

cells in the presence of insulin, transferrin and fibroblast growth factor (4F medium). This growth-promoting activity has been characterized as kallikrein-like protease(s). These results suggest that adipose precursor cells might be under the control of proteolytic events for their expansion through cell multiplication.

In addition to growth hormone (GH) and triiodothyronine ( $T_3$ ), fetuin, a bovine fetal protein (also present at high concentrations during embryogenesis in mammals), and a serum-derived adipogenic fraction (SDAF) are required to promote adipose conversion of Ob17 cells. In the presence of insulin, GH,  $T_3$  and fetuin, cells are able to express only early events of adipose conversion, such as the emergence of lipoprotein lipase (LPL), but remain unable to express late markers, such as glycerol-3-phosphate dehydrogenase (GPDH), and to accumulate triglycerides. SDAF is strictly required to induce a limited cell proliferation which is accompanied by the expression of the late markers of adipose conversion (terminal differentiation). Both events are strictly controlled by cAMP and modulated by messengers generated through inositolphospholipid breakdown.

Arachidonic acid, as a promoter of both the cAMP and the inositolphospholipid pathways, behaves as a mitogenic—adipogenic stimulus and is indeed able to substitute for SDAF and to induce terminal differentiation. Recent developments of these studies have allowed the characterization of prostaglandins, (prostaglandin ( $PGI_2$ ) and  $PGF_{2\alpha}$ ) as physiological effectors of this process *in vitro* and to propose that these metabolites could play a critical paracrine/autocrine role in the development of adipose tissue *in vivo*.

**Differentiation of rabbit adipocyte precursors in primary culture.** J. Nougues, Y. Reyne and J.P. Dulor (*INRA-ENSA, pl. Valia, 34060 Montpellier Cedex, France*)

The differentiation of adipocyte precursors derived from the stromal—vascular fraction (SVF) of rabbit perirenal adipose tissue was studied using two primary culture systems. Adipose conversion was assessed by the development of glycerol-3-phosphate dehydrogenase, acid: CoA ligase and lipoprotein lipase activities.

*Culture in a medium supplemented with serum or plasma.* Perirenal SVF cells from rabbit fetuses or 4 week old rabbits were not able to differentiate when maintained in a medium containing fetal calf or rabbit serum. In contrast, differentiation was induced when the cells were grown in a medium supplemented with rabbit plasma. The higher density of plasma—cultured SVF cells at the time of differentiation suggests that plasma has a positive effect on the post-confluent mitoses of susceptible cells. Mesenteric lymph or chylomicrons, added to the medium as lipid sources, enhanced greatly both lipid accumulation and enzymatic activities of differentiating cells.

*Culture in a chemically defined medium.* As rat adipocyte precursors are able to differentiate in a serum-free hormone-supplemented medium (ITT medium) containing insulin, transferrin and triiodothyronine (Deslex *et al.*, 1987; *Exp. Cell Res.* 168, 15—30), we tested this medium with SVF cells from the perirenal adipose tissue of 4 week old rabbits. ITT medium failed to induce adipose conversion or to a very low extent. Supplementation with growth hormone or fibroblast growth factor did not increase the proportion of differentiated cells. In contrast, 20—50% of SVF cells were able to differentiate in the presence of glucocorticoids (dexamethasone, corticosterone) within 2 weeks of culture. Sex steroids ( $\beta$ -estradiol, testosterone, progesterone) did not affect the differentiation process. The stimulatory actions of dexamethasone or insulin were dose-dependent and adipose conversion could be obtained at physiological concentrations of insulin and glucocorticoids. Insulin like growth factor-I was not able to replace insulin under our culture conditions and had only a slight effect when added along with dexamethasone and physiological concentrations of insulin.

In conclusion, glucocorticoids, in association with insulin, may play an important role in the development of rabbit adipocyte precursor cells.

**Adipose tissue metabolism and development in cold-acclimated piglets.** P. Herpin <sup>1</sup>, R. Bertin <sup>2</sup>, J. Le Dividich <sup>1</sup>, F. de Marco <sup>2</sup> and O. Douillet <sup>1</sup>

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An experiment involving 5 x 20 piglets was designed to examine the effects of chronic cold exposure (12°C for 3 weeks) on intermediate metabolism of lipids and plasma hormonal levels in order to assess the mechanisms involved in thermo-regulation of early-weaned piglets. The control piglets were maintained in a thermoneutral environment (24°C).

Cold-acclimated piglets fed *ad libitum* maintained a growth rate and a carcass composition similar to that of the controls, while increasing feed intake by 20% ( $P < 0.05$ ).

Lipoprotein lipase activity increased ( $P < 0.05$ ) by cold exposure in white adipose tissues (WAT) and heart. A large enhancement of lipogenesis was observed in WAT and, to a lesser extent, in the liver, while chemical composition of these tissues and fatty acid composition of the lipid extracts did not change significantly in the cold. However, a difference in the composition of internal and external WAT of control pigs was observed. These results together with the increased noradrenaline-induced lipolysis suggest an increase in fatty acid turnover in cold-acclimated piglets.

Urinary catecholamines (noradrenaline, adrenaline) and plasmatic thyroid hormones ( $T_3$ ,  $T_4$ ) gradually increase during cold acclimation and are defined as effectors of the thermogenesis. No changes appeared in cortisol, ACTH and insulin levels.

Our results suggest that early-weaned piglets adapt themselves to cold conditions by their increasing thermogenic capacities (increased feed consumption and heat production) and reducing heat losses (modifications of conformation), by a stimulation of lipid metabolism (lipogenesis, lipolysis, LPL activity) in white adipose tissue and a modification of the hormonal status (catecholamines and thyroid hormones levels). Enhancement of lipid metabolism in white adipose tissue is discussed in terms of cold-acclimation mechanisms.

**Biophysical and biochemical study of the plasma membrane of pig adipose cell.** C. Lhuillery <sup>1</sup>, C. Nicolas <sup>1</sup>, N. Benmansour <sup>1</sup>, D. Lacasa <sup>2</sup>, Y. Demarne <sup>1</sup>  
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In recent years, it has been stated that many functions of living cells are membrane-associated events. In various cell types, functional alterations have been related to membrane structural modifications. Little information is yet available concerning structure—function relationships in adipose cell membranes.

The objective of this study was : 1) to define the structural characteristics of adipose plasma membrane in the pig, and 2) to investigate the effects of diet, castration and adipose tissue location on these parameters. Castrated or male pigs were fed either a control diet or a polyunsaturated fatty acid-enriched diet (sunflower diet). In the different situations, adipose cells were collected from subcutaneous (S) and perirenal (P) tissues and a plasma membrane-enriched fraction was prepared on which structural and functional analyses were performed.

It was shown that the lipid matrix constituents of plasma membrane (cholesterol (CHOL), fatty acids (FA), and phospholipids (PL) could be modified by diet, castration and the origin of adipose tissue (P or S). Thus, the CHOL/PL ratio was increased by castration (regardless of the tissue) and increased by the sunflower diet (only in P tissue).

The degree of PL fatty acid insaturation increased with the sunflower diet and decreased by castration. Major modifications of PL classes concerned phosphatidyl choline (PC), phosphatidyl éthanolamine (PE) and sphingomyelin (SM) levels.

Membrane fluidity as assessed by fluorescence polarization studies was higher in S than in P membranes in the basic situation (male pigs fed the control diet) and increased in both tissues when animals were fed the sunflower diet.