A STEREOTAXIC ATLAS
OF THE
NEW ZEALAND RABBIT'S BRAIN

A STEREOTAXIC ATLAS OF THE NEW ZEALAND RABBIT'S BRAIN

By

IVAN URBAN
Rudolph Magnus Institute for Pharmacology
University of Utrecht
Utrecht, The Netherlands
formerly
Department of Anatomy
The Medical School
Bristol, England

PHILIPPE RICHARD
Institute de Physiologie and de Biochimie
Faculté des Sciences
Strasbourg, France
formerly
Laboratoire de Physiologie de la Lactation
Institut National de la Recherche Agronomique C.N.R.Z.
78 Jouy-en-Josas, France

CHARLES C THOMAS · PUBLISHER
Springfield · Illinois · U.S.A.

Published and Distributed Throughout the World by
CHARLES C THOMAS • PUBLISHER
BANNERSTONE HOUSE
301-327 East Lawrence Avenue, Springfield, Illinois, U.S.A.
NATCHEZ PLANTATION HOUSE
735 North Atlantic Boulevard, Fort Lauderdale, Florida, U.S.A.

ISBN 0-398-02431-6
Library of Congress Catalog Card Number: 76-184615

With THOMAS BOOKS *careful attention is given to all details of manufacturing and design. It is the Publisher's desire to present books that are satisfactory as to their physical qualities and artistic possibilities and appropriate for their particular use.* THOMAS BOOKS *will be true to those laws of quality that assure a good name and good will.*

Printed in the United States of America
P-4

Contents

A STEREOTAXIC ATLAS
OF THE
NEW ZEALAND RABBIT'S BRAIN

Introduction

HORSLEY AND CLARKE's invention of a stereotaxic instrument in 1908 (14) meant an introduction of a new three-dimensional way of locating intracranial structures. Their original idea relied on the assumption that there exists more or less constant relationship between intracerebral and skull structures in a particular animal species. Therefore, if one could select some prominent skull landmarks, preferably those less susceptible to individual variation of skull dimensions, as for example, meatus acusticus externus, margo infraorbitalis, and sutura sagittalis, one would be in a position to create a three-dimensional system of perpendicular basal planes defined by these landmarks. The location of any intracerebral structure could then be expressed in terms of its relationship to basal planes. According to these principles, Horsley and Clarke constructed the first stereotaxic instrument the main function of which was to orient the tools for intracranial intervention (an electrode etc.) into established basal planes. To achieve the correct orientation the head of the experimental subject was firmly fixed in the instrument in a manner securing identification of basal cranial planes with those of the apparatus. Favorable configuration of the chosen landmarks largely enhanced such fixation simply performed by insertion of a pair of horizontal ear bars into likewise oriented auditory meatus combined with a pair of infraorbital bracket bars pressing against infraorbital margins. In addition, such clear definition of fixation provided its good reproducibility within and between subjects.

Despite its progressive character, it was not until 1932 that the potential of the Horsley-Clarke method became fully understood due to the work of Ranson and his co-workers. These authors used the, until then, scarcely used stereotaxic technique for an extensive research of the cat diencephalon and the brain stem (15, 16), and the results achieved by this technique stimulated its further expansion and general acceptance in the research. Although the application of the method was initially restricted to the cat and the monkey for relative homogeneity of skull dimensions, the growth of brain research in the years that followed soon resulted in adaptation of stereotaxy to other animals, including birds. Of course, certain modifications had to be introduced in order to meet the requirements of new species, but basically the Horsley-Clarke concept of skull landmarks remained unchanged.

From recent articles reviewing the history and the development of the stereotaxic method (3, 7, 29), it became apparent that the rabbit, despite its popularity as an experimental subject, was not readily accepted into the family of animals to which the method was suited. The peculiar configuration of its auditory meatus—crucial landmarks and fixation points—was one of the main reasons. While in most animals auditory meatus are oriented horizontally and thus allow an easy insertion of straight horizontal ear bars, in the rabbit their longitudinal axes show a substantial deviation from the horizontal plane. Consequently, it was practically impossible to use conventional biaural head fixation (and also the corresponding Horsley-Clarke system of basal planes) unless some modifications were introduced.

There have been several attempts to solve the problem by seeking different ways to fixate the head in specially designed instruments; for example, against zygomatic arcs and front incisors. Bregma and Lambda skull sutures then served as the landmarks for definition of the stereotaxic planes. Harris was probably the first to use this approach in the rabbit for the purpose of implanting electrodes into the hypothalamus (10). However, a stereotaxic atlas based on these principles did not appear until 1954, compiled by Sawyer, Everett, and Green (30), and was later followed by that of Fifková and Maršala (5). It might be pertinent to mention in association with the latter technique that zygomatic arcs-front incisors fixation requires certain experience to master it and that the accuracy in reaching intracerebral targets, using the Bregma-Lambda reference system, was found by the present authors to be satisfactory only within the forebrain.

An alternative solution was described by Monnier and Gangloff in 1961 (20). These authors took advantage of a streotaxic plate originally designed by Hess (13) for multiple electrode implantations in the conscious cat and adapted its dimensions to the rabbit's skull. Similarly, as in the previous method, skull sutures were taken as the basic landmarks. However, while such approach obviated the need for a stereotaxic head holder, it was not suited for those experiments requiring restraint of the experimental subject.

So far, the solution reported by Chatelier and Buser (4) appears most attractive. The authors modified conventional horizontal ear bars according to the oblique angle of the rabbit's auditory meatus with the aim to facilitate their adequate insertion into the meatus for achievement desired rigid head fixation. Such modification, completed by an attachment for infraorbital fixation, permitted the adoption of classical landmarks with unaltered Horsley-Clarke basal planes defined as follows: the horizontal plane going through the middle of auditory meatus and infraorbital margins, the frontal plane intersecting perpendicularly the horizontal one in the center of the auditory orifices, and the sagittal plane being simply determined by sagittal suture. In addition, Chatelier and Buser technique circumvents the need for specially designed instruments, since any universal stereotaxic frame could be used, provided that the proper attachments are available.

We have adopted this approach because of the advantages it offers are simple and sufficiently accurate and because it provides more reliably reproducible head fixation than the methods mentioned above. In order to make full use of it we have prepared a corresponding stereotaxic atlas.

Materials and Methods

THIRTY-THREE one-year-old female New Zealand rabbits, weighing about 4.5 kg were used for preparation of the atlas. The animals, under urethane anesthesia (1.75 gm/kg), were biaurally mounted in the stereotaxic instrument (La Precision Cinematographique, Paris) provided with modified ear bars and the attachment for infraorbital fixation (for details see Chatelier and Buser [4]). Stainless steel needles were introduced into the brain bilaterally in frontal planes starting at A 0.0 mm and subsequently rostralwards at A 2.0, A 5.0, A 10.0, A 15.0, and A 20.0 mm (A 0.0 mm representing the middle of the auditory orifices—see Fig. 1) always 2 mm from the midline. The same procedure was repeated in the horizontal plane at H 2.0 mm and H 4.0 mm above H 0.0 mm on the left and at H 2.0 mm, H 4.0, and H 6.0 mm above H 0.0 mm on the right side (see Fig. 1). For

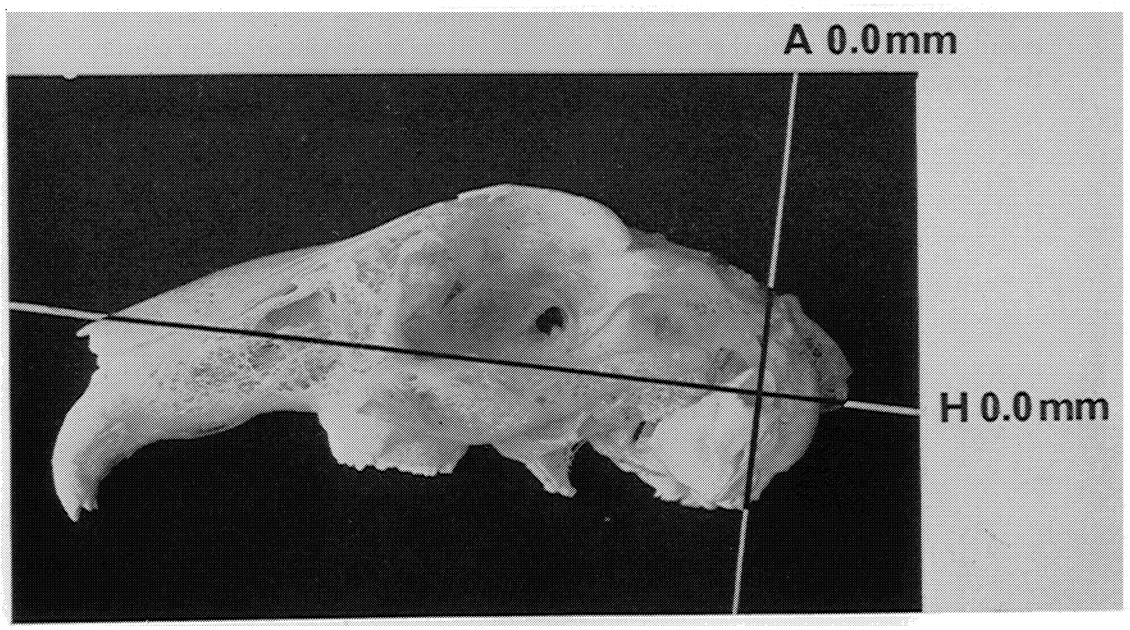

FIGURE 1. The lateral view of the rabbit's skull with marked horizontal (H 0.0 mm) and frontal (A 0.0 mm) Horsley-Clarke's basal planes.

the horizontal plane the needles were introduced 3 mm from the sagittal plane to avoid an interference of the horizontal tracks with those in the frontal planes. The main function of the tracks was to transfer stereotaxic planes intracranially and thus to facilitate later brain orientation.

The head of each subject was then perfused with 10% formalin via a carotid artery; the animal was decapitated and the brain was left *in situ* in