carnoy, or Lavdowsky and embedded in paraffin. For the DNA analyses, sections cut from the dwarf and normal bull tissues and smears from the seminal fluids were subjected to Feulgen microspectrophotometry as previously described (1, 6, 7). The validity of Feulgen microspectrophotometry for determining the DNA content in individual cells and spermatozoa has been repeatedly demonstrated and can be considered as established (8, 9). Unfortunately, it was not possible to make reliable counts of the chromosomes in this material.

Table I gives a survey of the DNA findings. It is evident from the table that the amounts of DNA carried in the cells of the dwarf somatic tissues are only a little more than half the basic diploid DNA content found in normal somatic tissues. Furthermore, the spermatozoa in the epididymis and seminal vesicles of the drawf have only half the haploid DNA content found in the spermatozoa of the normal bulls. Consequently, in spite of the low DNA content, the DNA *ratio* between the somatic cells and spermatozoa is also 2 : 1 for the drawf, as would be expected and as is found for bulls with the normal DNA content.

In contrast to spermatozoa found in epididymis and seminal vesicles, those still within the seminiferous tubules of the testis fall into two classes, namely, one with the normal haploid DNA content and the other with half this haploid value. It should also be noted that in the normal bull, as well as in the dwarf, the DNA values of spermatozoa found within the testis proper are somewhat higher

* This study was supported in part by a grant from the Brush Foundation, and in part by a grant from the Franchester Fertility Fund, Cleveland.

‡ *Received for publication, September 14, 1955.*

than those of spermatozoa found in the epididymis, seminal vesicles, and seminal fluids. These differences in sperms derived from different regions of the reproductive organ are rather puzzling and we have, at present, no convincing explanation for their occurrence.

spermatozoa in the testis, there is a clear separation between the DNA contents of spermatozoa and somatic cells from the dwarf and those from normal bulls. It may be stated that such low quantities of DNA as characterize this dwarf are quite exceptional among

TABLE I

Mean Amount of DNA (Feulgen Microspectrophotometry) in Individual Nuclei of Somatic Cells and Spermatozoa from Normal and Dwarf Bulls

Classification of bull	Type and No. (n) of cells measured		Mean amount* of DNA per nucleus
Normal	Liver	$n = 160$	3.25 ± 0.03 ($n = 123$) 6.42 ± 0.21 ($n = 37$)
	Kidney	$n = 26$	3.3 ± 0.10
	Spleen	$n = 23$	3.0 ± 0.08
	Spermatozoa, in epididymis, seminal vesicle, and seminal fluid.	$n = 140$	1.64 ± 0.02
	Spermatozoa, in testis	$n = 61$	$2.16 \pm 0\ 04$
Dwarf	Adrenal	$n = 30$	1.95 ± 0.04
	Brain	$n = 60$	1.85 ± 0.04
	Epithelium of seminal vesicle	$n = 35$	1.95 ± 0.01
	Spermatozoa, in epididymis, and seminal vesicle	$n = 115$	0.88 ± 0.03
	Spermatozoa, in testis	$n = 47$	0.99 ± 0.05 ($n = 25$) 1.80 ± 0.08 ($n = 22$)

* Arbitrary units.

The difference in the DNA content between the dwarf and normal bulls is even more striking if the DNA data for the *individual* cells are examined. In Fig. 1, such individual DNA data are graphed for somatic cells and spermatozoa from both the dwarf and normal bulls. It can be seen that with the exception of the DNA values in the

the considerable number of dwarf bulls whose tissues we have examined, and which stemmed from various strains in different parts of the country. In an extensive study of dwarf bulls from other herds now being carried out in our laboratories, we have never encountered such low DNA values in somatic tissues.

It is significant that here is an indi-

vidual that reached maturity but in which the nuclei of at least three different tissues contain an amount of DNA that is much lower than that which is carried in the somatic tissues of normal bulls. Further, as already pointed out, at least some of the sperms produced by it—and all those that reached the epididymis and the seminal vesicle—carry only half of the amount of DNA that is present in normal sperms. This would seem to indicate that even though the spermatogonial nuclei carried only half of the normal amount of DNA to start with, some sort of miotic process occurred nevertheless, and through this the quan-

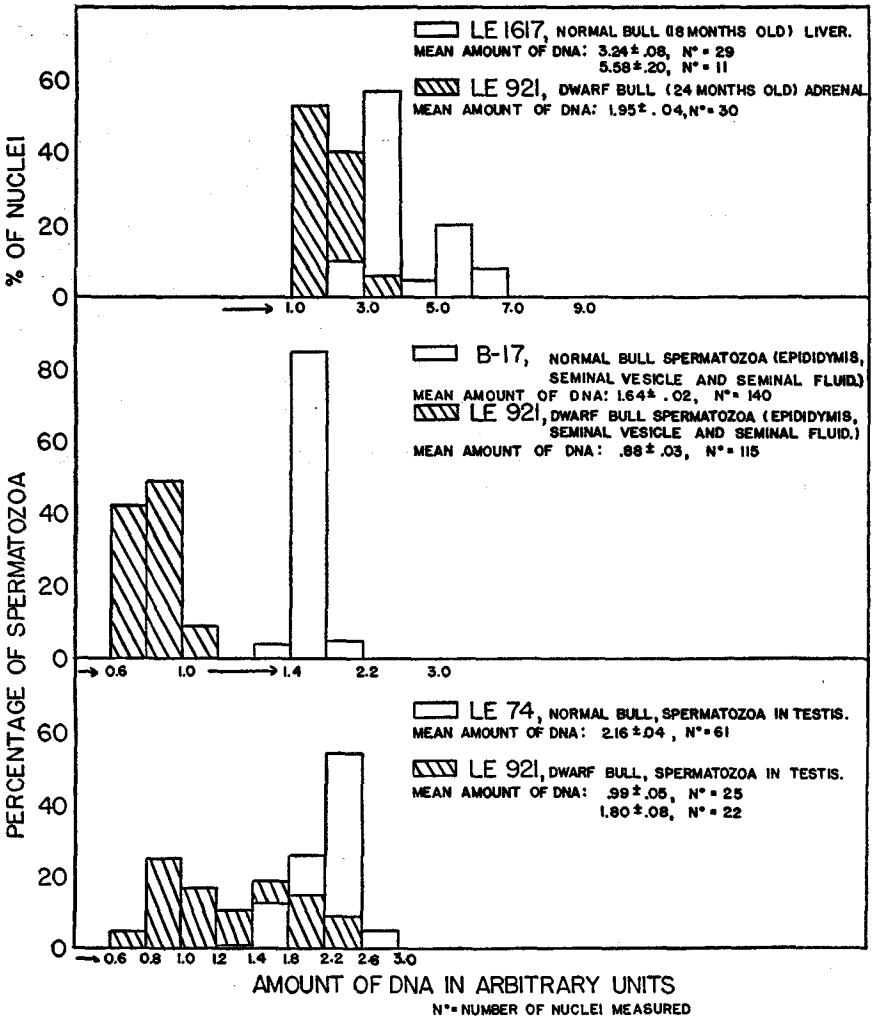

FIG. 1. Amount of DNA (microspectrophotometry) in individual nuclei of somatic cells and spermatozoa from normal and dwarf bulls.

tity of DNA was halved again. It is tempting to venture various explanations and interpretations of such an exceptional occurrence but in default of cytological data concerning the chromosome number and the meiotic behavior, and in view of our failure to obtain more material from the herd of cattle involved, such speculations would serve no useful purpose. However, it would seem worth while to put the case on record.

BIBLIOGRAPHY

1. Leuchtenberger, C., Schrader, F., Weir, D. R., and Gentile, D. P., *Chromosoma*, 1953, **6**, 61.
2. Leuchtenberger, C., Weir, D. R., Schrader, F., and Leuchtenberger, R., *Excerpta Med.*, 1954, **8**, sect. I, 418.
3. Leuchtenberger, C., Weir, D. R., Schrader, F., and Murmanis, L., *J. Lab. and Clin. Med.*, 1955, **45**, 851.
4. Leuchtenberger, C., Helweg-Larsen, H. F., and Murmanis, L., *Lab. Inv.*, 1954, **3**, 245.
5. Johnson, L. E., Harshfield, G. S., and McCone, W., *J. Hered.*, 1950, **41**, 177.
6. Leuchtenberger, C., *Chromosoma*, 1950, **3**, 449.
7. Leuchtenberger, C., The nucleoproteins in cell growth and division, *in* Statistics and Mathematics in Biology, (O. Kempthorne, T. A. Bancroft, J. W. Gowen, and J. L. Lush, editors), Ames, The Iowa State College Press, 1954, 557.
8. Pollister, A. W., and Ornstein, L., Cytophotometric analysis in the visible spectrum, *in* Analytical Cytology, (R. C. Mellors, editor), New York, The Blakiston Division, McGraw-Hill Book Co., Inc., 1955, 1.
9. Leuchtenberger, C., *Science*, 1954, **120**, 1022.