Sebaceous gland atrophy in the rat after a portacaval shunt

F. Magne, P. Bioulac[1], P. Sartor, J. Saric and C. Balabaud

Laboratoire des Interactions Cellulaires, Physiopathologie, Pharmacologie et Centre de Microscopie Electronique, Université de Bordeaux II, F-33076 Bordeaux (France), 3 December 1980

Summary. In the rat, 2 weeks after a portacaval shunt, the epithelium thickness of the skin is decreased, and so are the size and the number of sebaceous glands. These results are compatible with the hyperoestrogenic state associated with a testicular atrophy, observed after portacaval shunt.

Estrogens decrease sebum production[2]. They reduce the epidermal thickness and the size of sebaceous glands[3]. Likewise castration decreases the sebum production and the size of sebaceous glands[4,5]. Rats with a portacaval shunt have a testicular atrophy[6–10] but it is not known whether or not they have a reduced level of testosterone. In this model we should therefore expect an atrophy of the sebaceous glands. The aim of the present study was to ascertain this particular point.

Material and methods. 10 male Wistar rats (270–310 g), bred in our laboratory with free access to normal chow diet and water had either a portacaval shunt[11] or a sham portacaval shunt which consisted of all the surgical steps exept the anastomosis. At the time of surgery, before the laparotomy, a skin biopsy (approximately 5×10 mm, including s.c. adipose tissue) was performed on the right side of the thorax, 1 cm above the xiphoid process. 2 weeks after the operation under ether anesthesia, another skin biopsy was taken in the same conditions on the opposite side and the liver and testes were removed for other studies. Histologic sections were carefully oriented perpendicular to the skin surface, stained with hematoxylin and eosin (H.E.) and examined blindly. To obtain more objective results, the surface content of sebaceous glands was measured by planimetry. For each biopsy, one photograph was taken at random in the field at a magnification $\times 6.3$ and enlarged to a final magnification $\times 110$. At this magnification, a tissue area of approximately 2 mm^2 (2×1) was analyzed. Each photograph included the epithelium, the dermis and the hypodermis as illustrated in figure 1. For each photograph, the total number of sebaceous glands was counted and the total surface of the glands measured using a semi-automatic planimeter (MOP AMO_2, KONTRON, FRG). Results were compared using Student's t-test for paired data, p-values below 0.05 were considered significant.

Results. The effects of portal blood shunting on body weight, liver weight and weight of testes is shown in the table. 2 weeks after PCS, there is a severe liver weight atrophy; at that time testicular atrophy is minimal.

At the time of sacrifice, in the group of sham operated rats 4 out of 5 had regrown hair in the area shaved for the laparotomy, and 1 had retardation in hair growth. In the group of portacaval-shunted rats 4 out of 5 were hair-less in the same area and 1 had some very short hair. By light microscopy, the skin of rats with PCS was different from sham PCS rats (figs 1, 2): the thickness of epithelium appeared to be decreased, fat depots were absent or sharply reduced, and collagen density seemed to be increased. It was also obvious that the number and the size of sebaceous glands had decreased. Sebaceous glands were approximately half as numerous and smaller. Number of glands per photograph and gland area are shown in the table.

Discussion. As previously shown for female pubertal rats[7], male rats do not regrow hair after PCS. The time of observation was too short to see any significant loss of their hair[7]. The loss of hair and probably the impaired regrowth of hair after PCS can be explained by the hyperoestrogenic state[12], the testicular atrophy with or without a reduced plasma level of testosterone[13] and to a certain extent by malnutrition[14] and a decreased level of LH[15,16]. Our impression was that the epithelium thickness was decreased but this needs to be confirmed by morphometric measurements. The reduced size and number of sebaceous glands was obvious and was confirmed by planimetry analysis. Increasing the number of animals assayed and/or of skin samples examined would have led to an improved estimate of the mean sebaceous gland area. However, with the large differences induced by the PCS, the number of rats used and the number of sebaceous glands counted were quite sufficient to obtain results with a high degree of statistical

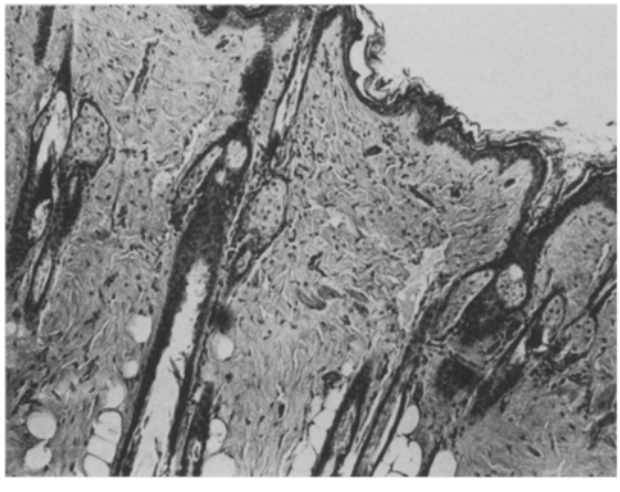

Figure 1. Skin biopsy of a rat before PCS. Normal aspect of skin histology. $\times 70$.

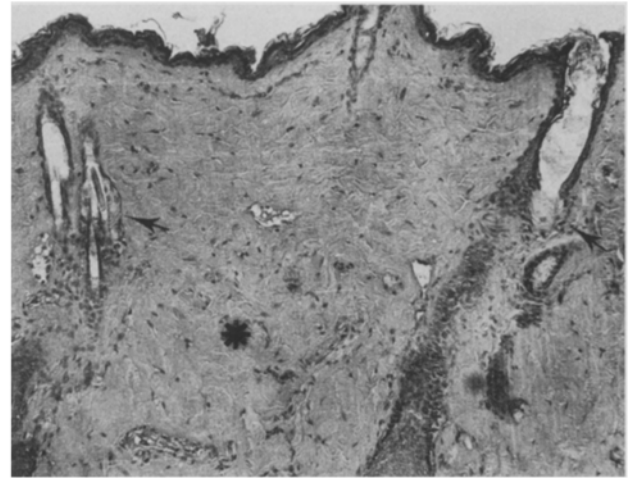

Figure 2. Skin biopsy of the same rat, 2 weeks after PCS. Compared with figure 1, sebaceous glands are less numerous and very atrophied (arrow), collagen density is increased (asterisk); subcutaneous fat depots are no longer present. $\times 70$.

Body weight, liver weight and area of sebaceous glands in rats with and without a portacaval shunt

N	Before PCS 5	After PCS 5	Before sham PCS 5	After sham PCS 5
Body weight (g)	307 ± 13[a]	241 ± 41[b]	291 ± 12	304 ± 18[c]
$\frac{\text{Liver weight}}{\text{Body weight}} \times 100$		2.15 ± 0.21		4.06 ± 0.35
$\frac{\text{Testes weight}}{\text{Body weight}} \times 100$		0.93 ± 0.07		0.99 ± 0.08
Number of sebaceous glands per micrograph	12.4 ± 2.7	6.2 ± 1.3[b]	10.8 ± 1.1	10.2 ± 2.1[c]
Area of sebaceous glands (mm^2)	598 ± 194	275 ± 88[b]	559 ± 170	567 ± 160[c]

[a] Mean ± ISD; [b] $p < 0.05$ vs the same group before PCS; [c] $p < 0.05$ vs the other group after PCS.

significance. The hyperoestrogenic state observed after PCS has several origins: among them, the porto-systemic shunt[17], the decreased hepatic blood flow[18], the increased conversion of androgen to oestrogens[19,10]. The atrophy of sebaceous glands after PCS is one more example of the importance of the skin as a hormonal receptor.

1 For reprint requests: P.B., Centre de Microscopie Electronique, Université de Bordeaux II, 146, Rue Léo Saignat, F-33076 Bordeaux Cedex (France).
2 F.J. Ebling, J. Endocr. *12*, 38 (1955).
3 F.J. Ebling, J. Embryol. exp. Morph. *5*, 74 (1957).
4 F.J. Ebling, J. Endocr. *15*, 297 (1957).
5 L.S. Sauter and A.V. Loud, J. Invest. Dermat. *64*, 9 (1975).
6 J. Bircher, Pharmac. Ther. *5*, 219 (1979).
7 P.J. Flynn and A.L. Kennan, Archs Path. *85*, 138 (1968).
8 R. Herz, V. Sautter, F. Robert and J. Bircher, Eur. J. clin. Invest. *2*, 390 (1972).
9 E. Dordal, S. Glacov and T.S. Debarros, Archs Path. *83*, 49 (1967).
10 M.H. Kyu and J.B. Cavanagh, Br. J. exp. Path. *51*, 217 (1970).
11 S. Lee, J.G. Chandler, C. Broelsch, Y.M. Flamant and M.J. Orloff, J. Surg. Res. *17*, 53 (1974).
12 F.J. Ebling, in: Dermatology in general medecine, p.126. Ed. T.B. Fitzpatrick, A.Z. Eisen, K. Wolff, I.M. Freedberg and K.F. Austen. Mac Graw-Hill Book Company, New York 1979.
13 L.J. Sholiton, L. Srivastava and B.B. Taylor, Steroids *26*, 797 (1975).
14 W.G. Pond, R.H. Barnes, R.B. Bradfield, E. Kwong and L. Krook, J. Nutr. *85*, 57 (1965).
15 S.K. Goolami, J.L. Burton and S. Shuster, Br. J. Dermat. *89*, 21 (1973).
16 P.E. Pochi and J.S. Strauss, Bull. N.Y. Acad. Med. (1975).
17 D.H. van Thiel, J.S. Gavaler, F.L. Slone, C.F. Cobb, W.I. Smith, K.M. Bron and R. Lester, Gastroenterology *78*, 81 (1980).
18 F.W. Ossenberg, P. Denis and J.P. Benhamou, J. appl. Physiol. *37*, 806 (1974).
19 G.G. Gordon, A.L. Southren, J. Vittek and C.S. Lieber, Metabolism *28*, 20 (1979).
20 J. Olivo, G.G. Gordon, F. Rafili and A.L. Southren, Steroids *26*, 47 (1975).

Retrograde axonal transport and transneuronal transference of horseradish peroxidase in the rat ciliary ganglion[1]

P. Gomez-Ramos and E.L. Rodriguez-Echandia

Dept. Morfologia, Facultad de Medicina, Universidad Autonoma de Madrid, Arzobispo Morcillo 4, Madrid 34 (Spain), 10 April 1981

Summary. Horseradish peroxidase was injected into the ciliary body of the eye, and 24 h later it was found in the perikaryon and dendritic processes of the ciliary ganglion neurons, as well as in nerve terminals presynaptic to those neurons. These results indicate that horseradish peroxidase was retrogradely transported and subsequently transneuronally transferred.

Substantial evidence exists for a bidirectional transfer of substances from one neuron to another. Transneuronal migration of radioactive material following anterograde axonal transport was first shown in the visual system[2] and later in several other systems[3-5]. Radioactive tetanus toxin has been shown to travel transneuronally following retrograde axonal transport[6,7]. The present report presents some evidence of transneuronal transfer of horseradish peroxidase (HRP) a protein transported intraneuronally mainly in the retrograde direction[8-10] but also anterogradely[11,12]. Preliminary results have been presented elsewhere[13].

10 male albino rats (200–250 g) of the Sprague-Dawley strain were used. In 8 animals, 1 mg of HRP (Sigma type VI) in 3 µl of distillated water was injected into the ciliary body of one eye under light ether anesthesia. 2 control animals were injected with distilled water. The animals were sacrificed 24 h after the injection, and perfused through the heart with 250 ml of 1% paraformaldehyde and 1% glutaraldehyde in 0.12 M phosphate buffer (pH 7.2). The brains and the ciliary ganglia homolateral to the injected eye were dissected out and immersed in the same double aldehyde fixative at 4 °C for 4–6 additional h. The material was soaked overnight in 0.12 M phosphate buffer containing 15% sucrose at 4 °C and the peroxidase activity was determined by the method of Graham and Karnovsky[14]. The ciliary ganglia were post-fixed in 1%