Brown Adipose Tissue

Edited by OLOV LINDBERG

ELSEVIER·NEW YORK·LONDON·AMSTERDAM

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OLOV LINDBERG

The Wenner-Gren Institute
University of Stockholm
Stockholm, Sweden

1970

AMERICAN ELSEVIER PUBLISHING COMPANY, INC.

NEW YORK

AMERICAN ELSEVIER PUBLISHING COMPANY, INC.
52 Vanderbilt Avenue, New York, N.Y. 10017

ELSEVIER PUBLISHING COMPANY LTD.
Barking, Essex, England

ELSEVIER PUBLISHING COMPANY
335 Jan Van Galenstraat, P.O. Box 211, Amsterdam, The Netherlands

International Standard Book Number 0-444-00080-1
Library of Congress Card Number 75-116710

PRINTED IN THE UNITED STATES OF AMERICA

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ADIPOSE TISSUE

Contributors

BJÖRN A. AFZELIUS *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 1–31)

TUDOR BARNARD *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 33–72)

KURT BRÜCK *Physiologisches Institut der Universität Marburg, Marburg an der Lahn, Germany* (*pp*. 117–154)

BARBARA CANNON *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 283–318)

W. H. COTTLE *University of Alberta, Edmonton, Alberta, Canada* (*pp*. 155–178)

ZDENEK DRAHOTA *Institute of Physiology, Czechoslovak Academy of Sciences, Prague, Czechoslovakia* (*pp*. 225–244)

R. L. DRYER *Department of Biochemistry, University of Iowa, Iowa City, Iowa* (*pp*. 197–206)

JOHN N. FAIN *Division of Biological and Medical Sciences, Brown University, Providence, Rhode Island* (*pp*. 207–224)

MARTIN J. HARDMAN *Nuffield Institute for Medical Research, University of Oxford, Oxford, England* (*pp*. 97–115)

KARL J. HITTELMAN *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 245–262)

Present address: Laboratory of Chemical Biology, Department of Zoology, University of Michigan, Ann Arbor, Michigan

DAVID HULL *Nuffield Institute for Medical Research, University of Oxford, Oxford, England* (*pp*. 97–115)

Present address: The Hospital for Sick Children, London, England

OLOV LINDBERG *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 245–262, 283–318)

K. G. MANN *Department of Biochemistry, University of Iowa, Iowa City, Iowa* (*pp*. 197–206)

Present address: Department of Biochemistry, Duke University, Durham, North Carolina

J. R. PAULSRUD *Department of Biochemistry, University of Iowa, Iowa City, Iowa* (*pp*. 197–206)

Present address: The Hormel Institute, University of Minnesota, Austin, Minnesota

MARTIN POE *Johnson Research Foundation, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania* (*pp*. 263–282)

STANLEY PRUSINER *Johnson Research Foundation, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania* (*pp*.263–282, 283–318)

NORA REED *Division of Biological and Medical Sciences, Brown University, Providence, Rhode Island* (*pp*. 207–224)

E. SCHÖNBAUM *University of Toronto, Toronto, Ontario, Canada* (*pp*. 179–196)

E. A. SELLERS *University of Toronto, Toronto, Ontario, Canada* (*pp*. 179–196)

JOSEF SKÁLA *The Wenner-Gren Institute, University of Stockholm, Stockholm, Sweden* (*pp*. 33–72)

On leave of absence from Faculty of Pediatrics, Charles University, Prague, Czechoslovakia

Present address: Faculty of Medicine, University of British Columbia, Vancouver, B.C., Canada

ROBERT L. SMALLEY *Kansas State Teachers College, Emporia, Kansas* (*pp*. 73–95)

G. STEINER *University of Toronto, Toronto, Ontario, Canada* (*pp*. 179–196)

Preface

Biologists have recently shown great interest in brown adipose tissue. Some of this activity has stemmed from the hope that a new material might release them from the frustrations encountered in previous work with more or less standard material, but part of the interest also arises from the study of a new system, the function of which was completely unknown until recent years. The rationale for the latter point of view is based on the unique anatomical features of brown fat and its specialized physiology, since the tissue is a powerful heat source under conditions of lowered muscular activity. This heat production is commonly described as nonshivering thermogenesis. We find here a simplified biochemical system suited for detailed enzymatic studies.

The progress of biology has frequently been intimately connected with the discovery of a suitable material for investigation. Pure bacterial strains, tissue cultures, the sea urchin egg, and the electric organ are all examples of materials on which fundamental studies have been made that provided insights of general biological applicability. It may sound paradoxical that it is the very specialized nature of these systems that makes them so suitable for studies of generalized importance, but the complex nature of biology causes the vast number of reactions that take place in less specialized cells to overshadow the process which may be of particular interest.

Twenty years of research on isolated mitochondria has contributed a host of information about this subcellular organelle and its reactions. Our knowledge of the adjustments or control of these reactions in the environment of the intact cell is, however, more a deduction from results on isolated mitochondria than from direct observations and experiments on intact cells or tissues. When, about twenty years ago, the mitochondrion became a favorite material of so many biologists, not the least reason was that these organelles contained the entire mechanism for a whole metabolic sequence, that of energy conservation. Even a brief glance at sections of brown adipose tissue in the electron microscope suggests that the metabolism of the mitochondria may dominate the biochemistry of the adipocytes. It now emerges that the role of the adipocyte of brown adipose tissue is to transduce the energy stored in fatty acids into heat, this process occurring in the mitochondria. Thus it is reasonable to assume—and experimental results support this—that many functional studies of brown fat adipocytes are in fact studies on the metabolism of mitochondria in a relatively undisturbed environment.

How the cell regulates its metabolism is a subject of interest for the biochemist as well as the physiologist and the clinician. As far as regulation of energy transduction is concerned, almost all hypotheses are based on the concept of so-called respiratory control. In this process the need of the system for covalent bonds with a high energy of hydrolysis controls the respiration of the cell. In a heat production which is not based upon the energy-waste of overall cellular metabolism, such bonds are not theoretically needed. Consequently we face the possibility of finding in brown fat an entirely new mechanism of metabolic control that has so far been studied only very little, and which can be expected to take place at least to some extent in other cells as well. The ability to isolate and stimulate the brown fat adipocyte with physiological agents, coupled with the possibility of studying such systems by means of the many sophisticated techniques developed in connection with studies of isolated mitochondria, forms a solid methodological foundation for this area of cellular research. Even more important, however, is the generosity of nature, which has provided us with a highly specialized cell that can be isolated in quantity, stimulated with hormones and neurotransmittors, and that can perform all the steps in one dominating sequence of reactions that starts from the very beginning of the receptor system and ends with the final end products of fatty acid oxidation.

The articles on brown adipose tissue presented in this volume have been selected to illustrate some of the problems, at different levels of organization, that can be studied on this newly exploited material.

OLOV LINDBERG

Stockholm, Sweden
Fall 1969

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CHAPTER 1

Brown Adipose Tissue:

Its Gross Anatomy, Histology, and Cytology

BJÖRN A. AFZELIUS

Introduction

Various roles have been assigned to the brown adipose tissue. Throughout the 300 years that it has been a subject of investigation, the prevailing opinions have shifted. Reviewers, such as Rasmussen (1) and Johansson (2), have recognized four main periods with respect to the interpretation of the tissue:

1. 1670–1817. Brown adipose tissue is regarded as part of the thymus.
2. 1817–1863. Brown adipose tissue is believed to be an endocrine gland, and by some authors an organ active in the formation of blood.
3. 1863–1902. Brown adipose tissue is considered to be a modified form of fat tissue serving as a reservoir for food substances.
4. 1902——. Brown adipose tissue is again believed to be an endocrine organ.

It is now possible to add a fifth period and at the same time to give an end date, 1961, to the fourth. Thus:

5. 1961——. Brown adipose tissue is regarded as a thermogenic effector, that is, a tissue with the function of heating the blood passing through it.

Smith (3) and Ball and Jungas (4) were the first to demonstrate clearly that the brown adipose tissue is the site of "nonshivering thermogenesis." They have pointed out the fundamental difference between the heat-insulating and fat-storing properties of the white adipose tissue and the heat-producing capacity of the brown adipose tissue.

Their discovery has greatly stimulated research in the biochemistry and physiology of the brown adipose tissue. This can be witnessed by the vastly increased number of publications on various aspects of this tissue. Articles on the gross and fine anatomy of the brown adipose tissue have hardly kept

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pace with the physiological ones. This is partly because good anatomical descriptions already exist. Previous reviews on the morphology of brown adipose tissue include those by Rasmussen (1), Rémillard (5), Fawcett (6), Barrnett (7), and Umahara (8).

The term "brown adipose tissue" seems to be the one in most common use. It will hence be applied here. Other terms used since 1920 for the same tissue include: hibernation gland (French: *glande hivernale*, German: *Winterschlafdrüse*, Italian: *ghiandola dell'ibernazione*), brown fat, brown tissue, brown body, multilocular adipose tissue, plurivacuolar fat tissue, glandular adipose tissue, lipoic gland, cholesterin gland, embryonal fat, immature adipose tissue, adipose tissue in formation, primitive fat. The profusion of terms reflects an uncertainty in the evaluation of the tissue. Occasionally, separate lobes of the brown adipose tissue have been described as endocrine glands and given such names as "glandula insularis cervicalis." The portion of the brown adipose tissue that has been studied the most is known as the interscapular fat, interscapular gland, etc. Brown adipose tissue is often abbreviated "BAT." Its cells are called brown adipocytes.

The other main adipose tissue in mammals is the white adipose tissue; this has also been called yellow adipose tissue, unilocular adipose tissue, and adult fat.

Gross Anatomy

Hedgehog

References (9) to (16) contain information on the anatomy of brown adipose tissue from the hedgehog, *Erinaceus europaeus*, either at the gross, histological, or fine structural level.

In the paper by Suomalainen and Herlevi (13) the brown adipose tissue is described as a "bilateral formation which extends over the neck, axillary region and anterior part of the back. Its lobes extend to the mediastinum and diaphragm and surround the large blood vessels of the thoracic cavity." This description is in agreement with one of the classical treatises on the hibernation, a monograph by Barkow (9), in which the term *Winterschlafdrüse* was introduced as well as the following terms for its different portions in the hedgehog: pars thoracica (along the breast bone), pars cervicalis (along the neck muscles), pars axillaris (beneath the key bones), and pars dorsalis (extending on the back from the level of the first thoracic vertebra). These terms are still common usage after a hundred years, except that the term "interscapular" has replaced the term "dorsalis." It is remarked both by Barkow (9) and Zirm (14) that the different portions of the organ are connected to each other by means of narrow bridges and hence form a unit or a single paired organ. There is no brown adipose tissue in the kidney or groin regions (12).

The brown adipose tissue makes up a substantial part of the hedgehog's body. Hoepke and Nikolaus (12) estimated that it amounts to 3.8% of the total body weight in an animal at the onset of hibernation (November) and 1.2% towards the end of hibernation (May). Corresponding figures given by Carlier (10) are 3.0% (October) and 0.9% (March). In June the tissue may be reduced to a few fibrous cords. The body weight of an adult hedgehog is 0.6–1.5 kg.

Bats

The brown adipose tissue of the bat seems to be largely confined to the two interscapular lobes, which meet at the dorsal midline. From the lateral angles of the lobes there are thin lateral arms curving around the sides of the neck. The appearance of the tissue is shown in Fig. 1, which is reproduced from the

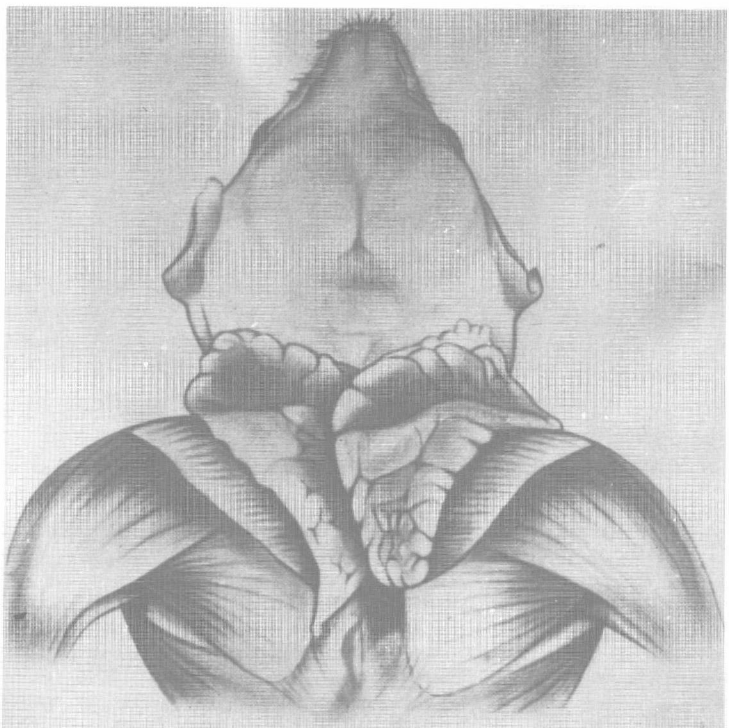

Fig. 1. Brown adipose tissue from the bat *Myotis lucifugus*. From G.-L. Rémillard. Annals of The New York Academy of Sciences, Volume 72, Article 1, Feb. 4, 1958.

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article by Rémillard (5). Rémillard has remarked that the brown adipose tissue of this species, *Myotis lucifugus lucifugus*, is salmon colored during hibernation and deep red during the awakened period.

In the adult big brown bat, *Eptesicus fuscus*, taken after 5 months of intermittent hibernation, the body weight averaged 14.3 g, and the brown adipose tissue 0.47 g, or 3.3% (17).

The Japanese bat species *Rhinolophus ferrum-equinum nippon*, studied by Umahara (8), lives in a much warmer climate than the Canadian species examined by Rémillard. The fat of the Japanese species, in contrast to the Canadian one, was found to have a biphasic seasonal weight curve with a period of fat depletion in March, corresponding to arousal from hibernation, and another even more pronounced period of fat depletion in August. This second minimum was correlated with a possible resting stage of the brown adipose tissue in the season of high and constant temperature.

The tropical fruit bat *Artibeus jamaicensis* has brown adipose tissue with unique histological properties to be described below in the section on microscopic anatomy (18). A comparison between the brown and the white adipose tissue of bats in histological and histochemical properties has been given by George and Eapen (19).

Rat

The rat *Rattus norvegicus* has been used in many morphological studies on the brown adipose tissue (6, 16, 20–45). The most complete description of the gross anatomy is given by Hammar (20). He stressed that masses of brown adipose tissue are found in many parts in the rat thorax and abdomen. In contrast to the situation in the hedgehog with its continuous, lobed "hibernation gland," there are several separate portions of the tissue in the rat abdomen: between the kidneys and penetrating the kidney hilus, surrounding the abdominal aorta along the foremost parts of the ureters, and in the groin region. The thorax region contains larger masses of brown adipose tissue. The largest portions are the paired masses of interscapular fat situated in a depression of the muscles in the middorsal line between the scapulae. Continuous with these masses are the smaller axillary portions and extensions between superficial and deep muscle layers. Most intermuscular fat is regarded by Hammar as brown adipose tissue. Other masses are found surrounding the descending aorta, within the mediastinum, and along the breast bone. There are also cervical portions appearing as thin strands along the front part of the neck. Hammar's description based on the laboratory rat is in agreement with a more recent report based on conditions on wild rats (26).

Brown adipose tissue amounts to 0.75–4.35 g; this according to Hammar, is equivalent to 0.60–1.65% of the body weight. The variation between different animals is said to be smaller than that of the white adipose tissue (0.53–8.49%). In contrast to the case in hibernators such as the hedgehog, there are no seasonal fluctuations (20, 26). When rats are kept in the cold (0–4°C), the amount of brown adipose tissue more than doubles (23, 33, 35, 37). Pagé and Babineau (23) estimated the increase in fat-free dry matter from the brown adipose tissue to be 70–80% after cold-acclimation. The increase in mitochondrial mass is proportionally still higher according to Smith *et al.* (38), or about 350% as expressed in mitochondrial nitrogen. In a cold-adapted rat weighing about 170 g it is possible to dissect out about 0.9–1.2 g of interscapular brown adipose tissue.* Cold-adapted rats have become a material of choice for students of brown adipose tissue.

The color and appearance of brown adipose tissue from well-fed rats is reminiscent of that of fish liver (20). Fat-depleted brown adipose tissue is dark red-brown, like a blood-filled rat liver. This color is attributed to the blood, as the isolated adipose cells are a light tan, appearing similar to a suspension of liver mitochondria (39).

Mouse

There are several papers containing information on the cytology or histochemistry of the brown adipose tissue from the mouse *Mus musculus* (16, 30, 36, 46–49). Descriptions of its gross anatomy seem more scanty. It appears that the anatomical distribution of brown adipose tissue in mice is not unlike that in rats. It amounts to 1–1.2% of the newborn mouse, which weighs about 1.4 g.

Hamsters

The Syrian hamster, *Mesocricetus auratus*, is a hibernator and can be bred in animal houses or obtained from pet shops. In these respects it seems unique. It is possible to select superhibernating and nonhibernating lines of Syrian hamsters (50). It is not surprising that the Syrian hamster has been used in investigations on the "hibernating gland" (15, 51–55).

Figure 2 shows the topography of the brown adipose tissue. As Aronson and Shwartzman (51) pointed out, the major portion of the brown adipose tissue is ventral to and between the scapulae, extending from the lower cervical level to the midthoracic plane. Other lobes extend beneath the scapulae and occupy the axillary apices, and thin extensions project rostrally between the posterior cervical muscles. There are only minute amounts around the thymus and the thyroid gland. The sympathetic nervous chain is surrounded by masses of brown adipose tissue in its thoracic course, but there is no other thoracic brown adipose tissue. The adrenal glands are sheathed by

*Personal communication, 1969: B. Cannon and A. Bergström, Wenner-Gren Institute, Stockholm.

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