When approaches based on homeostatic single samples fail to discern the mechanism underlying variability both in health and disease (in the latter case chronopathology quantifiable possibly as a rhythm alteration), a spectral analysis completed on a reference sample of test individuals from populations at different risk for a given cancer (such as that of the breast) may yield information of interest to both students of oncology and of chronobiology.

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- H.J. Lynch, Y. Ozaki, D. Shakal and R.J. Wurtman, Int. J. Biometeorol. 19, 267 (1975).
- G.M. Vaughan, R.W. Pelham, S.F. Pang, L.L. Loughlin, K.M. Wilson, K.L. Sandoch, M.K. Vaughan, S.H. Koslow

- and R.J. Reiter, J. clin. Endocr. Metab. 42, 752 (1976).
- M.D. Rollag and G.D. Niswender, Endocrinology 98, 482 (1976).
- L. Hedlund, M.M. Lischko, M.D. Rollag and G.D. Niswender, Science 195, 686 (1977).
- L. Wetterberg, J. neural Transm., suppl. XIII, 289 (1978).
- L. Wetterberg, J. Arendt, L. Paunier, I.C. Sizonenko, W. van Donselaar and T. Heyden, J. clin. Endocr. Metab. 42, 185
- L. Wetterberg, O. Eriksson, Y. Friberg and B. Vangbo, Clin. chim. Acta 86, 169 (1978).
- F. Halberg, F. Carandente, G. Cornelissen and G.C. Katinas,
- Chronobiologia 4, suppl. I (1977).
 F. Halberg, Y.L. Tong and E.A. Johnson, in: The Cellular Aspects of Biorhythms, p.20. Symposium on Biorhythms. Springer 1967.
- E. Katagiri, Osaka Igakki Zasshi 43, 315 (1944).
- T.K. Das Gupta and J. Terz, Cancer Res. 27, 1306 (1967).
- R. Hofstätter, Krebsarzt 14, 307 (1959).
- R.J. Reiter, J. exp. Zool. 191, 111 (1975).
- L. Vollrath, A. Kantazjian and C. Howe, Experientia 31, 458
- 16 F. Halberg, M. Engeli, C. Hamburger and D. Hillmann, Acta endocr., suppl. 103, 5 (1965).

A study of the enzyme activity in the seminal vesicles of castrated and hormone-replaced castrated mice

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Summary. Castration provokes a time-related decrease in weight, protein, β -glucuronidase and glucose-6-phosphate dehydrogenase activity of seminal vesicles. A dose-dependent stimulation of these parameters is obtained with 5a-DHT. Cyproterone acetate counteracts the stimulatory effects due to androgen. Acid and alkaline phosphatases remain largely unaffected by these treatments.

It is well-recognized that the function of the seminal vesicles is completely androgen-dependent, and that castration and administration of the antiandrogenic steroid cyproterone acetate results in an atrophy of these glands (for bibliographical details see Neumann²). Information, however, is lacking upon the androgen-dependency of several biochemical parameters. A more detailed study of these might be of use for a better comprehension of the physiology of this accessory sex gland.

This work was undertaken to study the time-related effects of castration, effects of androgen-replacement therapy and those of the antiandrogen cyproterone acetate on the enzyme activity in mice seminal vesicles.

Adult male mice (Swiss albino race) weighing 25-30 g were used. Castration was done via the scrotal route. Groups of 5 animals were sacrificed at 7, 14, 21, 28 and 42 days after castration. At the 6-week postcastration term, groups of remaining mice received daily s.c. injections respectively of 5 µg 5a-dihydrotestosterone (DHT), 50 µg DHT, 50 µg DHT+0.5 mg cyproterone acetate (CPA) and maize oil vehicle. The hormone treatment was continued for 15 days before the mice were sacrificed. All animals in experimen-

Influence of castration, 5α-dihydrotestosterone and cyproterone acetate on weight, protein content and enzyme activity in seminal vesicles of mouse^a (values are means ± SEM for 5 determinations)

Experimental groups	Organ- somatic index	Protein (µg/mg tissue)	β-glu- curoni- dase	Acid phospha- tase	Alkaline phospha- tase	Glucose-6- phosphate dehydrogenase
Intact control Castrated	9.82 ± 0.32	12.1 ± 6.28	4.68±1.01	1.73 ± 0.67	2.79 ± 1.76	543 ± 222
7 days 14 days 21 days 28 days 42 days 42 days Castrated + maize oil Castrated + 5 µg DHT Castrated + 50 µg DHT Castrated + 50 µg DHT	5.10 ± 0.57^{c} 2.10 ± 0.34^{c} 1.47 ± 0.21^{c} 1.07 ± 0.27^{c} 0.33 ± 0.09^{c} 0.46 ± 0.11 1.75 ± 0.23^{c} 5.81 ± 0.44^{c}	12.9 ± 3.01 11.5 ± 7.60 5.8 ± 3.01 4.4 ± 1.90b 1.1 ± 0.29° 1.9 ± 0.50 11.1 ± 2.60° 63.6 ± 9.11°	2.18 ± 0.57° 3.33 ± 1.68 3.41 ± 0.34b 1.74 ± 0.84° 0.90 ± 0.04° 0.70 ± 0.28 6.00 ± 1.35° 6.28 ± 0.66°	1.00 ± 0.08^{b} 1.12 ± 0.22 1.67 ± 0.09 1.44 ± 0.17 2.14 ± 0.51 2.90 ± 0.20 2.76 ± 0.81 3.36 ± 0.56	2.51 ± 0.80 2.28 ± 0.52 3.34 ± 0.87 2.83 ± 0.30 4.23 ± 1.07 4.22 ± 0.74 4.72 ± 1.67 5.23 ± 2.02	126 ± 79° 209 ± 31° 135 ± 13° 66 ± 19° 5.1± 1.1° 7.1± 0.4 134 ± 35° 711 ± 114°

^aEnzyme activities expressed as nmoles paranitrophenol liberated/µg protein min for acid and alkaline phosphatases, as nmoles phenolphthalein liberated/ μg protein min for β -glucuronidase and as pmoles NADP reduced/ μg protein min for glucose-6-phosphate dehydrogenase. b, d Significantly different respectively from intact and castrated + oil controls (0.02 < p < 0.05). c, e Significantly different respectively from intact and castrated + oil controls (p < 0.01).

tation were caged in groups of 5 each with free access to food and water. Each animal was weighed and killed by an overdose of ether. Seminal vesicles were quickly dissected out, weighed and homogenized for enzyme assay. Acid phosphatase phosphatase (EC $3.\overline{1}.3.2.$), alkaline (EC 3.1.3.1.), β -glucuronidase (EC 3.2.1.31.; $\hat{\beta}$ -GLR) and glucose-6-phosphate dehydrogenase (EC 1.1.1.49.; G6PD) were assayed as described earlier^{3,4}. Proteins were determined according to Lowry et al.5. Results were analyzed for significance using Student's t-test⁶. Values of p<0.05 were considered significant. Small pieces of glands were also fixed in Bouin's fluid for routine histological examination. A progressive decline in the weight of seminal vesicles was found over the 6-week period of castration (table). The protein content showed a significant decline only at 4- and 6-week postcastration terms. G6PD activity decreased significantly already after 7 days, maintaining the reduced value almost constant over the following 2 weeks. Thereafter it declined further, reaching almost 1% of the control value. β -GLR activity decreased at 1-week postcastration term. It increased at 2-week postcastration term and decreased again to reach nearly 20% of the control value. Acid phosphatase activity showed a significant decline only during the first week of castration. Subsequently it rose and maintained the control range. In contrast, alkaline phosphatase activity did not manifest any influence of castration. The weight, protein content, β -GLR and G6PD were significantly increased at 5 µg DHT/day. Except for β -GLR, the stimulation was greater (almost 5-fold) at the

higher dose level (50 µg DHT/day). The 2 phosphatases did not show any change in their activity in the androgenreplaced castrated mice. DHT-induced stimulation of organ weight, protein, β -GLR and G6PD was almost nullified by the simultaneous injections of CPA. A histological examination of the seminal vesicles showed that castration caused a progressive atrophy of the gland and a gradual disappearance of the seminal plasma. Androgen therapy induced tissue hypertrophy and increased secretion of seminal plasma. These effects were inhibited by CPA. The present results confirm those of Neumann² and Luttge et al.⁷ as far as the histology and organ weight are concerned. The present data also indicate that β -GLR and G6PD activity in the seminal vesicles is under the control of androgen.

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- F. Neumann, Gynec. Invest. 2, 150 (1971/72).
- M. Milone, R.K. Rastogi and L. Iela, Gen. Comp. Endocr. 26, 107 (1975).
- M. Milone and R. K. Rastogi, J. Endocr. 70, 329 (1976).
- O.H. Lowry, N.J. Rosebrough, A.L. Farr and R.J. Randall, J. biol. Chem. 193, 265 (1951).
- R.A. Fisher, in: Statistical methods for research workers, 12th
- ed. Oliver & Boyd, London 1954.
 W.G. Luttge, T.W. Jasper, C.S. Sheets and H.E. Gray,
 J. Reprod. Fert. 53, 45 (1978).

PARAPHYSICA

Editorial remark. In a past paper the author reported about experiments to map fields of physiologically effective stimuli of an unidentified nature and natural origin. The present contribution attempts to produce stimuli with similar physiological effects by well defined and well located artificial sources. This complementary report might be of general interest in view of the assumptions of practising sourcerers about the external agents they are looking for. The author seems critical enough not to imply the unproven identity of the natural and the artificial sources which produced the similar stimuli.

H.M.

Experimental investigation of the perceptibility of the artificial source for the dowsing agent. Progress report

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Summary. An artificial source for dowsing experiments is described. Investigations on the perceptibility of this source by turning on and off the water flow gives significant statistical results. The experiments for locating the source compared with blank experiments equally reveal perceptibility. An unexpected hysteresis in the reactions is observed when changing the source conditions in time and space. The behaviour of the sensitivity of the operator is deduced from many locating experiments.

In a first paper² we reported on the experimentally established spatial distribution of the reaction locations over natural reaction zones in buildings, and started to discuss the influence of the sensitivity of the operator on the location of the reaction. It resulted from many experiments that the rod, wand or fork (or pendulum) is merely an indicator of an unconscious muscular reaction of the operator, and that this indicator instrument is individual as long training had been done with it. The paper concluded with the statement that the dowsing reaction was due to an unknown agent which, for convenience, had been called D-agent (D for dowsing). This statement is justified when taking into account the large spectrum of observations in this field.

Remarks concerning the possible nature of the dowsing agent. Different investigators³⁻⁶ demonstrated a local correlation between reaction zones and disturbances of the magnetic field of the earth; they were even successful in producing the unconscious muscular reaction by induced variations of the magnetic field or by electromagnetic waves7. But even if such stimuli produce a reaction similar to that observed in a zone over a ground water stream, it cannot be concluded that the stimulus of the latter must be of the same nature. Repeated tests with our operators revealed no sensitivity to disturbances of the magnetic field. However it seems possible to train an operator to respond to various known stimuli.

W.A. Tiller⁸ and A.M. Comunetti² plead for an unknown