ANNUAL REPORT
OF
PROGRAM ACTIVITIES

NATIONAL EYE INSTITUTE

FISCAL YEAR 1971

U. S. DEPARTMENT OF HEALTH, EDUCATION, AND WELFARE
PUBLIC HEALTH SERVICE NATIONAL INSTITUTES OF HEALTH

ANNUAL REPORT

OF

PROGRAM ACTIVITIES

NATIONAL EYE INSTITUTE

Fiscal Year 1971

NATIONAL EYE INSTITUTE

ANNUAL REPORT

July 1, 1970 through June 30, 1971

TABLE OF CONTENTS

ANNUAL REPORT
NATIONAL EYE INSTITUTE
July 1, 1970 - June 31, 1971

STATEMENT OF THE INSTITUTE DIRECTOR
Carl Kupfer, M.D.

During the past year the National Eye Institute made significant progress toward becoming the fully functioning, independent organization envisioned by the Congress in its passage of the National Eye Institute Act.

Important staff positions were filled. Most notably, a new Associate Director for Extramural Research, Dr. George Brooks, was appointed to succeed Dr. Samuel S. Herman who retired from that position in March. Harold A. Kahn was appointed Chief of the newly created Office of Biometry and Epidemiology and Julian M. Morris was named Information Officer.

Our Advisory structure was strengthened with the creation of a Board of Scientific Counselors for the Intramural Program. Although there is at present no Clinical Director or Director for the Laboratory of Vision Research, the latter position will be filled early in FY 1972.

New guidelines and policies relating to the various research support mechanisms available to the Institute were drafted and put into effect which more closely defined these grants in terms of the research interests of the Institute.

The most pressing need of the Institute continues to be an adequate number of positions to provide backup and additional staff support to ongoing administrative and research activities and to initiate important functions for which there is presently no staff at all. Key areas requiring additional staff include extramural review and approval, program direction, biometry and epidemiology, program planning and review, and intramural research support.

ANNUAL REPORT
NATIONAL EYE INSTITUTE
July 1, 1970 - June 31, 1971

REPORT OF THE ACTING DIRECTOR OF INTRAMURAL RESEARCH

Carl Kupfer, M.D.

The Institute progressed toward its goal of creating a national resource for vision research with the reorganization and strengthening of the Intramural Program. The former Ophthalmology Branch was reconstituted as a Clinical Branch for patient studies and the Laboratory of Vision Research for experimental work. New equipment and renovated space for our Eye Clinic permitted the more efficient and extensive examination of patients and provided the setting for expanded clinical studies.

During the renovation of the new clinical area, temporary quarters necessitated a slowing-down of clinical research activities as noted by the number of out-patients seen from July 1, 1970 to June 30, 1971. This figure was 3241, of which 1348 were consults from other Institutes. However the in-patient clinical activity continued at a high level with 158 patients admitted for a total of 5904 in-patient hospital days.

Clinical studies during the year were primarily centered around investigations of the mode of action of medical therapy in glaucoma; retinal degenerations and dystrophies for which additional diagnostic studies are indicated; and inflammatory disease, with particular emphasis on Behcet's syndrome and sympathetic ophthalmia.

Basic research in the Laboratory of Vision Research included biochemistry of the retinal pigment rhodopsin, embryology of the lens and cornea, physiology of color vision, the coding of visual information from the eye to the brain, and the pharmacology of aqueous humor dynamics.

Serial No. NEI(I)-71 CB 006(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Design and Construction of Ophthalmic Instruments; Research in Methods of Evaluating Visual Processes.

Previous Serial Number: NEI(I)-70 O/OPS 006(c)
NDS(I)-56 O/OPS 300(c)

Principal Investigator: Ralph D. Gunkel, O.D.

Other Investigators: Peter Gouras, M.D.

Cooperating Units: None

Man Years:

Total:	0.8
Professional:	0.8
Other:	0.0

Project Description:

Objectives: Our objectives continue to be the promotion of the effectiveness of established procedures, techniques and instruments used in both clinical and laboratory work relating to the eye and its function, and wherever possible, to replace or supplement them with improvements.

The procedures involved consist mainly of subjective measurements for thresholds of visibility and color, recording of eye movements, electroretinography and photography of the eye, both external and internal.

Methods Employed: Collaboration with the Clinical and Research Associates is the primary mode of operation. Consultations and the pooling of ideas are encouraged.

Major Findings: Psychophysical tests were done on about 290 patients during the year, mostly for the purpose of establishing the state or stability of a tapeto-retinal degeneration or a toxic retinopathy.

A device for recording eye movements has been acquired and modified by the addition of an event marker on each channel for recording target position.

A new whole-field stimulator was constructed for use in electroretinography. It provides some new possibilities in the variation of brightness and color of background, with superimposed stimuli.

A simple portable adaptometer was designed for testing night vision in a mobile screening survey. Many small devices and modifications were made for the investigators in electrophysiology.

Significance to Biomedical Research and the Program of the Institute: Considerable stimulation and assistance has been provided for the new doctors arriving periodically, in addition to a measure of continuity in some of the on-going programs.

Proposed Course of Project: It is proposed that this project be continued in its present flexible form.

Honors and Awards: None

Publications:

Einaugler, R.B., Finkelstein, D., Gunkel, R.D., Brown, D.H., and Collins, E.: Retinitis pigmentosa and lymphocytic leukemia. Arch. Ophthal. (in press).

Serial No. NEI(I)-71 CB 030(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Studies of Parameters of Intraocular Pressure

Previous Serial Number: None

Principal Investigators: Carl Kupfer, M.D.
Douglas Gaasterland, M.D.

Other Investigators: Karyn Ross

Cooperating Units: None

Man Years:

Total:	2.9
Professional:	2.4
Other:	0.5

Project Description:

Objectives: The Glaucoma Laboratory is a new clinical research program established in the National Eye Institute during this year. The facilities which have been established include two complete laboratories wherein complete ocular examinations and evaluation of each of the seven important parameters of intraocular pressure can be carried out. The objectives are:

1. To measure the parameters of intraocular pressure in young normal volunteers.
2. To measure the parameters of intraocular pressure in patients with glaucoma.
3. To determine the effect of various pharmacologic agents on the parameters of intraocular pressure in the normal and glaucoma eye.

Methods Employed: The parameters evaluated include intraocular pressure, episcleral venous pressure, total facility of outflow, true facility, pseudofacility, aqueous humor flow and P_k of Goldmann. The present laboratory is the only one where such a complete study of aqueous humor dynamics is being conducted.

Major Findings: In young normal male subjects aqueous humor flow is nearly linearly related to intraocular pressure--as intraocular pressure increases flow decreases. The slope of the flow curve is the pseudofacility, a measure of vascular permeability. Acetazolamide causes a pressure-independent

decrease in the flow curve, with no alteration of pseudofacility. Topical l-epinephrine causes a pressure-dependent change of the flow curve, with reduction of pseudofacility and decrease of aqueous flow at intraocular pressure levels below 30 mmHg.

Significance to Biomedical Research and the Program of the Institute: A comparative evaluation of the patterns of alteration of flow curves induced by medications is expected to help understanding of the mechanisms of action of the medications. This is particularly significant for medications which may cause changes in ultrafiltration versus changes in secretion of aqueous humor. Younger patients, with a more intact vascular endothelium and more vigorous metabolic processes, may be more susceptible to the influence of medications which alter secretion, whereas older patients might be expected to be more affected by medications which alter ultrafiltration. Such findings will bear on the treatment of glaucoma.

Proposed Course of Project: These studies will be extended to patients with glaucoma.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 CB 031(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 20, 1971

Project Title: Lymphocyte Transformation in Sympathetic Ophthalmia

Previous Serial Number: None

Principal Investigator: Vernon G. Wong, M.D.

Other Investigators: Richard Anderson, B.S.
Paul O'Brien, Ph.D.

Cooperating Units: None

Man Years:

Total:	0.5
Professional:	0.25
Other:	0.0

Project Description:

Objectives: To determine the presence of sensitized peripheral lymphocytes to ocular tissues in sympathetic ophthalmia.

Methods Employed: Peripheral lymphocytes from eight patients with sympathetic ophthalmia diseases were studied for blastogenic transformation when incubated in the presence of ocular and non-ocular homologous tissue antigens.

Major Findings: Seven of eight patients with active sympathetic ophthalmia demonstrated lymphoblastic transformation in the presence of homologous human uveal-retinal extract. The mean blastogenic enhancement showed a significant ($p<0.05$) ten-fold increase (55.5) when compared to its own control incubated in the presence of skeletal muscle antigens (5.6). In the control group of patients without sympathetic ophthalmia, there was no significant increase in lymphocyte transformation.

Significance to Biomedical Research and the Program of the Institute: The following points can be derived from the above studies: 1) Patients with sympathetic ophthalmia contained sensitized lymphocytes to some component(s) of uveal-retinal tissues. 2) The sensitivity appears to be organ-specific since control cultures containing homologous skeletal muscle antigens

failed to evoke any significant degree of response. 3) This lymphocytic response indicates that sympathetic ophthalmia may represent a form of autosensitization of the delayed hypersensitivity type.

Proposed Course of Project: This project is being continued.

Honors and Awards: None

Publications:

Wong, V.G., Anderson, R., and O'Brien, P.: Sympathetic ophthalmia and lymphocyte transformation. Amer. J. Ophthal. (in press)

Serial No. NEI(I)-71 CB 023(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June, 1971

Project Title: Methotrexate Therapy of Selected Patients with Uveitis

Previous Serial Number: NEI(I)-70 0/0C 023(c)
NDS(I)-63 0/0C 1022(c)

Principal Investigator: Vernon G. Wong, M.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.3
Professional:	0.3
Other:	0.0

Project Description:

This project is being terminated this year, but may be continued in the future.

Serial No. NEI(I)-71 CB 024(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Cystinosis

Previous Serial Number: NEI(I)-70 0/0C 024(c)
NDS(I)-68 0/0C 1540(c)

Principal Investigator: Vernon G. Wong, M.D.

Other Investigators: Joseph Schulman, M.D.
J. Edwin Seegmiller, M.D.

Cooperating Units: Laboratory of Human Genetics, NIAMD

Man Years:

Total:	0.3
Professional:	0.25
Other:	0.05

Project Description:

Objectives: To develop a single, specific, and rapid method for the biochemical diagnosis of cystinosis based on analysis of a biopsy of conjunctiva.

Methods Employed: A small piece of bulbar conjunctiva is excised and is fixed immediately in absolute ethanol. Cystine is extracted in 0.5 ml 0.1N HCI and the amino acid is assayed by column chromatography.

Major Findings: In cystinotic patients the cystine contents were never less than 5 and usually greater than 30 mμ moles per 1/2 cystine per milligram of wet weight of tissue. Each specimen from 13 cystinotic patients (congenital and benign cystinosis) including those stored in ethanol for up to 2 years had an easily detectable quantity of cystine. The cystine contents of biopsies stored in ethanol from proved heterzygotes or normals (15 patients) were always less than 0.4 mμ/moles per milligram of wet weight.

Similar results were obtained in 4 samples (2 normals and 2 homozgotes) of conjunctiva which were analyzed fresh without processing in ethanol.

Significance to Biomedical Research and the Program of the Institute: The significance lies in the fact that by this method we have been able to distinguish cystinotic patients clearly from non-cystinotic ones in all

cases tested. Conjunctival biopsy has provided the physician a simple and a far less traumatic means of providing a specific diagnosis. Furthermore, tissue samples stored in absolute alcohol proved to be stable for many months at room temperature without fear of losing their cystine content.

Proposed Course of Project: The project is being continued.

Honors and Awards: None

Publications:

Wong, V.G., Schulman, J., and Seegmiller, J.E.: Conjunctival biopsy for the biochemical diagnosis of cystinosis. Amer. J. Ophthal. 70: 278-281.

Serial No. NEI(I)-71 CB 025(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Experimental Endogenous Immune Uveitis

Previous Serial Number: NEI(I)-70 0/0C 025(c)

Principal Investigator: Vernon G. Wong, M.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.4
Professional:	0.4
Other:	0.0

Project Description:

This project is being terminated this year, but may be continued in the future.

Serial No. NEI(I)-71 CB 032(c)
1. Clinical Branch
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Lattice Corneal Dystrophy

Previous Serial Number: None

Principal Investigator: Vernon G. Wong, M.D.

Other Investigators: R. A. Delellis, M.D.
G. G. Glenner, M.D.

Cooperating Units: NIAMD

Man Years:

Total:	0.3
Professional:	0.15
Other:	0.0

Project Description:

Objectives: Lattice dystrophy of the cornea is manifested by progressive opacities and transmitted as an autosomal dominant trait. The etiology of this disorder is unknown. The purpose of this study is to determine the nature of the corneal pathology.

Methods Employed: Corneal buttons from two related adults with corneal lattice dystrophy were removed for full thickness transplantation. The specimens were divided into three portions each for electron microscopic, histochemical and immunohistochemical studies. The latter method required the use of fluorescein labelled antisera to human amyloidotic spleen and liver tissues.

Major Findings: 1. Light microscopy examination: In sections stained with hematoxylin and eosin, deposits of densely eosinophilic material were noted throughout the corneal stroma. In polarized light, these areas exhibited an intense green birefrigence. In adjacent tissue sections, the deposits stained positively with PAS and were metachromatic with crystal violet. Congo red stained intensely along with positive staining of bright yellow fluorescence after thioflavin-T. 2. Electron Microscopy: Between bundles of collagenous fibers there were many large and small cords and plaques of moderately electron-dense reticular and finely fibrillar protein material. These electron-dense fibers measure 80-100Å in width and tend to be randomly arranged. 3. Immunohistochemical Studies: Amyloid deposits in unstained frozen sections exhibited

an intense blue autofluorescence. When reacted with antisera to human amyloid deposits, specific apple-green fluorescence was limited to the amyloid deposits. Tissues reacted with fluorescein-labeled normal rabbit serum remained unstained.

Significance to Biomedical Research and the Program of the Institute: 1. By using specific antisera, we have demonstrated for the first time that lattice dystrophy of the cornea is a form of amyloidosis. 2. It has been shown that the antigenic component of undenatured human amyloid deposits is a protein and this component has not yet been detected in normal human tissue. 3. It appears that this ocular dystrophy represents an isolated familial form of amyloidosis.

Proposed Course of Project: This project is to be continued.

Honors and Awards: None

Publications:

Bowen, R.A., Hassard, D.T.R., Wong, V.G., DeLellis, R.A., and Glenner, G.G.: Lattice Dystrophy of the cornea as a variety of amyloidosis. Amer. J. Ophthal. 70: 822-825, 1970.

Serial No. NEI(I)-71 LVR 016(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Protein Synthesis in the Retina

Previous Serial Number: NEI(I)-70 0/0M 016(c)
NDS(I)-68 0/0M 1533(c)

Principal Investigators: Jacqueline J. Bungenberg de Jong, Ph.D.
Paul J. O'Brien, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	1.0
Professional:	0.7
Other:	0.3

Project Description:

Objectives: An understanding of developmental impairment and progressive degeneration of the retina requires a thorough knowledge of the details of the normal processes which regulate photoreceptor cell renewal. This project was designed to provide a convenient system in which to study some of these processes, in particular the biosynthesis of rhodopsin.

Methods Employed: Standard biochemical methods were employed including incubation of retinas, cell fractionation, detergent extraction of membrane proteins and purification by column chromatography.

Major Findings: Radioactive amino acid was found to be incorporated into many subcellular fractions of the bovine retina. A major radioactive component of the photoreceptor outer segments was shown to be rhodopsin after extensive purification and demonstration of its sensitivity to light. The time course of rhodopsin labeling parallels the results observed *in vivo* where rhodopsin is formed in the inner segment and migrates to the outer segment.

Significance to Biomedical Research and the Program of the Institute: These results indicate that a reliable *in vitro* system is now available for

study of the photoreceptor renewal process in greater detail than would be possible in vivo since the environmental conditions are subject to laboratory control and manipulation.

Proposed Course of Project: Future studies will be concerned with the subcellular sites at which various components of the rhodopsin molecule are added, the metabolic controls which regulate these processes and the mechanism by which the rhodopsin molecule migrates to the outer segment and is incorporated into new disc membranes.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 001
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH

Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Ocular Morphogenesis

Previous Serial Number: NEI(I)-70 0/EE 01(c)
NDS(I)-69 0/EE 1661(c)

Principal Investigator: A. J. Coulombre, Ph.D.

Other Investigators: Jane L. Coulombre, B.S.
S. Goldberg, M.D. (guest worker)
D. Newsome, M.D.

Cooperating Units: Departments of Pathology and Ophthalmology, The Johns Hopkins University, Baltimore and Saint Christopher's Hospital for Children, Philadelphia.

Man Years:

Total:	4.20
Professional:	3.70
Other:	0.50

Project Description:

Objectives: We seek to identify and to characterize the tissue interactions which control the orderly growth of the vertebrate eye. Most congenital defects are attributable to interference with these interactions.

Methods Employed: Routine chemical, histochemical, experimental-embryological, microscopic, electron-microscopic, pharmacologic, tissue culture and autoradiographic techniques are used to analyze the development of the visual system of the domestic fowl. This year we developed a new technique for impregnating the nerve fibers of the retino-tectal projection with silver in whole mounts. This procedure makes possible a new attack on the mechanisms governing the establishment of an orderly retino-tectal projection.

Major Findings: During FY71 new information was obtained or published on:

I. Cornea: 1. The collagenous architecture of the corneal stroma is represented in the embryonic cell-free primary stroma which is secreted by the anterior epithelium of the cornea beginning at three days of incubation.

Each lamella of the primary stroma is deposited beneath the epithelium in its turn until the last of approximately fifty such layers appears at ten days of incubation. The geometry of the collagen deposited by the fibrocytes to form the secondary stroma is determined by the structure of the primary stroma. This may explain why the scar tissue which forms when the stromal architecture is disrupted by wounds is disordered and opaque. 2. The basal cells of the anterior corneal epithelium produce the tropocollagen which is deposited in the primary stroma. We have identified elongated vesicles in the basal cytoplasm of these cells which appear to be the final depots from which the collagen is secreted into the primary stroma. 3. Even after it begins to differentiate in this way the corneal anterior epithelium can be modulated by challange with appropriately inductive mesenchymes to form feathers or scales. This metaplastic transformation is not species specific since it can be elicited by mouse embryonic dermis. 4. L-azetidine-2-carboxylic acid produces lesions in the primary stroma which sink deeper with developmental time, confirming that the lamellae of the primary stroma are produced seriatim beneath the anterior epithelium of the cornea. 5. A human case of incomplete cleavage of the chamber angle was shown to involve displacement of corneal stromal tissue to the anterior face of the iris.

II. Lens: 1. Complete lenses are reconstituted in the lentectomized eye of the chick embryo when a piece of lens epithelium is introduced into the vacated orifice of the eye cup. 2. Lens epithelium retains its ability to form a closed vesicle long after the stage at which the lens vesicle normally forms. 3. Reconstituted lens vesicles grow to fill the orifice of the eye cup but do not overgrow this space. 4. Reconstituted lens vesicles regenerate a lens capsule. 5. The lenses which develop from reconstituted lens vesicles are always properly oriented with respect to the optic axis. Three factors were identified which actively control this orientation: contact of the lens with the margin of the eye cup; a mesenchymal influence which promotes lens fiber elongation; and a neural retinal factor which promotes an increase in lens cell volume. 6. The ability of the lens epithelium to respond to these influences decreases slowly with embryonic age.

III. Retina: 1. The induction by neural retina of neural retinal regeneration from the chick embryonic pigmented epithelium has a low species specificity since mouse embryonic neural retina also elicits neural retinal regeneration in the chick embryo. 2. The chick neural retina gradually loses its retinal inductive potency as a function of embryonic age. 3. The axons of the ganglion cells of the neural retina are oriented toward the choroid fissure as soon as they appear. They begin to fasciculate before they reach the fissure, forming two groups of bundles which enter the fissure at different angles. Nasally these bundles diverge to form an arcuate pattern in which axons are sparsely rather than densely aggregated, as has been believed hitherto. These findings limit the types of hypotheses that can be entertained concerning axonal guidance during embryogenesis.

IV. Sclera: 1. The cartilagenous sclera of the chick embryo is induced by the pigmented epithelium. 2. The inductive potency of the pigmented epithelium is restricted to a brief period early in embryonic development.

3. Each of the scleral ossicles is induced by a conjunctival papilla beneath which embryonic collagen is deposited prior to bone formation. When secretion of this collagen is aborted with l-azetidine-2-carboxylic acid the ossicles fail to develop. This strengthens the presumption that embryonic collagen produced beneath the papillae during brief periods early in embryonic development may induce the differentiation of osteocytes. 4. The collagen between the ossicular beds is not osteogenic and plays a role in determining the direction of overlap of the scleral bones.

Significance to Biomedical Research and the Program of the Institute: Identification and characterization of the tissue interactions which control ocular development is essential to an understanding of the etiology of most congenital eye defects. Particular attention is currently focused on the role of embryonic collagen in a number of inductive tissue interactions which are responsible for the differentiation of most, of not all, of the cell types in the eye. This class of epithelially produced collagens may be importantly involved in the development of most of the organs of the body.

Honors and Awards: None

Publications:

Coulombre, A.J.: Development of the vertebrate motor system. In Schmitt, F., Quarton, G., Melnechuk, T., and Adelman, G., (Eds.): The Neurosciences, Second Study Program. New York, N.Y., The Rockefeller University Press, 1970, pp. 108-116.

Coulombre, J. L., and Coulombre, A.J.: Influence of the mouse neural retina on regeneration of chick neural retina from chick embryonic pigmented epithelium. Nature 228: 559-560, 1970.

Coulombre, J. L., and Coulombre, A.J.: Lens development. V. Histological analysis of the mechanism of lens reconstitution from implants of lens epithelium. J. Exp. Zool. 176: 15-24, 1971.

Coulombre, J. L. and Coulombre, A.J.: Metaplastic induction of scales and feathers in the corneal anterior epithelium of the chick embryo. Develop. Biol. (in press)

Karlsberg, R., Jared, M., Emery, M., Green, W., Baldes-Depena, M., and Coulombre, A.J.: Iris and chamber angle anomalies in a child with multiple congenital anomalies. Arch. Ophthal. (in press)

Trelstad, R.: Vacuoles containing collagen in the embryonic chick corneal epithelium: an epithelium which produces collagen. J. Cell Biol. 48: 689-694, 1971.

Trelstad, R., and Coulombre, A.J.: Morphogenesis of the collagenous stroma in the chick cornea. J. Cell Biol. (in press)

Serial No. NEI(I)-71 LVR 026
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Physiology of the Primate Visual System

Previous Serial Number: None

Principal Investigators: Bruce Dow, M.D.
Peter Gouras, M.D.

Other Investigator: Pieter Padmos, Ph.D. (Guest Worker)
Eleanor Collins

Cooperating Units: Institute of Perception, Soesterberg, The Netherlands (for defraying costs of Dr. Padmos's stay in our laboratory).

Man Years:

Total:	2.8
Professional:	2.8
Other:	0.0

Project Description:

Objectives: The broad aim of this project is to understand the cellular organization underlying visual perception. We have concentrated on the visual system of the Rhesus monkey because its similarity to the human visual system allows for more direct comparisons with human perception about which there is a large volume of information.

The recent work has concentrated on the area of striate cortex subserving foveal vision since this is undoubtedly one of the most important parts of the primate visual system and this is a logical extension of our previous work on foveal retina. The specific objective of the current work is to understand how the perception of color and shape interact in the different layers and cells of foveal striate cortex.

Methods Employed: Our major tool is the ultrafine glass micro-pipette electrode which enables one to record electrically from outside as well as inside single neurons without significantly disturbing their function. The electrodes can be filled with conducting dye solutions (such as Procion yellow) which can be electrophoresed into a specific cell layer or in some

cases inside a specific cell from which responses have been recorded. We believe this to be a powerful tool for linking structure with function in complex neural centers such as visual cortex.

Major Findings: There are two general classes of cortical cells, one involved in color, the other not. The former are less common than the latter. The ones not involved in color processing appear to be involved primarily in the perception of shape. Of the color cells some are involved only in color, per se; others seem to take part in shape detection as well. In one region of visual cortex subserving a specific area of visual space color cells tend to be clustered together within a particular part of this region. Therefore there is some segregation for cells which process color from those which do not.

One of our most interesting finding indicates that there is some hierarchichal system of color processing within striate cortex which includes both broad and narrow band color sensitive cells as well as dichromatic and trichromatic ones.

We have been able to record from within single cells in striate cortex enabling us to sample dendritic activity which is undetectable by the more conventional extracellular recording techniques. This is an important parameter for distinguishing different cell types within the cortex and in addition allows us to inject and subsequently locate dyes within such single cell, which we have now succeeded in doing. This appears to be the first time that any cell has been recorded from and subsequently stained and identified in cerebral cortex.

Significance to Biomedical Research and the Program of the Institute: Such studies of retinal function at the cellular level should prove valuable for understanding vision and pathophysiology of retinal disease.

Proposed Course of Project: To continue linking function with specific cell in foveal striate cortex.

Honors and Awards:

Invited Lecture entitled: "Color coding in Striate Cortex" to be delivered before the 25th International Congress of Physiological Sciences in Munich, July 28, 1971.

Publications:

Gouras, P.: The function of the midget cell system in primate color vision in Symposium on Visual Processes in Vertebrates, Vision Res. (in press)

Serial No. NEI(I)-71 LVR 020(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Hereditary Blindness Among the Pingelapese People of the Eastern Caroline Islands

Previous Serial Number: NEI(I)-70 0/0C 020(c)

Principal Investigator: Richard Einaugler, M.D.

Other Investigators: Peter Gouras, M.D.
Daniel Finkelstein, M.D.
Ralph D. Gunkel, O.D.

Cooperating Units: Epidemiology Branch, C&FR, NINDS

Man Years:

Total:	0.6
Professional:	0.6
Other:	0.0

Project Description:

This project is being terminated.

Serial No. NEI(I)-71 LVR 004(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Electrophysiologic and Psychophysical Studies of Retinal Degenerations

Previous Serial Number: NEI(I)-70 0/OPS 004(c)
NDS(I)-63 0/OPS 1012(c)

Principal Investigators: Peter Gouras, M.D.
Daniel Finkelstein, M.D.
Richard Einaugler, M.D.

Other Investigator: Ralph Gunkel, O.D.

Cooperating Units: None

Man Years:

Total:	0.6
Professional:	0.6
Other:	0.0

Project Description:

This project is being terminated.

Serial No. NEI(I)-71 LVR 005
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Electrophysiological Studies of Mammalian Retina

Previous Serial Number: NEI(I)-70 0/OPS 005(c)
NDS(I)-63 0/OPS 1016(c)

Principal Investigators: Peter Gouras, M.D.
Mary Hoff, A.B.
Gunter Niemeyer, M.D., (Guest Worker)

Other Investigators: None

Cooperating Units: University of Zurich, Switzerland, for defraying costs of Dr. Niemeyer's visit to our laboratory.

Man Years:

Total:	2.1
Professional:	2.1
Other:	0.0

Objectives: The broad objective is to understand the mechanisms of visual processing in the retina. The current objective is to develop a system whereby it would be possible to record from within each of the different cell types in a mammalian retina.

Methods Employed: The principal technique involves the use of the isolated, perfused cat eye as a prototype mammalian system, a technique which has been originally developed in our laboratory. It has the great advantage of eliminating both vascular and respiratory pulsations which are a major handicap when using ultrafine pipettes to record from extremely small retinal cells.

Major Findings: Dr. Niemeyer has now succeeded in recording from single bipolar and horizontal cells in the perfused cat eye. His major success is in demonstrating that in the predominantly rod retina of the cat, bipolar cells can be detected which receive signals only from cones. This has important implications for understanding the specific connectivity between cells in the external plexiform layers. The presence of cone bipolar cell indicates that there must be some autonomy to the rod and cone signals processed by horizontal cells which intervene between receptor and bipolar cells in the external plexiform layer. It is the first bipolar cell that has ever been recorded from in mammalian retina.

Miss Hoff has demonstrated that neural transmission from receptors to bipolars to ganglion cells can be restablished after total interruption of the circulation to the perfused mammalian eye for as long as two hours. This indicates that the mammalian central nervous system is much more tolerant of ischemia than is generally believed. Knowledge of this has given us considerable more time to set up the perfused eye preparation during the cannulation procedure. It also has some interesting implications for the acute treatment of human retinal vascular occlusions.

Significance to Biomedical Research and the Program of the Institute: Understanding the cellular physiology of the mammalian retina can only lead to a better understanding of abnormal states observed clinically.

Proposed Course of Project: Attempts will be made to classify horizontal and bipolar cells in cat retina by intracellular recording techniques.

Honors and Awards: None

Publications:

Gouras, P. and Hoff, M.: Retinal function in an isolated, perfused mammalian eye. Invest. Ophthal. 9: 388-399, 1970

Gouras, P.: Light and dark-adaptation, In Fuortes, M.G.F. (Ed.): Handbook of Sensory Physiology. New York, N.Y., Springer-Verlag, Vol. VII (2). (in press).

Gouras, P.: S-potentials. In Fuortes, M.G.F. (Ed.): Handbook of Sensory Physiology. New York, N.Y., Springer-Verlag, Vol. VII (2). (in press)

Gouras, P.: Electroretinography: Some basic principles. Invest. Ophthal. 9: 557-569, 1970.

Finkelstein, D. and Gouras, P.: Visual electrophysiology, An introduction to the ERG, EOG, ERP and VER. Int. Ophthal. Clin. 9: 857-881, 1969.

Finkelstein, D. and Gouras, P.: Human electroretinogram near the absolute threshold of vision. Int. Ophthal. Clin. 9: 1073-1081, 1969.

Serial No. NEI(I)-71 LVR 010(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Chemistry of the Vitreous Body

Previous Serial Number: NEI(I)-70 0/0CH 010(c)
NDS(I)-67 0/0CH 1433(c)

Principal Investigator: Ralph J. Helmsen, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.4
Professional:	0.3
Other:	0.1

Project Description:

Objectives: To isolate tissue-specific sialoproteins and/or sialoglycans from vitreous and to characterize these macromolecules by physical and immunological techniques.

Methods Employed: Distinct proteins were isolated and fractionated from vitreous of cattle by use of glass bead column chromatography and/or salt precipitation plus membrane ultrafiltration. Colorimetry was employed to determine sialic acids present in the tissue.

Major Findings: A rapid separation of hyaluronic acid from the soluble proteins and nucleotides of undialyzed bovine vitreous was achieved by a new technique which employs pressure chromatography on columns of glass beads with pore diameters of 370A°. Samples up to 5 ml were successfully processed by this procedure and the method is suitable for work with single eye preparations.

A vitreous protein with a molecular weight between 50,000 and 100,000 was isolated by use of perchloric acid precipitation and membrane ultrafiltration. This substance gave a single band when stained after polyacrylamide gel electrophoresis.

Significance to Biomedical Research and the Program of the Institute: Isolation of a tissue-specific sialoprotein or sialoglycan from vitreous in large amounts would provide source material for immunological studies

on the production of uveitis in experimental animals. If such studies were successful, further investigations with the macromolecule modified chemically might suggest an approach for control of the clinical condition in humans.

Proposed Course of Project: New glass bead materials will be examined for efficacy in large scale isolation of vitreous sialoproteins. Ganglioside contamination of the various connective tissue fractions will be assessed.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 027(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Induction of Buphthalmos in Chicks by Feeding a High Level of Glycine

Previous Serial Number: None

Principal Investigator: Ralph J. Helmsen, Ph.D.

Other Investigators: Max Rubin, Ph.D. (University of Maryland)
Douglas Gaasterland, M.D.

Cooperating Units: Department of Poultry Science, University of Maryland

Man Years:

Total:	0.98
Professional:	0.7
Other:	0.28

Project Description:

Objective: To study the physical and chemical factors which control the size and shape of vitreous during development of the eye as well as maturity.

Methods Employed: Weight determinations were made on the total eye and various ocular tissues. Colorimetry was employed to measure the quantity of each of the major macromolecules in dialyzed vitreous.

Major Findings: The feeding of 8% glycine to newly hatched chicks raised for a period of 7 weeks on a basal diet supplemented with 5% gelatin and 10mg % nicotinic acid produces a depression in body growth and an enlargment of the eyeball (buphthalmos). Elevation of the level of gelatin from 5 to 8% and reduction of glycine from 8 to 6% in the diet results in elimination of the growth depression with retention of the eye effect. No change in intraocular pressure or facility of aqueous humor outflow was observed in those birds fed glycine when compared to controls which did not receive the amino acid.

Significance to Biomedical Research and the Program of the Institute: Chicks grown on a high-glycine diet represent a new model for the study of buphthalmos in experimental animals. This state appears to differ from the photo-induced eye enlargement in chickens and the recessive buphthalmia

observed in rabbits in that no evidence of glaucoma was detected. These animals in addition provide a useful experimental tool for the study of the biochemical changes which take place in developing vitreous.

Proposed Course of Project: Glycine-rich peptides and various proteins will be assessed as to their ability to substitute for gelatin in enhancing the effect of glycine in the induction of buphthalmos. An attempt will also be made to extend the time for development of the eye enlargement in the growing chick by injection of high glycine solutions into fertilized eggs before hatching.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 028(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Antibody in Tears Following Intranasal Instillation of Inactivated Virus: II Use of poly I:C to Stimulate Higher Titers of Anti-vaccinia Antibody in Rabbit Tears

Previous Serial Number: None

Prinicpal Investigator: Harry L. S. Knopf, M.D.

Other Investigators: Morris Glassman, M.D.
Neil Blacklow, M.D.
Walter L. Cline
Vernon G. Wong, M.D.

Cooperating Units: Laboratory of Infectious Diseases/NIAID

Man Years:

Total:	1.5
Professional:	1.25
Other:	0.25

Project Description:

Objectives: There were three major objectives:
I. To determine if the rabbit was a suitable experimental model for the investigation of antibody function in tears.
II. To determine if an inactivated virus (vaccinia) vaccine could stimulate antibody in tears when administered by various routes.
III. To determine if an adjuvant poly I:C- would enhance the production of tear antibody if administered with the vaccine.

Methods Employed: Formalin-inactivated or live vaccinia virus was administered intranasally or intradermally to 15 animals. Complement fixation (CF), hemagglutination inhibition (HI), neutralization (NT), sucrose gradient ultracentrifugation and indirect fluorescence were used to assay anti-viral activity.

Immunoglobulin concentration in tears was determined by a modification of antibody-in-agar technique (single, radial diffusion). The method was adapted for use with a commercial antiserum to rabbit 7S globulin. It allowed simultaneous measurement of IgA and IgG concentrations in secretions.

Major Findings: I. Rabbits responded to intranasal vaccination in much the same way as humans; they developed increased titers of specific Ab in nasal secretions and serum, with a less marked response in tears.
II. Poly I:C increased antibody titers in both serum and tears:

A. The geometric mean NT titers in serum were as follow:

Intradermal with poly I:C = 1/830
Intranasal with poly I:C = 1/400
Interdermal-live = 1/370
Inranasal inactivated alone = 1/40

B. Tear antibody was just above baseline for the intranasal vaccine group (without adjuvant). The other groups had nearly equivalent levels of tear antibody activity; but the intranasal -poly I:C animals were usually slightly higher than other groups.

III. The peak titers, which were noted after the fourth dose of vaccine, began to decrease within one week. This indicated the adjuvant was short-lived in its effect.

IV. No toxicity was noted in animals receiving poly I:C.

Significance to Biomedical Research and the Program of the Institute: There are four basic conclusions which can be derived from the experiments:
I. The rabbit appers to be a good experimental animal for the study of secretory antibody and external infections of the eye.
II. Antibody titers at mucosal surfaces can be specifically and significantly increased by the appropriate application of antigen. It remains to be shown, however, that the antibody in tears has a significant functional role in ocular infections.
III. Use of adjuvants such as poly I:C may provide a means by which vaccination can produce higher titers of protective antibody with fewer doses of vaccine.
IV. Inactivated vaccinia may find use as a pre-treatment, to be administered prior to live vaccine in order to decrease the complications of the latter.

Proposed Course of Project: To be continued.

Honors and Awards: None

Publications:

Knopf, J.L., Bertran, D.M. and Dapikian, A.Z.: Demonstration and characterization of antibody in tears following intranasal vaccination with inactivated type 13 Rhinovirus: A preliminary report. Invest. Ophthal. 9: 727-734, 1970.

Serial No. NEI(I)-71 LVR 029(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Antibody in Tears Following Intranasal Instillation of Inactivated Virus: III Examination of the Role of Tear Antibody in Experimental Vaccinia Conjunctivitis

Previous Serial Number: None

Principal Investigators: Harry L.S. Knopf, M.D.

Other Investigators: Morris Glassman, M.D.
Neil Blacklow, M.D.
Walter L. Cline
Vernon G. Wong, M.D.

Cooperating Units: Laboratory of Infectious Diseases, NIAID

Man Years:

Total:	1.0
Professional:	0.75
Other:	0.25

Project Description:

Objectives: As a continuation of the previous study, this project had a single major objective: to determine the relative role of secretory antibody in experimental conjunctivitis.

Methods Employed: The animals from the previous study were challenged with live homologous vaccinia virus by dropping it on the unscarified eye. Clinical observations were made and cultures were taken for 2-3 weeks after infection. Evidence for a modifying affect by serum and/or tear antibody was sougnt in the analysis and correlation of (a) days of clinical illness (b) quantity and length of virus shedding and (c) serologic changes in tears and sera.

Major Findings: Preliminary results are: 1) Virus shedding in the vaccinated animals appeared to be decreased in all vaccinees as compared to controls. However, this change was independent of the route of administration of vaccine. 2) The degree of virus shedding and clinical illness did not appear to correlate well with tear antibody, and was more dependent on serum levels. 3) One vaccinated animal developed iritis during the challenge study, but all others developed only mild conjunctivitis.

Significance to Biomedical Research and the Program of the Institute: The results of the experiment are preliminary, but they suggest several conclusions: 1) In experimental vaccinia conjunctivitis, it does not appear that tear antibody plays a significant role in prevention of illness. However, it may have some other modifying influence which is not yet apparent. 2) Iritis occurred in one animal, and may represent a hypersensitivity reaction. 3) Humoral antibody did not seem to be the sole factor in disease modification. This suggests that delayed hypersensitivity may play a role and should be investigated. 4) The rises in tear antibody titer after infection or vaccination may represent a method for diagnosing conjunctivitis of unknown or suspected viral etiologies.

Proposed Course of Project: To be continued.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 008(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Chemistry of Rhodopsin

Previous Serial Number: NEI(I)-70 O/OCH 008(c)
NDS(I)-65 O/OCH 1210(c)

Principal Investigator: Marc S. Lewis, Ph.D.

Other Investigators: Dr. Melvin Gottlieb, Laboratory of Physical Biology, NIAMD

Cooperating Units: None

Man Years:

Total:	0.8
Professional:	0.8
Other:	0.0

Project Description:

Objectives: To study the structural and functional aspects of the bovine rhodopsin molecule.

Methods Employed: Particles were prepared by sonically disrupting bovine rod outer segments which had been purified by gradient centrifugation. Velocity and equilibrium studies were performed on these particles in the analytical ultracentrifuge. Lysolecithin, a phospholipid which may be considered to be a representative model of the phospholipids which are a part of the visual pigments, was also studied using equilibrium techniques in the analytical ultracentrifuge.

Major Findings: The need for a high-intensity, monochromatic light source operating at a wavelength greater than 600 nm. for the ultracentrifugal studies on rhodopsin led to the development of a method of using a He-Ne laser for this purpose. The laser has proved to be extremely satisfactory for this purpose, giving improved optical performance of the ultracentrifuge without causing any apparent bleaching of rhodopsin.

The molecular weight and partial specific volume of the sonicated particles were calculated by using the molecular dimensions obtained from electron microscopy with data from the analytical ultracentrifuge. The molecular weight was found to be 1.1 million and the partial specific volume was found

to be 0.883. This partial specific volume indicates a relatively high lipid content for these particles, which is consistent with their membrane origin. No effect due to bleaching was found for either the molecular weight or the partial specific volume. Sedimentation velocity studies showed that there was no significant change in either shape or volume of the particles as a result of bleaching. These results are consistent with the observation, based on circular dichroism studies, that there is no apparent loss of helical content in the particles as a result of bleaching.

Sedimentation equilibrium studies were performed on solutions of synthetic 1,3 palmitoyl lysolecithin in order to study the thermodynamics of the self-association of this phospholipid. Micellar size ranged from 170 to 190 monomer units; the change in standard free energy of association was calculated to be in the range of 5.3 to 5.9 kcal/mole of lysolecithin monomer, with the exact values depending upon the electrolyte present in the solvent system. Lysolecithin has been demonstrated to be effective for the solubilization and extraction of rhodopsin from bovine rod outer segments.

Significance to Biomedical Research and the Program of the Institute: In addition to its usefulness for ultracentrifugal studies on rhodopsin, the laser light source represents a significant advance for ultracentrifugal analysis generally, giving increased precision because of the greater optical resolution obtainable.

The absence of any effect of bleaching on the size or shape of the sonicated rod outer segment particles is particularly significant because it demonstrates that the changes in size and shape alleged to occur with bleaching of detergent solubilized rhodopsin do not appear to have this effect on the behavior of ensembles of rhodopsin with other molecules in the rod outer segment membrane. This is not intended to imply that there are no conformational changes as a result of bleaching, but rather that the changes which do take place are of a more subtle nature than would be implied by studies on solubilized rhodopsin, and must take into account the interaction of the rhodopsin with the other proteins and the lipids of the rod outer segment membrane.

The studies on lysolecithin have several important implications. They suggest that the change in free energy of association of phospholipids with the hydrophobic region of rhodopsin will be at least as great as the change of free energy of self-association of lysolecithin. They indicate that the change in free energy occuring when lysolecithin disrupts the outer segment membrane and solubilizes rhodopsin is greater than the free energy change for the formation of the lysolecithin micelle. Lysolecithin appears to have great potential as a physiological solubilizing agent for rhodopsin as well as serving as a model compound for studying the association of phospholipids with rhodopsin.

Proposed Course of Project: Further studies along the same lines as those described above are planned with particular emphasis on studies on the association of lysolecithin with rhodopsin.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 009(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Physical Chemistry of Model Gel Systems

Previous Serial Number: NEI(I)-70 0/0CH 009(c)
NDS(I)-63 0/0CH 1217 (c)

Principal Investigator: Marc S. Lewis, Ph.D.

Other Investigators: Richard Shrager, Physical Sciences Laboratory, DCRT

Cooperating Units: None

Man Years:

Total:	0.2
Professional:	0.2
Other:	0.0

Project Description:

Objectives: To study the physical and chemical parameters of model systems which are pertinent for transparency and opacity of gel systems. In particular, the objectives are to isolate and to characterize the soluble proteins of lenses of various species at various ages under both normal and pathological conditions and to relate the properties of the various proteins to the structure and function of the lens.

Methods Employed: Because of the evidence that some of the lens proteins appear to behave as either reversible or irreversible associating systems, current work in this area has been devoted to improving methods of studying systems of this type. In particular, emphasis has been given to the development of a powerful and sophisticated method of computer analysis which would be suitable for the analysis of ultracentrifugal, light-scattering, and osmotic pressure data from such systems.

Major Findings: Programs have been developed which have been quite successful in the analysis of synthetic data and also in the analysis of ultracentrifugal data for carbonmonoxy hemoglobin (furnished by Dr. Arlan Gottlieb, Department of Medicine, University of Pennsylvania). These programs utilize the Modelaide system, permitting the investigator to interact with the computer during the analysis, and greatly facilitating obtaining meaningful results, since the data and the analysis are presented graphically on a

cathode ray tube display system. It was found that the carbonmonoxy hemoglobin system could be best represented as a monomer-dimer-tetramer-octamer system. Since, at normal concentrations, hemoglobin is in the tetrameric form, the dimer and monomer represent dissociation products, and the octamer may be thought of as a dimer of the normal tetrameric form. The standard free energies of dissociation were found to be 8.27 and 9.52 Kcal/mole for the formation of the dimer and the monomer, and -3.70 kcal/mole for the formation of the octamer. This implies that at physiological concentrations, a significant amount of the hemoglobin has undergone association to this form.

Significance to Biomedical Research and the Program of the Institute: The techniques of analysis of interacting systems which have been developed are of applicability to a wide variety of problems beyond the analysis of the lens proteins. Of particular interest here are the possibilities of studying the interactions of a variety of proteins and other macromolecules of retinal origin with the visual pigments. These techniques have already been applied in the study on the self-association of the phospholipid lysolecithin.

The analysis of hemoglobin has extended our knowledge of the physical properties of this very important macromolecule. The discovery that hemoglobin can undergo association beyond the normal tetrameric form indicates a need for further studies on the degree of association of hemoglobin at physiological concentrations and the relationship of this association to the ability of the hemoglobin to bind oxygen.

Proposed Course of Project: It is planned to apply the developed computational techniques to appropriate problems of interest in vision science, extending these methods and developing new methods as needed.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 013(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Study on the Pharmacodynamics of Various Agents Affecting the Intraocular Pressure

Previous Serial Number: NEI(I)-70 0/OPH 013(c)
NDS(I)-56 0/OPH 301(c)

Principal Investigator: Frank J. Macri, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	1.0
Professional:	1.0
Other:	0.0

Project Description:

Objectives: To determine the pharmacodynamics of agents able to alter the intraocular pressure with a view toward finding more effective compounds and to possibly further the understanding of mechanisms which maintain the intraocular pressure.

Methods Employed: Studies are made on the enucleated, arterially perfused eye. Perfusate is channeled either through the ophthalmic artery in the cat to nourish the entire eye, or through one long posterior ciliary artery in the cat, monkey and rabbit to nourish the fore part of the eye which is responsible for formation of anterior chamber fluid. Drugs and other test substances are added to individual bottles of fluid which can then be introduced into the system by stopcock control. Temperature and rate of arterial flow are easily regulated. The rate of aqueous humor formation was determined by the use of an I^{125} tracer (RISA).

Major Findings: 1. An unpredictable and erratic number of perfused preparations were encountered during the year which failed to respond to drug administration. It was assumed at the outset that the lack of response was due to unphysiologic conditions or to some intrinsic toxin. A study was made to determine the role on the system of the gases: air, oxygen, oxygen and carbon dioxide in various quantitative concentrations and combinations. Alterations of gaseous concentrations had no salutatory effect in increasing

the responsiveness of the eye. The end result of our investigation indicated that the polyethylene tubing and bottles used in the system were releasing some toxin (probably plasticising material) which could render the preparation unresponsive. The perfusate set-up has been modified so that polyethylene no longer remains as a contaminating source.

Experiments currently being performed deal with the ability of autonomic nervous system drugs to alter the rate of aqueous humor formation. Additionally, similar studies are being performed with sympathetic and p-sympathetic nerve stimulation.

Significance to Biomedical Research and the Program of the Institute: The arterially perfused enucleated eye has permitted research to be done on the eye which heretofore could not be accomplished. Due to the abolishing of a vascular pulse, microelectrode determination of ERG potentials now have been accomplished on the retina. Physiologic and pharmacologic studies are also being performed on an essentially "living" eye without the complicating factors of a variable and drug responsive blood pressure and unknown nervous stimuli from the brain.

Proposed Course of Project: It is planned to devote most of our energies to determine whether aqueous humor formed in the isolated eye is similar to that of the intact eye. This will be done by studying compounds which are known to be secreted out of and into the eye. Use of metabolic inhibitors will also be utilized and leakiness of the blood-aqueous humor barrier will be determined. Should these findings indicate a normal physiologically functioning eye, drug studies will then be performed to determine how aqueous inflow and outflow can be altered.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 014(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: The Role of Vasculature in the Maintenance of Intraocular Pressure

Previous Serial Number: NEI(I)-70 O/OPH 014(c)
NDS(I)-59 O/OPH 600(c)

Principal Investigator: Frank J. Macri, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.0
Professional:	0.0
Other:	0.0

Project Description:

Objectives: To determine if and by what mechanisms the dynamics of the intraocular basculature affect the eye pressure and to demonstrate the pharmacologic response of this vasculature to certain drugs.

Major Findings: This project was put in abeyance. It is planned to continue this study using enucleated, arterially perfused eye. Problems encountered in this phase of research are outlined in Project 600(c). As soon as it can be ascertained that the eye is physiologically normal, this project will then be continued.

Serial No. NEI(I)-71 LVR 021(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Clinical and Research Pupillography

Previous Serial Number: NEI(I)-70 0/OC 021(c)

Principal Investigator: David A. Newsome, M.D.

Other Investigators: John Marquardt, M.D. (for problem #2 under "Objectives")

Cooperating Units: NIH-DCRT (Miss Mary Lee Dante)
Dept. of Neurology, Washington Hospital Center (Dr. Ramon B. Jenkins)

Man Years:

Total:	0.6
Professional:	0.6
Other	0.0

Project Description:

Objectives: Clinical pupillography provides documentation, localization and diagnosis of any lesions present in a patient's nerves of the pupillary light reflex pathway. Research problems involving the pupil in FY71 included: 1) examination of the pupillary sphincter muscle in patients with myasthenia gravis, a serious muscle-nerve disease; and 2) study of the effects of various detergents and bacteriostatic chemicals used in some commercial eye drops on the iris muscles in an attempt to learn more about the well-known but unexplained phenomena of pupil widening and lid droop associated with the use of steroid eye drops by patients.

Methods Employed: Pupillary movements in response to bright light flashes were recorded simultaneously from both eyes with the electronic infra-red pupillograph and then analyzed for abnormalities of size or contour. Because of the large amount of data in the study of myasthenic patients, a computer program was developed so that measuring, averaging, and statistical computations could be performed by machine. No previous programs existed that treat pupillographic data in its pure form.

Major Findings: 1) In FY71 46 patients were referred from the clinical services for pupillograms. Of this group, 75% had various lesions of the pupillary reflex arc.

2) Compared with an age-matched group of normals, 3 of 15 patients with well-documented myasthenia gravis had an abnormal amount of iris sphincter fatigueability. Whether the pupils are involved in this disease has been controversial for some time; this is the first study of the state of the pupils using such a refined measuring and recording technique.

3) In Rhesus monkeys, administering the detergents and preservatives that accompany dexamethasone (a steroid) in a commercial eye drop into the eye produced lid droop and pupillary widening, while the dexamethasone alone did not. There appeared to be a toxic effect on the iris muscles. These compounds are being studied in man using topical application.

Significance to Biomedical Research and the Program of the Institute: This study provides an accurate, objective method of diagnosing pupillary abnormalities thus improving patient care, and also expands knowledge of the condition of the iris muscles and nerves in various disease states and under the influence of various pharmacologic agents used by ophthalmologists.

Proposed Course of Project: Patients will be tested as they are referred from the clinical services.

Honors and Awards: None

Publications:

Newsome, D.: After-image and pupillary activity following strong light exposure. Vision Res. (in press)

Newsome, D.A. and Einaugler, R.B.: Tonic pupil following retinal detachment surgery. Arch. Ophthal. (in press)

Thompson, H.S., Newsome, D.A. and Lowenfeld, I.E.: The fixed dilated pupil. Sudden iridoplegia or mydriatic drops? A simple diagnostic test. Arch. Ophthal. (in press)

Serial No. NEI(I)-71 LVR 015(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Synthesis of Sugar-Containing Polymers in Retina

Previous Serial Number: NEI(I)-70 0/OM 015(c)
NDS(I)-68 0/OM 1531(c)

Principal Investigator: Paul J. O'Brien, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.8
Professional:	0.5
Other:	0.3

Project Description:

Objectives: Many cell interactions are mediated by the sugars on the cell surface. The same is true of interactions between cells and macromolecules and of intermolecular attractions. A number of events in the renewal of photoreceptor outer segments could be regulated in this way. One such process is the orientation and association of rhodopsin, a glycoprotein, with other molecules in the assembly of photoreceptor disc membranes. The purpose of this project was to develop an *in vitro* system in which to study the synthesis of the carbohydrate component of rhodopsin as well as to assess its role in outer segment renewal.

Methods Employed: Ordinary biochemical techniques were used, for example, incubation of retinas, cell fractionation, detergent extraction of membrane proteins and purification by column chromatography.

Major Findings: Radioactive glucosamine was incorporated into many components of the photoreceptor outer segment of the bovine retina. One radioactive component was shown to be rhodopsin following extensive purification and demonstration of its sensitivity to light. Time course studies indicated that at least some of the sugars are added to the rhodopsin polypeptide in the inner segment.

Significance to Biomedical Research and the Program of the Institute: These experiments demonstrate that an *in vitro* system can be used to investigate the details of the biosynthesis of sugar-containing macromolecules and that the sugar residues of rhodopsin may be necessary for its migration to the photoreceptor outer segment and subsequent incorporation into disc membranes.

Proposed Course of Project: Future studies will determine the subcellular sites at which all the sugar residues are added to rhodopsin, and the necessity of having a full complement of sugars on rhodopsin before it is capable of either binding retinal or forming disc membranes.

Honors and Awards: None

Publications: None

Serial No. NEI(I)-71 LVR 011(c)
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Biological Oxidations in the Retinal Pigment Epithelium

Previous Serial Number: NEI(I)-70 0/OCH 011(c)
NDS(I)-68 0/OCH 1530(c)

Principal Investigator: Hitoshi Shichi, Ph.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.2
Professional:	0.2
Other:	0.0

Project Description:

Objectives: This project is being terminated.

Serial No. NEI(I)-71 LVR 012
1. Laboratory of Vision Research
2.
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Biochemistry of Visual Pigments

Previous Serial Number: NEI(I)-70 O/OCH 012(c)
NDS(I)-69 O/OCH 1660(c)

Principal Investigator: Hitoshi Shichi, Ph.D.

Other Investigator: None

Cooperating Units: None

Man Years:

Total:	1.0
Professional:	1.0
Other:	0.0

Project Description:

Objectives: For an understanding of the molecular mechanism of night vision in which the glycolipoprotein rhodopsin is involved as the photoreceptor, elucidation of the chemical structure of rhodopsin is essential. Therefore, attempts were made to investigate a possible function of lipid in the structure of rhodopsin.

Methods Employed: Such biochemical methods as centrifugation, column chromatography and spectroscopic analysis.

Major Findings: (1) When rhodopsin is associated with the retinal outer segment membrane, phospholipid may be involved in maintaining a preferred conformation (native conformation) of opsin and the asymmetric structure of the chromophore of rhodopsin. (2) For the regeneration of rhodopsin from the bleached product, phospholipid is required; it probably stabilizes the native conformation of opsin which is essential for regenerability.

Significance to Biomedical Research and the Program of the Institute: In order that vision is not impaired, it is essential that rhodopsin is regenerated from the bleached product. The present finding on the phospholipid requirement for the regenerability of rhodopsin may provide a clue for clinical research on congenital night-blindness.

Proposed Course of Project: (1) Characterizarion of the nature of interaction between rhodopsin and phospholipid. (2) Elucidation of the chemical structure of rhodopsin.

Honors and Awards: None

Publications:

Shichi, H.: Circular dichroism of bovine rhodopsin. Photochem. Photobiol. (in press)

ANNUAL REPORT
July 1, 1970 - June 30, 1971

OFFICE OF BIOMETRY AND EPIDEMIOLOGY

Office of the Chief

The Office was created during the year and Harold A. Kahn reported for duty as Chief in January 1971. Eleven publications by Harold A. Kahn are included as part of the National Heart and Lung Institute Annual Report.

The Office began to provide strong statistical consultation to and work in close contact with the large collaborative studies of disease treatment and natural history of interest to the National Eye Institute. Fred Ederer, a biostatistician with considerable experience in clinical trials, was hired and assigned this area as his principal responsibility.

Statistical consultation was provided to intramural scientists and to the National Center for Health Statistics.

Section on Blindness Statistics

The Section continued its administration of the Model Reporting Area for Blindness Statistics (MRA). During the year the 1968 booklet on statistics for the MRA was completed and sent to the printer. A paper on "Estimated Statistics on Blindness in the U. S. - 1968" was prepared and is currently being revised prior to submission for publication. Staff of the Section provided consultation to the National Advisory Eye Council, to the National Center for Health Statistics, and to members of the NEI intramural staff. Visits were made to New York State and Tennessee to discuss MRA procedures.

Publications

Anderson, P.H. (Ed.): 1969 Conference of Model Reporting Area for Blindness Proceedings. DHEW, PHS, NIH. U. S. Government Printing Office, 1970

Anderson, P.H. (Ed.): Statistics for 1967 on Blindness in the Model Reporting Area. DHEW, PHS, NIH. U. S. Government Printing Office, 1970

ANNUAL REPORT
July 1, 1970 - June 30, 1971

OFFICE OF BIOMETRY AND EPIDEMIOLOGY

Section on Ophthalmic Field and Developmental Research

I. Personnel

During the past year, loss of positions vacated by departing personnel effected a continued reduction in staff. Dr. Michael Gilman, Ophthalmologist, completed his two-year assignment in June and will not be replaced. Dr. Albert York, who was transferred to the Section from the NEI Extramural Programs in September 1970, also completed his two-year military tour in June. At the end of the fiscal year, Mrs. Doris Collie, Medical Technician, and Dr. J. T. Schwartz, Section Head, will comprise the total professional staff of the Section.

II. Progress

New investigations have been curtailed in favor of advancing those studies currently underway. Present activity is outlined under the headings: (A) Twin Studies, (B) Instrumentation Development, and (C) Other Activity.

A. Twin Studies

Expansion of the local Twin Register for Eye Examinations during the past year was limited to twins having specific qualifications for participation in ongoing projects. Current investigations are as follows:

1. Effect of Treatment on the Progression of Myopia

Myopia or nearsightedness is the world's most common cause of defective vision. Clinical methods of controlling accommodation are employed in attempt to retard the progression of myopia but the effect of such treatment remains unsettled.

This is a prospective study undertaken to assess the effect of a specific treatment in retarding the progression of myopia. The study population consists of 25 pairs of young monozygotic twins who are similarly myopic. One twin receives standard spectacle correction as the control; the other is given specially prescribed bifocal spectacles and topical, short-acting cycloplegic eyedrops.

During the past fiscal year, photographic and diagnostic ultrasound systems were assembled in order to provide measurements of the size of intraocular compartments and the curvature of various refractive surfaces of the eye. Baseline measurements were obtained and treatment/control programs were begun. Participants in this study will continue under careful observation for three years.

2. Genetic role underlying parameters of ocular motility

This is a study of the relative roles of environment and heredity in the development of parameters of ocular motility such as vergence ability, heterophoria, and AC/A ratio. These measurements are undertaken among a select population of 60 monozygotic and 60 dizygotic sets of twins to provide comparison of concordance in order to estimate the relative role of genetic factors. Knowledge of the contribution of inheritance to AC/A ratio is important because of their intricate clinical relation to strabismus. It has been reported that approximately half of the cases of esotropia are associated with an abnormally high AC/A ratio and half of the cases of exotropia are associated with an abnormally low AC/A ratio.

This investigation was implemented and all clinical data were acquired during the past year.

3. Heritability of the effect of corticosteroids on intraocular pressure

This investigation is designed to examine a popular hypothesis that the ocular hypertensive response to topical steroids is inherited as a simple autosomal trait. This hypothesis bears directly upon a current and widely accepted concept which ascribes a genetic etiology for chronic simple glaucoma. If the findings of the present twin study indicate genetic determination of the steroid response, there would be need for further research to define thoroughly the relationship between steroid responsiveness and chronic simple glaucoma. If inheritance is found not to play a significant role, the evidence would suggest a need to reexamine the popular basic hypothesis of autosomal inheritance of the steroid response.

Data on ocular pressure response to steroid provocative testing have been collected among a study population of 158 participating twins. Extensive data were also collected on the metabolic and cardiovascular status of these subjects. Data analysis will be completed during the next fiscal year.

4. Study of strabismus among twins

This is a retrospective study undertaken among all strabismic twins identified in our local register. Among 52 sets of twins with strabismus, 45 had primary strabismus; 30 of these had esotropia and 14 exotropia. This is a larger series than any single study of strabismus among twins reported to date. The data supported a significant hereditary role underlying both esotropia and exotropia. An unexpected, yet interesting, finding was the presence of an abnormality of 3-dimensional vision among non-strabismic cotwins in monozygotic twinships discordant for esotropia. This finding bears importantly on current concepts of the pathophysiology and natural history of strabismus. Specifically, this observation is inconsistent with a popular theory that the motor deviation of strabismus precedes the development of sensory abnormality and that the latter is a secondary adaptation to the motor abnormality. This finding suggests instead that the sensory abnormality may in itself be inherited and contribute to the evolution of strabismus.

During the past year, twins of special interest received further examination. A final report is being prepared for publication.

5. Dermatoglyphics and the determination of twin zygosity

This study is directed toward establishing criteria for determining the zygosity of twins on the basis of fingerprint data. Finger and palm prints were prepared for over 1,000 members of the twin register. In collaboration with the National Institute of Child Health and Human Development, multiple dermatoglyphic characteristics will be studied in detail to determine the extent of congruency for monozygotic and dizygotic twins. Such analysis may permit a reduction in the probability of error involved in the diagnosis of zygosity by blood typing alone. This would be of value for studies of eye disease as well as other conditions.

6. Collaborative study of X_g^a blood group incompatibility in fetal loss

Findings of a recent twin study on X_g^a blood group incompatibility, undertaken in collaboration with the Children's Diagnostic and Study Branch, National Institute of Child Health and Human Development, were described in the last annual report. A protocol for a follow-up family study on X-chromosomal inactivation was prepared during the past year and will be undertaken in collaboration with the same coinvestigators.

7. The heritability of human salivary isoamylase

A study of the heritability of human salivary amylases was undertaken in collaboration with the Human Genetics Branch, National Institute of Dental Research. Results of studying salivary specimens from 183 pairs of twins suggested essentially complete genetic determination for one of eight separable amylase fractions. This pattern of heritability for the salivary amylases has not heretofore been described.

8. Collaborative study of genetic factors in cardiovascular disease

Medical History, physical examinations or clinical laboratory determinations relating to cardiovascular disease were obtained for over 80 pairs of adult twins. In collaboration with the Field Epidemiologic Research Section of the National Heart and Lung Institute, these data are being studied to evaluate the role of heredity with regard to clinical parameters of cardiovascular disease.

B. Instrumentation Development

This is a prototype development project undertaken to demonstrate our present capability for providing a television image of the ocular fundus in a fashion which will permit automated recovery and treatment of genuinely useful clinical information. The purpose, method of approach, and previous progress of this project have been described in earlier reports.

The contractor is presently working on the specific objective of providing and storing a high resolution fundus image and treating this image in such a manner that representation of the optic nerve head is computer-acceptable for the purpose of estimating the cup/disk ratio. This ratio provides information of fundamental value in the diagnosis and management of glaucoma. If this contract objective is accomplished, television ophthalmoscopy will hold demonstrated near term value for clinical practice and for population screening programs. Further, a successful system could serve as the forerunner for automated acquisition of other clinically useful measurements (e.g., vessel size, circulation time) requiring similar technical capability.

C. Other Activity

Collaboration, consultation and services rendered to other groups

The Head of the Section on Ophthalmic Field and Developmental Research serves as consultant to the Department of Ophthalmology, USPHS Hospital, Baltimore, Maryland; as Clinical Assistant Professor of Ophthalmology, George Washington School of Medicine, Washington, D. C.; and as consultant to the National Health Examination Survey, National Center for Health Statistics, HSMHA, PHS. He also serves on the

Committee on Standardization of Tonometers, American Academy of Ophthalmology and Otolaryngology. The Section currently undertakes active collaboration with sections of the National Heart and Lung Institute, National Institute of Child Health and Human Development, and the National Institute of Dental Research.

III. Proposed Course for the Section

When new investigations are permitted to be undertaken, following completion of current studies, the following are proposed for early consideration:

1. Collaborative study on the treatment of myopia

The treatment employed in our twin study on controlling the progression of myopia was selected as maximum therapy which would not interfere with ordinary daytime function of accommodation. Practitioners interested in this problem employ additional methods of treatment including deeper and longer-acting medications. A prospective clinical trial undertaken in collaboration with selected clinicians is proposed. The responsibility of this section would include study design, study management and clinical measurement of components of refraction.

2. Study of the heritability of components of refraction

This will be a comparison of the agreement among monozygotic and dizygotic twinships for total refraction, corneal power, anterior chamber depth, anterior and posterior lens curvature, lens thickness and axial length of the eye. The study population will consist of members of the twin register and will utilize those systems of ultrasound and photographic examinations which were assembled for the twin study on myopia.

3. Pilot study of viral antibodies in acute iridocyclitis

The cause of acute iridocyclitis remains an enigma. This is a study of the relationship between acute iridocyclitis and serologic evidence of previous or coexisting viral infection. A series of cases experiencing first episode of acute iridocyclitis will be identified and matched with appropriate controls. Acute and convalescent blood sera will be tested with a series of 40 virus antigens. Antibody response will be compared to identify viral agent(s) found in significant association with the disorder. This study to be undertaken in collaboration with the Laboratory of Infectious Diseases, Perinatal Research Branch, National Institute of Neurological Diseases and Stroke (# NEI9CF - 70 E 008).

Serial No. NEI (CF) - 70 E 001
1. Collaborative and Field Research
2. Section on Ophthalmic Field and Developmental Research
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Twin Register for Eye Examinations (TREE)

Previous Serial Number: NDS (CF) - 66 E 1254

Principal Investigator: J. Theodore Schwartz, M.D.

Other Investigators: Michael Gilman, M.D.
Albert T. York, M.D.

Cooperating Units: None

Man Years:

Total:	0.8
Professional:	0.6
Other:	0.2

Project Description:

Objectives: To maintain a local register of twins as a resource for investigations on the heritability of ocular characteristics, case-control studies and studies of the early natural history of chronic disorders.

Methods employed: This Section has compiled a register of over 700 pairs of monozygotic and dizygotic twins for the purpose of ophthalmic investigations. All twins reside in the metropolitan Washington, D.C. area. The twin population is interviewed and given a general eye examination. Blood type, fingerprints and ocular photographs are obtained. On the basis of information developed through the first examination, subsets of the twin population are selected to participate in specific investigations.

Major findings: Ocular abnormalities found among the twin population are being compiled. A series of twins with normal visual acuity and negative history for eye disease has participated in a study of the inheritance of the hypertensive reaction to topically instilled corticosteroid (steroid) eye drops. A subset of young identical twins concordant for myopia has been assembled to study the influence of treatment on the progress of myopia. A series of 52 twinships with strabismus has been identified for study of inheritance of this disorder. A series of adult monozygotic and dizygotic twins has been studied for inheritance of AC/A ratio and other parameters of ocular motility. Detailed descriptions of these investigations are given in separate project reports.

The following investigations are being undertaken in collaboration with other Institutes: (1) a study of fetal loss associated with Xg^a blood type incompatibility between mother and offspring, undertaken in collaboration with the Children's Diagnostic and Study Branch, National Institute of Child Health and Human Development, (2) a study of the heritability of blood lipid characteristics being undertaken in cooperation with the Molecular Disease Branch, National Heart and Lung Institute, (3) a study of genetic factors in cardiovascular diseases being undertaken by the Field Epidemiologic Research Section of the NHLI, (4) a study of the criteria for determining the zygosity of twins on the basis of fingerprint data, being undertaken in collaboration with the Children's Diagnostic and Study Branch, NICHHD, and (5) a study of the heritability of human salivary amylases being undertaken in conjunction with the Human Genetics Branch, National Institute of Dental Research.

Significance to biomedical research and the program of the Institute: Comparison of agreement among monozygotic and dizygotic twins with regard to physical characteristics is valuable as an indication of the relative roles of heredity and environment in the expression of these characteristics. This register serves as a resource to identify appropriate populations for such studies as well as investigations on therapeutic effectiveness.

Proposed course: It is proposed that this twin register continue to be maintained and expanded as a resource for direct and collaborative clinical investigation.

Honors and Awards: None

Publications:

Schwartz, J.T.: A twin register for eye studies and need for collaboration: In Gedda, L. (Ed.): Proc. First Int'l. Symp. Twin Studies. Published in Acta Gen. Med. et Gem. (Roma) 19:344-348, 1970.

Plato, C.C., Mann, J.D., Schwartz, J.T., and Leyshon, W.: The role of Xg^a blood group incompatibility in fetal loss. Twin Study. In Gedda, L. (Ed.): Proc. First Int'l. Symp. Twin Studies. Published in Acta Gen. Med. et Gem. (Roma) 19:235-237, 1970.

Wolf, R.O., Taylor, L.L., Niswander, J.D., and Schwartz, J.T.: The heritability of human salivary amylases. Arch. Oral Bio. (In press)

Serial No. NEI (CF) - 70 E 004
1. Collaborative & Field Research
2. Section on Ophthalmic Field and Developmental Research
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Effect of treatment on the progression of myopia

Previous Serial Number: Same

Principal Investigator: J. Theodore Schwartz, M.D.

Other Investigators: Albert T. York, M.D.

Cooperating Units: None

Man Years:

Total:	1.0
Professional:	0.75
Other:	0.25

Project Description:

Objectives: To assess the effect of a specific treatment in retarding the progression of myopia.

Methods employed: This is a three-year study among a population of 25 pairs of young, monozygotic twins who are similarly myopic. One cotwin receives standard spectacle correction as the control; the other is managed using specially prescribed bifocal spectacles and topical, short-acting cycloplegic eye drops. The essential advantage in working with MZ twins in this investigation lies in the complete match on genetic constitution for the treated twin and his cotwin control. Key biologic variables of age, race, sex, period of gestation and maternal age are inherently controlled as are certain environmental factors common to the shared domicile. The study population was selected from our Twin Register for Eye Examinations.

Historical data including maternal, perinatal, growth history, family history, diet, development and past medical and ophthalmic history have been obtained. Detailed general ocular examination was undertaken. Critical measurements include refraction, corneal curvature, corneal thickness, anterior chamber depth, anterior lens curvature, posterior lens curvature, lens thickness, vitreous length and overall axial length.

During this past year photographic and ultrasound systems were assembled for the purpose of measuring the size of intraocular compartment and the curvature of refractive surfaces of the eye. These

hardware systems were pretested, base-line measurements were obtained on participating twins, and the study population was placed on appropriate treatment/control regimes.

Major findings: Study in progress.

Significance to biomedical research and the program of the Institute: Myopia is by far the world's most common cause of defective vision. Among environmental factors of suggested etiologic importance, one widely held theme, recurrent throughout the literature, relates the progression of myopia to prolonged use of the eyes for near tasks. Methods of treatment have been directed toward limiting accommodation and the effort of near work. Although clinical impressions and published data regarding the effect of such treatment are promising, the role of controlled accommodation on the progress of myopia remains unsettled. This study will provide a careful appraisal of the effectiveness of a clinically acceptable method of controlling accommodation.

Proposed course: The study population will be re-examined at least twice per year for a minimum of three years.

Honors and Awards: None

Publications: None

Serial No. NEI (CF) - 70 E 003
1. Collaborative & Field Research
2. Section on Ophthalmic Field and Developmental Research
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: A study of strabismus in twins

Previous Serial Number: Same

Principal Investigators: Michael Gilman, M.D.
J. Theodore Schwartz, M.D.

Other Investigators: None

Cooperating Units: None

Man Years:

Total:	0.4
Professional:	0.3
Other:	0.1

Project Description:

Objectives: To study concordance rates for esotropia and exotropia in twins. To examine the role of inheritance in sensory anomalies in strabismus.

Methods employed: A retrospective study of 649 pairs of twins revealed 52 twinships with strabismus. The majority of these twins were re-examined with emphasis on parameters of extraocular muscle and 3-dimensional vision function.

Major findings: (1) Concordance rates for esotropia among MZ and DZ twin populations are consistent for inheritance causing motor anomaly of this type; (2) Concordance rates for exotropia are consistent for inheritance causing motor anomaly of this type; and (3) Analysis of MZ twins concordant and discordant for esotropia suggests that the ability to see 3-dimensionally may be under genetic influence.

Significance to biomedical research and the program of the Institute: These data suggest a hereditary etiology for both esotropia and exotropia. The finding of an abnormality in 3-dimensional vision in MZ cotwins discordant for esotropia suggests that fusion need not occur as a secondary adaptation to the motor deviation. Instead, the fusion abnormality may be inherited separately and contribute to the evolution of strabismus.

Proposed course: The discordant MZ esotropic twins are undergoing further motor and sensory evaluation in depth. A report is being prepared for publication.

Honors and Awards: None

Publications: None

Serial No. NEI (CF) - 70 E 005
1. Collaborative & Field Research
2. Section on Ophthalmic Field and Developmental Research
3. Bethesda, Maryland

PHS-NIH
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Twin study of parameters of ocular motility

Previous Serial Number: Same

Principal Investigators: Michael Gilman, M.D.
J. Theodore Schwartz, M.D.

Other Investigators: None

Cooperating Unit: None

Man Years:

Total:	1.0
Professional:	0.8
Other:	0.2

Project Description:

Objectives: To estimate the relative roles of heredity and environment underlying the development of parameters of ocular motility such as heterophoria, vergence ability, AC/A ratio.

Methods employed: A select population of 60 sets of MZ and 58 sets of DZ twins have been studied; each cotwin underwent measurements of heterophoria, vergence ability, and AC/A ratio determination. Concordance rates between MZ and DZ populations will be compared.

Major findings: In analysis.

Significance to biomedical research and the program of the Institute: Current evidence suggests that strabismus is largely inherited. By estimating heritability of characteristics of ocular motility, which appear related to strabismus, this study will provide direction for further clinical investigation of strabismus. Thus, if this study shows clinical parameters of ocular motility to be inherited, then their further investigation among families of propositus strabismus cases would be of interest. Such effort would be directed toward identifying matings which might hold a predictive relationship to strabismus.

Proposed course: Data collection has been completed. A report is in preparation.

Honors and Awards: None

Publications: None

Serial No. NEI (CF) - 70 E 002
1. Collaborative & Field Research
2. Section on Ophthalmic Field and Developmental Research
3. Bethesda, Maryland

PHS-NEI
Individual Project Report
July 1, 1970 through June 30, 1971

Project Title: Heritability of the effect of corticosteroids on intraocular pressure

Previous Serial Number: NDS (CF) - 69 E 1783

Principal Investigators: Frank H. Reuling, M.D.
J. Theodore Schwartz, M.D.

Other Investigators: Manning Feinlieb, M.D.

Cooperating Units: NAS-NRC Twin Panel
Epidemiologic Research Section, NHLI

Man Years:

Total:	0.2
Professional:	0.1
Other:	0.1

Project Description:

Objectives: To assess the role of genetic factors in determining the intraocular pressure response caused by topical application of steroid eye drops. Humoral and metabolic factors which may correlate with the steroid response are also being studied.

Methods employed: A sample of 79 pairs of monozygotic and like sex dizygotic twins over 15 years of age were examined according to a standard protocol. Dexamethasone 0.1% eye drops were instilled three times per day for four weeks and the examination is repeated. Data were gathered on family history of various diseases, various measures of intraocular tension before, during and after four weeks of steroids, and anatomical observations such as gonioscopy, corneal thickness, cup/disk ratio were recorded. In addition, blood chemistries including post prandial glucose and lipoprotein fractions were obtained. Physical examinations were performed by members of the Field Epidemiological Research Section of the NHLI.

The protocol for this study was approved by the NAS-NRC Follow-Up Agency which granted access to those twins in their panel who reside in the Washington-Baltimore metropolitan area. Five pairs of these twins are included in the study.

Major findings: In analysis.

Significance to biomedical research and the program of the Institute: If inheritance is found to play a major role in the transmission of the steroid response, this research would offer strong support for the need of further studies to define thoroughly the relationship between steroid responsiveness and chronic simple glaucoma. If inheritance is found not to play a significant role, the evidence would suggest the need to re-examine a popular hypothesis suggesting simple autosomal transmission of the hypertensive response to steroids. The efforts of two collaborating units, the Molecular Disease Branch of NHLI and the Field Epidemiologic Research Section of NHLI who are conducting studies on the twins participating in this study may, in addition to their own results, give some insight into the determinants of the steroid response.

Proposed course: Final data on 158 twins have been collected. Analysis of these data will be completed in the next fiscal year.

Honors and Awards: None

Publications: Reuling, F.H. and Schwartz, J.T.: Heritability of the effect of corticosteroids on intraocular pressure: In Gedda, L. (Ed.): Proc. First Int'l. Symp. on Twin Studies. Published in Acta Gen. Med. et Gem (Roma) 19:264-267, 1970.

CONTRACT NARRATIVE
Section on Ophthalmic Field and
Developmental Research
C&FR, NEI

JEFFERSON MEDICAL COLLEGE OF PHILADELPHIA (HSM 110-69-185)

Title: Television Ophthalmoscopy Development: Feasibility Testing for Geometric and Temporal Studies

Contractor's Project Director: Thomas Behrendt, M.D.

Current Annual Level: No additional funding.

Objectives: The contractor shall develop techniques of television ophthalmoscopy to improve and automate clinical tests outlined below (P4) which have application in basic research, epidemiology, and medical screening; design experimental approach to specifically determine which of these tests are most feasible and would be most productive in widespread use; then pursue and perfect the selected tests with knowledge and review of the Project Officer.

In performance of contract, the contractor shall:

1. Assemble suitable components of the instrumentation system including high-power strobe light source, fundus camera, television image tubes, scan generators, image monitors, signal amplifiers, and electronic recording devices.

2. Perform comparative evaluation of several types of television equipment, and develop electronics techniques appropriate to the ophthalmology tests listed below.

3. Conduct pilot clinical trials using human subjects to determine the suitability and acceptability of television ophthalmology in a clinical context.

4. Produce electronic images and data useful for objective analysis and measurement and document diagnostic parameters for the following clinical tests:

   (a) Geometric studies including measurement of optic nerve head parameters and retinal blood vessels.

   (b) Temporal studies including measurement of retinal artery blood pressure, pulse transmission time, circulation time, and fluorescein patterns.

5. Based on the results of items 1,2,3,4, the contractor shall:

   (a) Provide a detailed report of the television ophthalmoscopy equipment and techniques utilized.

(b) Delineate the type and extent of useful medical knowledge obtained in each clinical test attempted.

(c) Provide technical specifications for construction of television ophthalmoscopy systems by electronics consultants.

(d) Recommendations for optimal applications, widespread use, and further development of the television ophthalmoscopy system.

Major findings: The project director has devised a method of handling the televised fundus image which is capable of providing more descriptive information on geometric characteristics than is available using photographic techniques. This advantage results primarily from use of a return beam vidicon tube which is capable of extremely high resolution and stores the fundus image while it is converted into digital form. The digital representation is recorded on computer magnetic tape. A computer can then be used to manipulate the stored information. At present the data acquisition hardware is assembled and is operational.

Assembly of an information display system is in progress. Using this system, an operator will be able to change the magnification and contrast of the fundus image using selected portions of the stored information. By such interaction, geometric characteristics of clinical interest will be selected and portrayed in a form amenable to computer analysis.

Modifications of the system are proposed for similar treatment of time-dependent characteristics as described above.

Significance to NEI program and biomedical research: This is the first system designed to interface television ophthalmoscopy and computer image analysis. With this system, it will be possible to generate automatic measurements of selected characteristics such as cup/disc ratio and geometric characteristics of retinal vessels. This procedure will be directly applicable to clinical evaluation and population screening.

Proposed course of project: The contractor is presently finishing the original contract. Upon completion, results will be evaluated and further action decided.

ANNUAL REPORT
OFFICE OF INFORMATION
NATIONAL EYE INSTITUTE
July 1, 1970 - June 30, 1971

The Office of Information was established during the past year to disseminate information about Institute programs and activities and to increase public awareness of vision problems and how research can lead to their alleviation.

The Office also played a role in fostering the development of internal channels of communications within the Institute, and in encouraging the flow of information among the various NEI components. Editorial assistance was provided by the Office for a variety of administrative projects, including the preparation of materials used in conjunction with Congressional appropriations hearings and program planning exercises.

GENERAL

During the early months of the fiscal year, the Information Officer's time was spent primarily in the establishment of the Office, including the organizing of information sources related to the Institute's programs and the programs of other government and private organizations in the vision health field and on visual disease and current vision research, and the organization of information materials and files inherited from the NINDS Information Office. The Information Officer visited the public relations directors of the National Society for the Prevention of Blindness and Research to Prevent Blindness, two major voluntary organizations in the vision field, to determine the nature and extent of their activities and their potential relationship to the Eye Institute's efforts in this area. Liaison was also established with the AAO, AOA, and Metropolitan Washington Prevention of Blindness Society.

The most immediate initial need of the Information Office was the establishment of a system for handling a sizable number of public inquiries and Congressional correspondence that was being answered on an ad hoc basis by other Institute staff members, including the Director, and the proper disposition of the many daily telephone inquiries coming into the Institute. (A total of 568 written public inquiries and 1300 telephoned inquiries were handled during the year; 20 Congressional inquiries were answered. An estimated 1700 requests for publications were received and handled during the year.

A second immediate need was to assist the Director with preparation of a number of speeches to be given before various professional and voluntary organizations. A total of eleven speeches were prepared for the Director and his immediate staff during the year.

PRESS RELATIONS

The Information Office prepared seven announcements on the appointment of key Institute staff members and new members of the National Advisory Eye Council. Four other releases were prepared on research and program developments. The Office assisted reporters from The New York Times,

Washington Post, National Geographic Magazine, Medical World News, Associated Press, Hospital Practice, MD Medical Newsmagazine, Family Health, Reader's Digest, Good Housekeeping, and Family Circle Magazine in preparing articles related to the Institute's programs.

EXHIBITS

The Office prepared two exhibits during the year. One, intended as a general program exhibit illustrating the history, objectives, and research interests of the Institute, was presented at the 1970 Meeting of the American Academy of Ophthalmology and Otolaryngology. A revised version of this exhibit emphasizing laboratory rather than clinical research was prepared for the Spring 1971 Meeting of the Association for Research in Vision and Ophthalmology.

A second exhibit, designed to encourage vision screening by non-ophthalmic physicians, particularly for visual acuity and amblyopia in children and glaucoma in adults, was presented at the Annual Meeting of the Medical Society of the State of New York and the American Medical Association Spring 1971 Meeting in Atlantic City. The latter exhibit was also loaned to the Prevention of Blindness Society of Metropolitan Washington for use in conjunction with their glaucoma screening activities.

PUBLICATIONS

In addition to the two professional monographs and two lay interest publications inherited from NINDS and the PHS Chronic Disease Program, the Office prepared a fact sheet on the Institute for general distribution and to be used with its program exhibit. Another fact sheet providing non-ophthalmic physicians with detailed procedures for visual screening of children and adults to be used in conjunction with the screening exhibit was prepared.

A series of contracts was awarded for the drafting of manuscripts on 16 public information pamphlets on a variety of vision related topics. A typewritten fact sheet on contact lenses was developed as an interim measure to handle requests from the public on this subject. Stocks of publications from other government agencies and voluntary and professional organizations relating to the Institute's programs were also obtained and maintained for the benefit of visitors to the Office.

RADIO AND TELEVISION

The Office arranged for the interview of the Institute Director on the Discussion: NIH program, broadcast locally on radio station WGMS. Two radio 60-second spot announcements, one on glaucoma, the other on amblyopia, were prepared for inclusion in the regular Search for Health Radio Features, prepared centrally at NIH. Arrangements were made for the Institute Director to appear locally on three segments on the NIH Reports television program (WRC-TV) and to be interviewed on the WTTG-TV Panorama Show.

MISCELLANEOUS

The Office prepared 5 articles for the NIH Record. It drafted a Save Your Vision Week message for President Nixon along with two other Presidential messages and one Secretarial. Four Weekly Reports to the Deputy Director for Science, NIH originated in the Office. NEI contributions to the NIH Almanac, Scientific Directory and Bibliography, and DHEW Annual Report were prepared. The Office developed and maintains a mailing list for keeping interested persons and organizations informed about Institute activities.

PROBLEMS

The Office's main problem is that of staff. Presently the Information Officer and his Secretary-Assistant comprise the Office, which limits its activities to those things which "must be done". There is little time for planning and launching new projects which would further the information objectives of the Institute. In particular, additional staff would enable the Office to cover and publicize better the activities of the Intramural Research program which is now almost entirely neglected; follow more closely the activities of Institute grantees to keep better informed of current research developments and to encourage public release of new developments; explore ways of utilizing the broadcast media more effectively; and in general to plan a more coordinated and less ad hoc public information program.

One specific major problem which is foreseen for the coming fiscal year is the storage and distribution of the 16 public information pamphlets we will be publishing. Without any attempt at promotion, the Office already handles a large number of requests for publications each week which are the result of past promotion activities by the agencies which originally published them. The projected increased demand for our own leaflets will overburden the present staff.

ANNUAL REPORT
JULY 1, 1970 THROUGH JUNE 30, 1971
EXTRAMURAL PROGRAMS

PROGRAM REPORT

The Extramural Program area is responsible for planning, developing, evaluating, and administering the National Eye Institute's grants program. Through its grants and awards, the Institute supports research and training on problems related to the prevention, diagnosis, and treatment of visual disorders; on problems related to rehabilitating the blind and the partially sighted; and on fundamental studies essential to the understanding of the eye and the visual system. Since the total program is relatively small, it has not been subdivided for administrative purposes into program areas; however, the research supported by the Institute emphasizes retinal, choroidal, and corneal disease, as well as studies on glaucoma, cataract, and disorders of the visual system. The Institute also supports research on uveitis, chemical toxicity, physical trauma, developmental defects, and on disorders of the oculomotor system.

RESEARCH HIGHLIGHTS

For the purpose of this report, research highlights are categorized under the following areas; diseases of the retina and choroid, corneal and infectious diseases, glaucoma, cataract, the visual system, and oculomotor and other disorders. These highlights represent a selection of the research that has been published by NEI supported investigators during the past year. The selection does not attempt to include all of the significant work published, but instead attempts to represent examples of the research sponsored by the Institute.

Diseases of the Retina and Choroid

The retina is the photoreceptive tissue lining the eye that converts light into nervous impulses which are transmitted to the brain creating the sense of sight. It is nourished by the choroid, the adjacent specialized vascular tissue exterior to the retina. Disorders of these tissues include: retinal detachment, diabetic retinopathy retinal and macular degenerations, congenital anomalies and circulatory disturbances.

In retinal detachments, the inner and outer layers of the retina separate. An increasing visual loss accompanies progressive detachment. Progression of this disorder can often be halted by using laser beams or photocoagulators to "spot weld" the retina to the underlying tissue. It is desirable, if possible to move the detached tissue back into place and "spot weld" it there. An investigation using ultrasound has shown that this form of energy can be used to move detached retinas. 1/ Equipment that concentrates and propagates directed ultrasonic energy was first used to move air bubbles introduced into the eyes of rabbits. These experiments

demonstrated the radiation pressure effect of ultrasound within the vitreous cavity. When subsequent studies of the eyes showed there had been no adverse effect on the ocular tissues, attempts were made to move retinal detachments in humans. In detachments complicated by a retinal tear, ultrasound successfully moved most detachments back into place where they could be surgically reattached. Less success was achieved where no retinal tear was observed.

A simple office procedure was developed which can distinguish between retinal abnormalities that have their origin in deficient photopigment (rhodopsin) and those that have their basis in neural abnormalities. 2/ An instrument which flashes a distinctively shaped ray of light upon the retina is used. Repeated flashes of the distinctive pattern are focused on the retina of patients whose eyes have become adapted to the dark. Subsequent examination of the retina should reveal a distinctively shaped area of bleached pigment. Failure to detect such a bleached area in the absence of other explanations is indicative of a photopigment abnormality.

An investigation of the drug chloroquine provides an experimental model to evaluate drugs that are potentially toxic to the retina. 3/ Chloroquine and hydroxychloroquine are drugs that have been used for antimalarial medication and for the therapy of rheumatoid arthritis. Taken in large amounts over extended periods these drugs can induce retinal damage in man. The length of time between onset of therapy and the onset of visual loss, and the low incidence of retinal damage in patients receiving these drugs hinders studies on ways to influence this undesirable side effect. Laboratory animal investigations are also difficult because of the prolonged treatment necessary to produce retinal damage and because large dosages are systemically toxic. These difficulties were overcome when it was discovered that doses of chloroquine that are small in relation to the entire animal but large in relation to the tissues of the eye, produce retinal toxicity when injected directly into the eye. Experiments with cats showed that doses which are at the threshhold for inducing retinal changes in the injected eye do not produce any overall toxicity in the animal or in the uninjected eye. At threshhold doses changes occur in the eye's electrical potential within 6 to 24 hours after the injection. Retinal damage detectable by light microscope studies occurred about 40 hours after injection. With this technique, laboratory investigations of short duration can be brought to bear on the nature of chloroquine retinal damage. Indeed, the technique has already been used in an electron microscope study of ultrastructural changes induced in the retina by chloroquine. 4/ This study showed that the first and by far the most serious damage occurs in the photoreceptor cells. In these photoreceptors most of the damage is confined to the part of the cell that contains the light sensitive pigments.

A strain of rat with an inherited retinal degeneration is being studied extensively in the hope that it may become an experimental model for human retinal pigmentary degeneration. One study of these "dystrophic rats" described the progression of retinal degeneration. 5/ In normal animals rod-shaped photoreceptor cells continually produce photosensitive materials, and the excess material is removed by an adjacent tissue, the pigment

epithelium. In the dystrophic rat the pigment epithelium is unable to remove the excess, and the material accumulates as an abnormal extracellular layer. Initially the photoreceptor cells continue their production of photosensitive material; but as the layer of excess material accumulates, the new production slows and finally ends. As the abnormality progresses, the removal of excess material finally begins (through the inner retinal layers which do not ordinarily provide this function) but not before degeneration has become irreversible.

A genetic investigation of a North Carolina family spanning four generations revealed a specific pattern of inheritance of macular degeneration. [6] Seventy members of the family were examined and 25 of these persons were afflicted. Variation in the condition defied classification by age of onset, clinical appearance, or visual function, but taken all together the analysis showed that the condition can be attributed to a dominant gene that is inherited independent of sex. Incidentally, in this family, and in two others, macular degeneration was associated, although not invariably, with another genetic disease, amino-aciduria, which is detected from urine samples. An understanding of the genetic basis for the inheritance of macular degeneration provides important information that can be useful in genetic counseling.

Retinal disorders characteristically occur in patients with sickle-cell hemoglobin C disease. Intraocular bleeding and retinal detachment are the ultimate result. The natural history of this disorder was studied in 24 patients. [7] The progressive stages are: (a) the obstruction and closing off of retinal blood vessels; (b) a fusion of the retinal blood vessels; (c) new blood vessel development and a growth of fibrous tissue; (d) intraocular bleeding; and (e) retinal detachment. A finer classification of the progression of the disease within these stages was also accomplished. Knowledge of the natural history of retinal disorders in these patients and the classification of the progression of their retinal pathology provides the basis for further investigation and will be useful in evaluating therapeutic procedures.

Fluorescein angiography is a new and powerful diagnostic tool which permits direct viewing of blood flow within the vessels of the retina and choroid thereby making it possible to differentiate various diseases of the retina and choroid and to locate the site of any leakage or blockage of blood flow in these tissues. In this technique, fluorescein is injected into the veins. Subsequent observations can trace the movement of fluorescein in the vessels of the eye. Several studies published during the past year illustrate the value of this technique. In one such study, cavernous hemangioma could be distinguished from other vascular malformations of the retina. [8] Cavernous hemangioma is a retinal tumor composed of thin walled saclike structures filled with dark venous blood. This tumor has the appearance of a cluster of grapes attached by a broad base to the retina or to the optic nerve head. The fluorescein study showed little or no blood leakage from these tumors and indicated that they have minimal growth potential. Since these tumors may not require therapy, distinguishing them from retinal lesions that require therapy is very important. In another study, fluorescein angiography

was valuable in describing various stages of presumed ocular histoplasmosis as an objective way to document changes in hemorrhagic lesions during trials of different therapies. [9] Nonhemorrhagic lesions could be identified as areas of retinal swelling in which there was a late diffusion of fluorescein. In large hemorrhagic lesions a network of fine capillaries was always demonstrated and changes in the size and shape of the lesions corresponded to changes in this capillary network. Fluorescein was used in two studies of retinitis pigmentosa, an abnormality of a pigmented layer in the eye. One study showed that a diffuse mottled hyperfluorescence of the retina combined with extensive abnormalities of the retinal blood vessels and occasional areas of exposure of the deeper, larger choroidal vessels identified the disease.[10] In the other study, fluorescein angiography was helpful in defining the relationships between three different aspects of retinitis pigmentosa: (a) changes in the retinal circulation, (b) changes in pigment distribtuion, and (c) changes in choroidal circulation. [11] All advanced cases showed a marked degeneration of small choroidal blood vessels in the region of pigment changes. Continuing studies will investigate whether early changes in these vessels always portend pigmentary changes. Fluorescein angiography has also been used to interpret the functional and pathological significance of choroidal sclerosis, a condition in which choroidal blood vessels are more visible than usual and vary considerably in size, color, and visibility at different sites. [12] Patients with choroidal sclerosis not associated with visual loss usually had normal fluorescein angiograms. Patients with choroidal sclerosis associated with visual loss always had abnormal fluorescein angiograms. A study of the clinical course and pathogenesis of macular edema also made use of fluorescein angiography. [13] This swelling of the central retina frequently occurs after the removal of a cataract. The swelling may cause a rupture of the vitreous, a jellylike substance that fills the eye. The accurate diagnosis of this condition is enhanced by observing a characteristic fluorescein pattern.

Corneal and Infectious Diseases

The cornea is the tough transparent outer coat that covers the front of the eye. All light that stimulates vision passes through this perfectly transparent tissue. It not only admits light but also focuses the light rays on the retina. In addition, the cornea serves as a protective layer for the delicate inner parts of the eye. Fundamental studies on the structure and metabolism of the cornea, on the control of ocular infections, and on corneal healing aim to avoid the development of cloudiness, scarring, and surface irregularities in this organ. Other studies attempt to perfect corneal transplantation, already a successful surgical procedure.

Measuring the permeability of the innermost layer of the cornea may become a useful technique in diagnosing corneal disease. [14] The method monitors the movement of fluorescein through the corneal endothelium after anesthesia. Fluorescein is introduced into the eye by iontophoresis. An electrode made of an agar gel and fluorescein is held in contact with the patient's cornea. The other electrode, a metal rod, is held by the patient and a very weak current is passed through this closed circuit. Fluorescein

permeates the cornea and the aqueous humor, a fluid located directly behind the cornea. A steady state is reached in several hours in which fluorescein leaves the cornea and the aqueous humor at equal rates. The concentration of fluorescein in the cornea and in the aqueous humor is determined at repeated intervals with an objective slit-lamp fluorophotometer. The movement of fluorescein measures the permeability of the corneal endothelium. Permeability measured this way was found to be remarkably invariable in the corneas of rabbits and normal living patients. Permeability did not change with age nor were there differences between the two eyes of normal patients. However, preliminary observations in patients suggest that with corneal disease present the endothelial permeability increases markedly. Furthermore, the increase may preceed changes in corneal thickness, a common indication of corneal disease.

One of the most serious complications of herpetic keratitis, a viral infection of the cornea, is an ulceration which may lead to scarring of the corneal surface or even to holes in the cornea. The cause of this damage is poorly understood since the virus itself does not have the ability to degrade collagen; the substance which composes the cornea. Recent findings indicate that infection induces the corneal production of an enzyme, collagenase, which can break down the corneal substance. 15/ Corneal tissue from diseased and healthy animal eyes were placed on a collagen gel, and the condition of the collagen gel near the tissue was observed. The gel remained intact around healthy corneal tissue and degenerated around diseased tissue. The mechanism of collagenase release may ultimately explain why some infections develop ulcerations and others do not. In the meantime these findings offer the possibility of using collagenase inhibitors in combination with specific antiviral drugs in the therapy of viral infections of the cornea.

Two drugs, adenine arabinoside and cytarabine, showed promise for the chemotherapy of an ocular viral infection. 16/ Adenine arabinoside was extremely effective in preventing the spread of herpes virus in artificially infected laboratory animals and the drug showed no apparent ocular or systemic toxicity. Cytarabine is a highly effective antiviral drug but causes corneal toxicity when used alone in effective dosages. However, preliminary observations indicate that adenine arabinoside in combination with non-toxic doses of cytarabine may be a usable and effective therapy for deep seated viral infections.

Inducers of interferon, a substance produced within the body that helps fight off virus infections, are being widely studied. One of these inducers is a synthetic molecule of ribonucleic acid (RNA) called polyinosinic-polycytidilic acid (poly I-C). One investigation indicated that repeated topical ocular applications of poly I-C failed to increase its effectiveness in rabbits and that the therapeutic effect in man is very weak. 17/ However, another investigation showed that injections of poly I-C into rabbits induced the production of interferon which appeared in blood serum and in the fluids of the eye. 18/ Intraocular levels of interferon were greatest when the inducer was injected into the blood stream, however, intraocular injection showed that the eye itself is a source for interferon production. Continuing

investigations are aimed at the possible use of producing interferon within the eye during viral infection of the iris and of the uvea.

An animal study of a corneal fungus infection indicated an effective therapy which combines treatment with pimaricin and potassium iodide with a weak solution of the corticosteroid, dexamethasone. [19] Previous work had shown that the drug, pimaricin, is experimentally and clinically effective in the treatment of superficial fungal infections of the cornea. Severe intraocular inflammation accompanies the fungal infection and when steroids are used alone as anti-inflammatory agents they aggravate fungal infection. Nevertheless, a therapy was worked out in which the fungal infection is controlled by pimaricin and the inflammation is suppressed by a diluted topical application of corticosteroid.

The hydrophilic or soft contact lens was used successfully as a healing aid for corneal disorders. [20] Soft contact lenses made of loosely cross-linked polymers of 2-hydroxyethyl methacrylate were originally developed as an alternative to conventional contact lenses to take advantage of their pliable and non-abrasive characteristics. Soft lenses were used to treat patients suffering from superficial and painless corneal ulcers which had not responded to conventional therapy. Since previous animal experimentation indicated that these lenses aggravate active corneal infections, only patients whose ulcers were free of bacteria and virus were treated. The treatment started with the removal of the outer layer of the cornea. The corneal surface was then washed and the soft lens was put in place. The lens remained in place for 1 week, while the corneal tissues gradually regenerated. Complete regeneration usually occurred within 5 days. The ulcers healed completely, and comfort was restored. The exact way in which soft lenses aid healing is uncertain, but they may serve as a bandage, shielding the ulcer from irritation as well as providing a structural framework for the regenerating tissues.

Another study explored the use of silicone rubber as a material suitable for contact lenses. [21] This study indicated that silicone rubber lenses do not induce clinical signs of irritation and do not interfere with the metabolism of the cornea. Conventional contact lenses may hamper metabolism of the cornea by interfering with the normal supply of atmospheric oxygen. A cornea deprived of its oxygen supply becomes thickened and less transparent. Silicone rubber lenses were tested because this material is highly permeable to oxygen and carbon dioxide, and the transmission of light is about equal for all colors of the spectrum. The encouraging results of this experimental study on rabbits point to clinical trials to determine if silicone rubber lenses will be amenable to clinical standards.

An instrument was developed that holds promise for simplified examinations of human corneal material before it is used for corneal transplants. [22] A number of criteria have been proposed and are in use for assessing the viability of donor corneas, but these assessments are carried out on samples removed from the donor cornea. A "specular" microscope now offers a means to critically examine the donor cornea intact. It is possible with this microscope to measure the thickness of all the layers of the cornea

without exposure, handling, or washing of an intact globe. The effectiveness of these measurements as a routine eye bank method for evaluating donor eyes is being investigated.

A study of several techniques for the freeze-preservation of corneas for transplantation identified a successful method. [23] The best results in experiments using rabbits' eyes were obtained with a mixture of 7 percent dimethylsulfoxide and 10 percent sucrose as a vehicle in a liquid nitrogen freezer. Frozen corneas were thawed in normal serum or albumen at room temperature. Transplants with these corneas gave good experimental and clinical results. Freeze-perserved human corneas used in more than 120 transplants gave clinical results as good as transplants of refrigerated tissues. Although much remains unknown of the metabolism, biology, and immunology of freeze-perserved corneas, the methods may no longer be considered experimental. True tissue banking can now be done thus providing corneas for transplantation when needed and thereby eliminating the waste of donor materials.

In current practice corneal surgeons prefer that donor eyes come from relatively young individuals. [24] A recent study indicated, however, that the success of complete corneal transplantation was independent of the age of the donor. Approximately 400 transplantations were studied. Clear grafts were obtained in 89 percent of the cases that had favorable prognoses. Clear grafts were obtained in 69 percent of the cases that had moderately favorable prognoses, and clear grafts were obtained in 50 percent of the cases which had unfavorable prognoses. Although the age of donors ranged from below 50 years to over 70 years, success of transplantation was unrelated to donors' ages. These results should make more corneas available for transplantation.

Glaucoma

Glaucoma is a group of diseases characterized by the elevation of pressure within the eye. The pressure elevation in suseptible individuals leads to damage of the optic nerve and if left untreated may progress to total irreversible blindness. The loss of vision can be arrested or prevented by medication or surgery. Glaucoma is initially symptomless, however, and an irreparable visual loss may occur before the disease is detected. An important goal, therefore, is the early identification of individuals likely to lose vision so that treatment can be initiated.

Retinal blood vessels converge at the optic disc, a circular area in the back of the eye that is devoid of photosensitive cells and where retinal nerve fibers also converge as they leave the eye. The size of the depression in the center of the optic disc, the physiological cup, appears to be genetically determined. The relation of the size of the cup to the size of the disc, the cup-disc ratio, was studied as an aid in early diagnosis of glaucoma. [25] The cup-disc ratio was observed in individuals free of ocular complaints who had glaucomatous visual field defects. Such individuals exhibit the earliest clinical stage of glaucoma. The study showed that enlargement of the optic cup occurs early in the course

of glaucoma and parallels the magnitude of the vision defect. Furthermore, in the early stage of glaucoma, the cup-disc ratio is characteristically unequal in the two eyes. The ratio, therefore, becomes an important tool in suspecting and detecting this otherwise symptomless disease. The simplicity of this procedure should be invaluable in reducing the frequency of the false positive tests common to current procedures.

Corticosteroids, a group of related compounds used to suppress inflammation, are known to increase intraocular pressure in the relatives of glacuoma patients. A corticosteroid provocative test has been developed which is becoming an important tool in investigating the genetics of primary open-angle glaucoma and in identifying glaucoma-prone individuals. Questions have been raised, however, as to the possible irreversible intraocular pressure and visual field changes induced by the short-term medication. A recent study confirmed the safety of this provocative test. 26/ In 500 consecutive provocative tests with topical steroids,all elevations of intraocular pressure were completely reversible. Pressures returned to normal within 10 days in 98 percent of the patients, and within 21 days all eyes were back to normal. The visual field was observed in 70 patients in which pronounced elevation in intraocular pressure was provoked. In all instances, changes recorded during medication were promptly reversed when topical corticosteroids were discontinued. A 4-year follow-up indicated no detectable permanent changes. Nevertheless, it must be remembered that patients who are near-sighted or have a family history of glaucoma are especially prone to develop a rise in intraocular pressure in response to corticosteroid eye drops. The prolonged use of such medication to alleviate contact lens irritation and discomfort may lead to the development of glaucoma and cataracts in high risk groups. The need for precaution in prescribing corticosteroids for contact lens patients suffering from corneal irritation was illustrated by two case histories. 27/ Two young women developed glaucoma with visual loss and cataracts in both eyes after prolonged medication with cortisone.

Pressure within the eye normally increases as a response to drinking water. In patients with damaged optic nerves, this response is sometimes modified. To examine the role of the central nervous system in the control of intraocular pressure, studies were performed on rabbits in which one of the two optic nerves was severed. One study showed that neither the basic intraocular pressure nor the basic normal outflow of ocular fluids was affected by cut optic nerves. 28/ The response to drinking water and to oral doses of isosorbide, a drug whose osmotic effect reduces intraocular pressure, was also studied. Severed optical nerves did not change the effect of very large doses of water or of isosorbide. At lesser doses, however, eyes with severed nerves were less responsive than eyes with intact optic nerves. In another study, alcohol which also affects osmolarity and drugs known to change intraocular pressure by influencing the normal outflow of ocular fluids were studied. 29/ The effect of alcohol was altered in eyes with severed optic nerves. The condition of the optic nerve had no significant effect on the influence of pilocarpine, epinephrine hydrochloride, or acetazolamide. These experiments indicate that the central nervous system is involved somehow in the control of intraocular pressures but that

the involvement is limited to osmotic effects and does not impinge upon the facility of ocular fluid outflow.

An investigation of the effect of exercise on intraocular pressure in human beings confirmed that the pressure within the human eye falls after exercise. [30] The fall in pressure is accompanied by a significant rise in blood lactate, an increase in the osmolarity of blood plasma, and an increase in blood acidity. The largest drop in pressure and the greatest changes in blood chemistry occur immediately following exercise. Another study showed that similar results are obtained in rabbits, thus the rabbit provides an experimental model for further investigations. [31] Experimental alteration of blood osmolarity, lactate, and acidity implicated only changes in acidity and osmolarity in the fall of ocular pressure after exercise. The animal model may provide a means by which further studies can explore the effect of blood chemistry on intraocular pressure.

An acute form of glaucoma is caused by an anatomic abnormality in which the iris blocks the normal flow of ocular fluids. Very high intraocular pressures result. The abnormally high pressures can be eliminated by surgery which provides an opening in the iris. The use of laser beams to produce the opening was explored in rabbits, cats, monkeys, and autopsied human eyes. [32] The results suggest that ruby laser coagulation may be used effectively for this purpose in brown eyes but not in blue eyes. The argon laser also shows promise but has not been tried in blue eyes. Should laser surgery prove effective it would have significant advantages over the present procedures. Potentially it provides an opportunity to quickly abort an acute glaucoma attack. There would be no need to hazard intraocular surgery, and the procedure could be performed on an outpatient basis.

The use of alpha chymotrypsin, a protein-digesting enzyme in cataract extraction has been associated with elevated intraocular pressures. Glaucoma was produced experimentally by injecting the enzyme in the eyes of owl monkeys. Subsequent studies by electron microscopy showed that in glaucomatous eyes particles obstruct the channels through which ocular fluids normally pass. Most of the particles obstructing the outflow channels were identified as fragments of the tissues dissolved by the enzyme. [33]

An investigation of 64 patients with primary open-angle glaucoma who had undergone uncomplicated cataract surgery indicated that cataract extraction not only restores vision but improves the control of glaucoma and frequently reverses glaucomatous visual field losses. [34] It had already been demonstrated that glaucomatous visual field defects are reversible and that reverses are most likely to occur with the lowering of intraocular pressure when the field defect is of recent origin. In glaucoma patients who underwent cataract extraction, visual field recovery occurred in 15 of 21 eyes with defects of less than one year's duration. Only two of 19 eyes recovered visual field losses when defects were of longer duration than one year. As a result of these observations, cataract extraction where indicated has been used alone for patients showing progressive field loss and poorly controlled intraocular pressure. Previously such patients would have been candidates for primary glaucoma surgery or for combined cataract extraction

and glaucoma surgery. Subsequently, only two eyes, considerable fewer than one would have predicted, required glaucoma surgery.

Cataract

A cataract is a loss of transparency or a cloudiness of the ocular lens. The most common type is senile cataract which appears to be a part of the general aging process of the human body. Cataracts can also result from congenital defects, systemic and infectious diseases, and injuries to the eyes. The exact cause of most kinds of cataract remains uncertain, but changes in the normal balance of soluble and insoluble lens proteins seem to be involved. The development of a medical procedure for the prevention or treatment of cataract awaits a better understanding of the chemical nature of these opacities and of the metabolism of the lens. One such study was an investigation of the role of glutathione in lens metabolism. 35/ This compound is present in high concentrations in the lens, but its physiological role has never been clearly understood. A technique in which lenses are cultured in test tubes was used. By making specific additions to the culture media, and by observing uptake of radioactive tracer chemicals by the lens, inferences about the normal role of various chemical compounds were possible. The study indicated that glutathione may be necessary to maintain certain sulfhydryl groups on the lens membrane in a form required for proper membrane function.

The ocular lens contains an unusually high concentration of myoinositol, a cyclic sugar alcohol. The concentration is 70 times higher in the lens than in the aqueous humor, a fluid which bathes the lens. The role of this high concentration of myoinositol in the lens and the mechanism for maintaining this high concentration is under investigation. It has been shown that myoinositol is accumulated in the lens against a concentration gradient and that it is actively transported into the ciliary body and across the blood-aqueous barrier by a highly specific carrier-mediated system. The transport mechanism requires an expenditure of energy, is dependent on the potassium ion, is sensitive to certain drugs such as ouabain, and is sensitive to the sodium ion concentration. 36/ It has been shown that myoinositol is an essential requirement for the growth of normal cells in culture and for the preservation of the transport of amino acids by cells cultured *in vitro*. A similar function can be tentatively assigned to myoinositol in the lens. 37/

Rats on a high galactose diet have an increased tendency to develop cataracts, and the formation of galactose-induced cataracts in these rats can be delayed by the addition of a high level of fat in the diet. 38/ The mechanism for the delay in cataract formation appears to be an increase in the supply of acetylcoenzyme-A, and therefore, of energy. A high level of galactose appears to induce cataracts by reducing the amounts of at least two fatty acids, presumably in the lens fiber membranes, and the deficiency of these fatty acids may contribute to the rapidity of the formation of mature cataracts.

The ionic composition of the lens consists of a high potassium and a low sodium and chloride concentration in contrast with the bathing aqueous and

vitreous humors which have high sodium and chloride and low potassium levels. The lens is surrounded by a membranous noncellular capsule composed of a collagen-like material. This capsule is underlayed at the front of the lens by a single-layered cell membrane. The role of the capsule in maintaining ionic levels of potassium, sodium, and chloride were studied in cultured toad lenses. [39] It was already known that puncturing both the capsule and the outer single-cell membrane of the lens or the removal of both layers allows the free passage of ions between the lens and its bathing medium. The capsule was removed from toad lenses and the lenses were incubated in culture where they maintained their ionic concentration even more efficiently than in intact lenses. The addition of metabolic inhibitors altered the movement of ions, as would be expected, but surprisingly implicated both oxidative and anaerobic processes. Since the capsule is the principal site of cholinesterase in the lens, cholinesterase inhibitors were also introduced into the medium. Only one of these, demecarium, affected the movement of ions. The presence of this drug increased sodium gain in the lens (but not potassium loss) but did so in the presence or the absence of the capsule. Thus, demecarium's effect does not appear to depend on its ability to inhibit cholinesterase.

Rapid forming but reversible cataracts have been described in the lenses of experimental animals and have been attributed to excessive evaporation through the cornea. This explanation was reinforced by a study of the sodium, potassium, and water content of the lens and of the aqueous humor after the induction of these reversible opacities in hamsters. [40] Extreme dilation of the pupil by phenylephrine was accompanied by an increase in the sodium and potassium concentration and a decrease in the water content of the lens. There was a simultaneous increase in sodium concentration in the aqueous humor leading to the formation of a cataract 30 to 60 minutes after exposing the cornea to air. No such changes occurred when the eyelid remained closed. These observations, which were consistant with another similar but independent study, [41] suggest that evaporation through the cornea produces concentrated aqueous humor which then dehydrates the lens.

Although cataract surgery has become a safe and satisfactory practice, technical improvements in the procedure are still being achieved. A 5-year evaluation showed that an aspiration (suction) technique is superior to conventional surgery for removing cataracts in infancy, childhood, and adolescence and may also be used successfully in older patients. [42] The anterior chamber of the eye is entered through clear cornea with a small tapered knife. The lens capsule is cut open and the lens material is stirred for several minutes. The knife is withdrawn, a hollow needle is inserted and the lens material is sucked out. The aspiration technique generally leads to better visual results and fewer post-operative complications than conventional techniques. The procedure is done in one session. The small incision simplified post-operative care and shortens hospitalization. Congenital cataracts have long been a problem to ophthalmologists. Conditions that interfere with normal retinal image formation during infancy can cause irreversible vision loss. Ideally, congenital cataracts should be removed as early in life as possible, but the surgeon has been faced with the difficult question of when to operate and what surgical technique to use. Now

this single and simple aspiration procedure can remove the onus associated with congenital cataract surgery in the past.

Visual System

When light is flashed in an eye,there is an immediate electrical activity (the early receptor potential, ERP) which is associated with light induced changes in the photopigment rhodopsin. In a recent study, intracellular recordings of the early receptor potential were accomplished. 43/ The recordings were obtained from the retina of the gecko and other small vertebrates with exceptionally large retinal cells. The receptor cells were penetrated by potassium chloride filled, glass micropipette electrodes. After an electrical response was recorded, a dye was injected into the cell through the same micropipette, and a subsequent microscopic examination identified the cell and the segment of the cell from which recording had been accomplished. Recordings from the inner and from the outer segments of the photoreceptor cells showed no differences. However, recordings from within and from without receptor cells showed that the early receptor potential reverses its polarity the moment the plasma membrane is penetrated. There is also a difference in the amplitude recorded intracellularly and extracellularly. These observations suggest that the early receptor potential is produced either across the plasma membrane or across a structure electrically connected with the plasma membrane. Since the early receptor potential is a pigment response and the visual pigment is contained in the discs of the outer segment, the membrane origin of the early receptor potential indicates an electrical connection between the discs and the plasma membrane. This observation establishes a relation between plasma membrane structure and electrical function in photoreceptor cells.

The possibility that chemical mediators participate in the transmission of information in the visual system was studied in the eye of the horseshoe crab. 44/ The simplicity of this primitive marine animal's eye makes it very useful in vision research. Neural cells of the eye were impaled with glass micropipettes filled with a potassium chloride solution. These micropipettes served as electrodes to measure spontaneous and light stimulated nerve impulses and changes in membrane potential. Twelve substances were screened for their effect on the electrical activity of impaled cells. They were tested by replacing the sea water which bathed the preparation with sea water solutions of the chemicals or by applying these solutions at selected sites. The results implicated several chemicals in the transmission of information in the visual system. The chemicals implicated were: 5-hydroxytryptamine, gamma-aminobutyric acid, and the following catecholamines; dopamine, norepinephrine, and epinephrine.

Most studies of the electrical activity of the retina that employ intraretinal microelectrode recording have been concerned with the nerve impulse activity of ganglion cells and with a slow potential complex, the intraretinal electroretinogram(ERG). A different slow potential, the proximal negative response (PNR) which has previously been observed but largely ignored has now been intensively investigated. 45/ This electrical potential was studied by inserting microelectrodes into frog retinas prepared in a way that left the

posterior segment of the eye in the orbit. The electrodes recorded extracellular responses at various depths in retinas stimulated by flashes of sharply focused spots of light. The proximal negative response becomes confused with the electroretinogram when the stimulating light spot has a large diameter. However, by using a finely focused small spot, the proximal negative response can be definitely identified. It has a distinctive wave form, which has a transient "on" response characterized by a rapid and sharp negative initial peak followed by a gradual decay. A similar "off" transient peak is seen with moderate flash intensities. The proximal negative response has an amplitude-intensity relation in which the amplitude of the initial negative peak increases with increased flash intensity. The proximal negative response does not reverse its polarity during the retinal penetration of the electrode, but its amplitude has a maximum at a depth in the retina that corresponds to the layer of amacrine cells. These and other observations were cited as evidence that the proximal negative response is an extracellular field potential of the amacrine cells. These results should enhance future efforts to analyze relations between amacrine cell function and the visual process.

A study on the influence of oculomotor activity on visual processing was stimulated by the observation that the visual system must possess a mechanism for distinguishing between patterns of retinal activity produced by movement in the environment from the similar patterns produced by the movement of the eyes. [46] Maintenance of the spatial stability of the perceived visual world in the face of constant eye movements requires an interaction between incoming visual information and information related to the internal generation of eye movements. Previous studies demonstrated that light flashes delivered in conjunction with eye movements produce visual evoked responses (an electrical potential in the visual cortex of the brain induced by visual stimulation and recorded by electrodes placed on the scalp) that differ from visual evoked responses (VER) produced when no eye movements occur. Such changes presumably reflect alterations in the activity of individual neurons making up the visual pathway. A study of the electrical activity of single neurons in the lateral geniculate body (LGB) and of the retinal receptive fields of those neurons attempted to localize the origin of the VER changes that accompany eye movements. Cats were anesthetized. Movement in one eye was physically restrained, and this eye was presented with visual stimuli in the form of light flashes. Movements of the other eye which was shielded from light were induced by electrically stimulating the controlling lateral vestibular nucleus. Visual evoked responses elicited during eye movements and without eye movements confirmed previously observed differences. A microelectrode was then advanced into the lateral geniculate body (on the side of the head opposite to the eye receiving visual stimulation) until the activity of a single neuron was being recorded. The electrical activity of that neuron was examined and its retinal receptive field was mapped. Of the twelve units studied, none demonstrated any change in the organization of their receptive fields or in their electrical response to light stimulation as a result of eye movements. It was concluded that interactions must occur at a level central to the lateral geniculate body. Comparable studies of the visual cortex neurons in the brain are underway.

Another study investigated the electrical activity and the corresponding retinal field of neuron cells of the brain in an effort to understand binocular stereoscopic depth perception. [47] In man and in monkeys, the visual nerve fibers end in the so-called striate cortex of the brain, an area also known as area 17 of Brodmann. The interaction of binocular light stimulation was studied on cells in area 18 which is located just to the rear of the striate cortex. About half of the cells produced approximately equal electrical responses to light in either of the two separate eyes. There was approximate summation of responses when the two eyes are stimulated together, and the relative position of stimuli within the two receptive fields was generally not critical for a cell to show maximum response. The other half of the cells in area 18 were more specialized. These cells were designated "binocular depth cells." Stimulation of either eye separately gave no response or only weak responses in these cells, whereas stimulation of the two eyes together resulted in very brisk responses. The retinal fields of some of these cells corresponded exactly in the two eyes, but for others there was a disparity in the position of the two receptive fields. The displacement of the field in one eye relative to the field in the other was usually at right angles to the receptive field of orientation. Vertically oriented fields are thus hortizontally displaced, whereas with oblique fields there is a vertical component to the disparity. An examination of the frequency with which binocular depth cells are encountered suggests an organization in which cells representing a given stereoscopic depth are grouped together in columns and are separated from columns of cells concerned with orientation. This study was done with monkeys. Another study done with cats indicated a similar organization.[48] In a "constant depth column" the receptive fields of binocular neurons cover a small retinal area, and they are laid out in almost identical arrays in the two eyes. The depth column as a whole views a thin sheet of visual space, floating at some distance from the cat. In a "constant direction column," the fields of the binocular units are all superimposed on the retina of the eye on the opposite side of the head. The column as a whole views a cylinder of visual space directed toward that eye. This column arrangement is probably important for space perception. Activity in only one depth and one direction column would specify the orientation and the three dimensional locus of an object in space.

The electrical impulses that leave the eye through an individual fiber of the optic nerve are influenced by an area of the retina, the receptive field, that includes many photoreceptive cells. Generally, receptive fields are organized in such a way that impulses generated by the center of the field are antagonized by the responses of the surrounding area. If for example, light falling upon a certain center increases the electrical impulses sent through the corresponding nerve fiber, light falling on the surround would tend to diminish the impulses. Receptive fields without antagonistic surrounds have been reported in some visual systems but have not previously been commonly found in mammalian retinas. A study of the field organization of the rat retina, however, indicates the existence of such fields. [49] Microelectrodes were placed in individual fibers of the optic nerves of anesthetized rats. Light spots were projected on a screen in the rats' line of sight. The receptive field of the nerve fiber that had been impaled by the microelectrode was located by moving the light spot until

an electrical response was recorded. The size of the receptive field was determined by varying the size and intensity of the spot. The nature of the antagonistic surround was measured by varying the intensity of the light stimulating the center and of the light stimulating the surround. The results indicated great variability in the antagonistic effect of receptive field surrounds. In the rat, some ganglion cells have no demonstrable antagonistic surround. Others have surrounds which are effective in antagonizing the response of the center. Still others are so effective that when their activity is integrated over the entire area of the receptive field they dominate a center.

Another investigation of receptive fields was concerned with the sensitization effect. [50] The threshold required for a small spot of light on the retina to stimulate an electrical response is influenced by the pattern of illuminance falling on the surrounding area of the retina. For example, the increment threshold for a small spot of light located at the center of an illuminated disc varies with the diameter of the disc. When this phenomenon was studied in several different subjects, consistent differences between the subjects were found in the disc diameter necessary to produce a maximum increment threshold. For at least some subjects, this peak diameter also seems to shift as the illuminance level changes. These individual differences which will have to be taken into account in future studies of the sensitization effect probably result from differences in retinal image quality and differences in the spatial extent of the centers of receptive fields.

A coloring naming study was directed toward a better understanding of the mechanism for color vision in humans. [51] Previous reports indicated that the central fovea, the retina's center of greatest visual clarity, is sensitive to only two of the primary colors. The specific form of this dichromacy resembled tritanopia or "blue-blindness." This observation rose from experiments in which individuals were asked to match colors and is open to two interpretations; (a) there is a virtual absence of blue receptors in the fovea, and (b) the apparent "blue-blindness" results from a kind of adaptation in the blue system in which discrimination is impaired by prolonged fixation. A color naming experiment was used to test the adaptation hypotheses because it is a sensitive method, it utilizes short test flashes to which the subject does not have time to adapt, the flash can be superimposed on backgrounds of various colors so that the general state of adaptation can be manipulated, and the effects of fixation eccentric to the central fovea can be measured. Subjects were asked to fix their gaze on a gap in a thin vertical luminious line in an otherwise dark field. They were then asked to identify flashes of colored light projected in the gap or to its right or to its left. Repeated trials gave frequency data which when analyzed indicated that the central fovea is not dichromatic. Neither does adaptation arising from the steady fixation required to make color matches explain all of the features of foveal and small-field "blue-blindness." Apparently blue receptors are scarce but not entirely lacking in the central fovea.

Oculomotor and Other Disorders

Normally the fixation of an eye is such that the image of a visual object falls on the fovea, the central area of the retina that provides clearest vision. In strabismic or amblyopic eyes, fixation may place the image of a visual object elsewhere. Fixation was studied in patients with strabismic amblyopia, that is in patients with crossed eyes and impaired vision unrelated to any other apparent anomaly. [52] The study was intended to clarify the extent to which fixation changes occur in different positions of gaze and also whether such changes are related to muscular deficiency and changes of visual acuity. Fixation and visual acuity were determined with the eyes in a primary position and also at several different directions of gaze. The study revealed that changes of fixation occur frequently when the eye moves away from the primary position. These changes of fixation were explained on a mechanical basis rather than by implicating a hypothetical sensory feedback mechanism. Thus, in patients with a deficiency of motor function, fixation becomes more eccentric as the gaze changes because the eye cannot be moved far enough to keep the image of the visual object on the fovea or, in the case of an amblyopic, on the area of eccentric fixation to which the reflex has become adapted. In patients that exhibited changes of fixation at different directions of gaze, visual acuity was not always altered by a shift of fixation toward or further away from the fovea. In amblyopic eyes, therefore, visual acuity and fixation behavior cannot generally be correlated. This point is of utmost importance in planning surgery for eccentric fixation. Unless it can be shown that a significant improvement occurs in any position of gaze because fixation is closer to the fovea, surgery is unwarranted.

Egocentric localization pertains to optic localization of visual objects in their relation to the observer's body and the observer's tactile, visual, and conceptual image of his own position. Past-pointing is an error of egocentric localization observed in patients with recent paralysis of extraocular muscles. If the patient while his sound eye is covered is asked to point toward an object located in the field of action of the defective muscle, his finger will point beyond the object and toward the field of action of that muscle. Past-pointing is a clinical phenomenon which usually disappears after several weeks. It is of limited diagnostic value but of considerable fundamental interest. A recent study helped to distinguish between theories advanced to explain this phenomenon. [53] Errors of egocentric localization were studied in patients with extraocular muscle paralysis of recent onset. The retinal position of an image was also recorded when afflicted eyes were turned to the right or to the left. The results were contrary to the widely held view that the angle of past-pointing is determined by the degree to which the image on the retina is removed from the fovea. The study supports a theory that egocentric localization of objects in space is approximately correct as long as there is no discrepancy between the nervous energy expended to move the eyes in a certain direction and the magnitude of the actual eye movement. Past-pointing is caused by a disproportion between motor innervation and its effect. For example, when a nervous impulse is given to move an eye sideways toward an object and, because of paralysis, the eye moves little or not at all, the patient judges the object to be beyond its actual location, and he points past it.

Fifty-nine patients with myopia (nearsightedness) in only one eye were studied. 54/ This condition is commonly associated with amblyopia, an impairment of vision accompanied by no detectable organic disorder. Therapy with glasses and/or patching resulted in improvement of visual acuity and in binocular vision in 25 percent of the cases. The degree of improvement was greatest in the milder cases of amblyopia and myopia, and when therapy was initiated at an early age. The presence or absence of crossed eyes had no influence on the result of therapy. This study shows that early detection and therapy between the ages of 2 and 4 years leads to partial or complete recovery of visual acuity in the affected eye. Until all children have a complete eye examination between 2 and 4 years of age, this condition will usually continue to go unnoticed until the patient has passed the optimal age for therapy. There is an obvious need for visual screening of all pre-school children to detect unilateral refractive errors.

Observations over a 3-year period in a pediatric ophthalmology clinic indicated that optic nerve hypoplasia is more common than previously thought. 55/ This defective or incomplete development of the optic nerve is characterized by marked visual impairment, a reduced response of the pupil to light, a small optic disc, and evidence that the defect has been present since birth. It is usually, although not invariably, associated with crossed eyes, wandering eye movements, or rapid involuntary movement of the eye. Where only one eye is afflicted, the proper diagnosis is extremely important to avoid patching the sound eye, the proper treatment for other forms of strabismus but unwarranted for unilateral optic nerve hypoplasia.

A study on an electrical potential generated in the eye (the electroretinogram or ERG) and in the brain (visually evoked response or VER) indicated that these measurements may be used to determine the refractive condition of the human eye. 56/ A special form of test pattern was used in which the overall light intensity was held constant while individual elements of the pattern fluctuated in luminance. The eye recognizes these fluctuations and responds with large electrical potentials. Small errors of refraction tend to reduce the size of these potentials and large refractive errors cause the potentials to approach zero. The VER is more sensitive than the ERG to small refractive changes, probably because the VER emphasizes responses in the central visual field. This method of refraction is more cumbersome and time-consuming than other techniques, but the electrical responses are more certain to be valid because they depend on the same initial receptive processes as vision itself. This study demonstrates a relationship between detailed focusing of an image on the retina and electrical activity in the brain.

A preliminary experiment tested the effect of heavy particle radiation from thermal neutron capture by boron-10 for the treatment of tumors of the eye. Several factors combine to suggest this therapy in the eye. These include the fact that boron can be introduced directly into the anterior chamber of the eye and the short range and, therefore, the high degree of localization of the radiation. A pentaborate solution was injected into the anterior chambers of rabbits' eyes in which tumors had previously been implanted. Then the area was subjected to a beam of thermal neutrons from

a nuclear reactor. Wherever the boron was placed, thermal neutron irradiation produced high energy ionizing radiation with a discrete localization and a dosage limited only by the boron concentration. Full tumor inhibition was achieved in 14 of 25 eyes, of which 6 survived functionally and anatomically intact. Eight, however, suffered irradiation damage. Of the remaining 11 eyes, all showed some tumor inhibition. Continued studies are in progress. [57]

A technique was developed which supplements x-rays with ultrasonic examination for locating foreign bodies in the eye. [58] The technique takes advantage of the fact that ultrasound can be used to detect non-metallic objects and that one of the ultrasound techniques (B-scan) can offer a two dimensional display, thus providing an orientation of intraocular dimensions. The B-scan technique has the added advantage of simultaneously demonstrating intraocular pathology such as lens dislocation, retinal detachment, blood clots, and anatomic malformations. The technique proceeds from x-ray localization as a first step followed by two ultrasound procedures. The ultrasound and x-ray data can be presented on a single chart in a way that enhances foreign body localization.

REFERENCES

1. Karlin, D. B.
   Arch. Ophthal. 84:409-414, 1970.
2. Enoch, J. M.
   Amer. Jour. Ophthal. 70:995-996, 1970.
3. Berson, E. L.
   Invest. Ophthal. 9:618-628, 1970.
4. Smith, R. S. and E. L. Berson
   Invest. Ophthal. 10:237-246, 1971.
5. Herron, W. L., B. W. Riegel, and M. L. Sears
   Invest. Ophthal. 10:54-63, 1971.
6. Lefler, W. H., J. A. C. Wadsworth, and J. S. Sidbury
   Amer. Jour. Ophthal. 71:224-230, 1971.
7. Goldberg, M. F.
   Amer. Jour. Ophthal. 71:649-665, 1971.
8. Gass, J. D. M.
   Amer. Jour. Ophthal. 71:799-814, 1971.
9. Hyvärinen, L., R. J. Lerer, and D. L. Knox
   Amer. Jour. Ophthal. 71:449-459, 1971.
10. Krill, A. E., D. Archer, and F. W. Newell
    Amer. Jour. Ophthal. 69:826-835, 1970.
11. Hyvärinen, L., A. E. Maumenee, J. Kelley, and S. Cantollino
    Amer. Jour. Ophthal. 71:17-26, 1971.
12. Archer, D., A. E. Krill, and F. W. Newell
    Amer. Jour. Ophthal. 71:266-285, 1971.
13. Cogan, D. G., and S. V. Guzak
    Amer. Jour. Ophthal. 71:291-297, 1971.
14. Waltman, S. R., and H. E. Kaufman
    Amer. Jour. Ophthal. 70:45-47, 1970.
15. McCulley, J. P., H. H. Slansky, D. Pavan-Langston, and C. H. Dohlman
    Arch. Ophthal. 84:516-519, 1970.
16. Kaufman, H. E., E. D. Ellison, and W. M. Townsend
    Arch. Ophthal. 84:783-787, 1970.
17. Centifanto, Y., M. R. M. Goorha, and H. E. Kaufman
    Amer. Jour. Ophthal. 70:1006-1009, 1970.
18. Weissenbacher, J., N. Schachter, M. A. Galin, and S. Baron
    Arch. Ophthal. 84:495-498, 1970.
19. Newmark, E., A. C. Ellison, and H. E. Kaufman
    Amer. Jour. Ophthal. 71:718-722, 1971.
20. Leibowitz, M., and P. Rosenthal
    Arch. Ophthal. 85:163-166, 1971.
21. Burns, R. P., H. Roberts, L. F. Rich
    Amer. Jour. Ophthal. 71:486-498, 1971.
22. Hoefle, F. B., D. M. Maurice, and R. C. Sibley
    Arch. Ophthal. 84:741-744, 1970.
23. Polack, F. M.
    Amer. Jour. Ophthal. 71:505-515, 1971.

24. Forster, R. K., and M. Fine
Arch. Ophthal. 85:42-47, 1971.
25. Armaly, M. F.
Invest. Ophthal. 9:425-429, 1970.
26. LeBlanc, R. E., R. H. Stewart, and B. Becker
Invest. Ophthal. 9:946-948, 1970.
27. Burde, R. M. and B. Becker
JAMA 213:2075-2077, 1970.
28. Krupin, T., S. M. Podos, and B. Becker
Amer. Jour. Ophthal. 70:214-220, 1970.
29. Podos, S. M., T. Krupin, and B. Becker
Invest. Ophthal. 9:492-495, 1970.
30. Marcus, D. F., T. Krupin, S. M. Podos, and B. Becker
Invest. Ophthal. 9:749-752, 1970.
31. Marcus, D. F., T. Krupin, S. M. Podos, and B. Becker
Invest. Ophthal. 9:753-757, 1970.
32. Zweng, H. C., G. L. Paris, A. Vassiliadis, H. Rose, and J. Hayes
Arch. Ophthal. 84:193-199, 1970.
33. Anderson, D. R.
Amer. Jour. Ophthal. 71:470-476, 1971.
34. Bigger, J. F., and B. Becker
Amer. Jour. Ophthal. 71:335-340, 1971.
35. Epstein, D. L., and J. H. Kinoshita
Invest. Ophthal. 9:629-638, 1970.
36. Varma, S. D., B. Chakrapani, and V. M. Reddy
Invest. Ophthal. 9:794-800, 1970.
37. Cotlier, E.
Invest. Ophthal. 9:681-691, 1970.
38. Hatcher, H., and J. S. Andrews
Invest. Ophthal. 9:801-806, 1970.
39. Toyofuku, H., and P. J. Bentley
Invest. Ophthal. 9:959-965, 1970.
40. Peterson, L. H.
Invest. Ophthal. 10:147-150, 1971.
41. Bronson, L. J., and M. Lazar
Invest. Ophthal. 10:144-146, 1971.
42. Ryan, S. J., and G. K. von Noorden
Amer. Jour. Ophthal. 71:626-630, 1971.
43. Murakami, M., and W. L. Pak
Vision Research 10:965-975, 1970.
44. Behrens, M. E., and V. J. Wulff
Vision Research 10:679-689, 1970.
45. Burkhardt, D. A.
Jour. Neurophysiol. 33:405-420, 1970.
46. Michael, J. A., and L. Y. Ichinose
Brain Research 22:249-253, 1970.
47. Hubel, D. H., and T. N. Wiesel
Nature 225:41-42, 1970.

48. Blakemore, C.
Jour. Physiol. 209:155-178, 1970.
49. Partridge, L. D., and J. E. Brown
Vision Research 10:455-460, 1970.
50. Teller, D. Y., and B. Lindsey
Vision Research 10:1045-1055, 1970.
51. Ingling, C. R., H. M. O. Scheibner, and R. M. Boynton
Vision Research 10:501-511, 1970.
52. von Noorden, G. K., and E. M. Helveston
Amer. Jour. Ophthal. 70:199-204, 1970.
53. von Noorden, G. K., S. Awaya, and P. E. Romano
Amer. Jour. Ophthal. 71:27-33, 1971.
54. Rosenthal, A. R., and G. K. von Noorden
Amer. Jour. Ophthal. 71:873-879, 1971.
55. Edwards, W. C., and W. E. Layden
Amer. Jour. Ophthal. 70:950-959, 1970.
56. Millodot, M., and L. A. Riggs
Arch. Ophthal. 84:272-278, 1970.
57. Krohn, D. L., R. Brandt, R. F. Straub, and J. F. Robertson
Amer. Jour. Ophthal. 69:1047-1052, 1970.
58. Coleman, D. J., and S. L. Trokel
Amer. Jour. Ophthal. 71:84-89, 1971.

ADMINISTRATIVE HIGHLIGHTS

Program Development and Analysis

During the year, the Extramural Program of the National Eye Institute prepared an Information and Policy Statement on program project, research center, and outpatient research center grants. The Statement provides guidance to potential applicants for new and renewed support. It also provides guidance to the Institute's advisors for evaluating applications. The effect of this Statement is to distinguish clearly between grants made solely for core research resources and facilities (research center grants) and the grants made for the direct support of specified research (program project and outpatient research center grants). The Statement emphasizes the requirement for a full and adequate documentation of specific research plans in applications for program projects and outpatient centers.

The Institute is developing its procedures for administering cooperative research projects. These large projects which involve many institutions providing data for central analysis require greater involvement of Institute staff than do conventional research projects. In addition to assigning a project officer from its Extramural Program, the Institute will assign a statistician from its Office of Biometry and Epidemiology to monitor the progress and effectiveness of cooperative projects. This administrative arrangement is being implemented with the initiation of a new diabetic retinopathy cooperative study.

At the request of the National Advisory Eye Council, staff has prepared the document "Research Grants Supported by the National Eye Institute." A discussion during the June 1970 meeting of the Council pointed to the desirability of providing brief descriptions of the research projects supported by National Eye Institute grants. The document is a compilation of abbreviated abstracts and represents the Institute's first step in describing and analyzing its research grants program for the Council.

Budget and Finance

Budgetary restrictions imposed by operating under a continuing resolution and by the magnitude of the budget request required funding of research grants at levels below recommended amounts. Throughout most of the year these reductions averaged 10 percent for new, renewal, and continuation research grants. The negotiated reductions allowed the Institute to fund a moderate proportion of its competitive applications. When the appropriation and the apportionment provided additional funds, the Institute made partial restorations so as to average a 6 percent reduction. The Institute was selective in restoring funds; the most meritorious projects received the highest percentage restorations. The Institute also reactivated and awarded several applications previously inactivated because of insufficient funds.

For the second year, the Institute implemented a Council plan for coping with the deficit in funds available for training grants. The plan called

for (a) reduced funding of non-competing grants by an average of 18 percent and (b) reduced funding of competing renewal grants by order of priority score to the extent made possible by the remaining funds. Following this plan, the Institute was able to award five renewal applications for ongoing training programs. Applications for the renewal of four ongoing programs that had been recommended for approval could not be funded. Five approved proposals for new and supplemental support also went unfunded. This continues a trend of decreasing numbers of training programs supported by the Institute. The number of NEI training programs supported by FY 1969, 1970, and 1971 funds was 58, 52, and 47 respectively.

Funds available for postdoctoral and special fellowships allowed the Institute to make 66 awards, an increment over recent years. The Research Career Development Award program remained at approximately a constant level. Nevertheless, replacement of terminating awards during FY 1971 made 10 new awards possible.

Associate Director's Report

Honors and Awards

Dr. Samuel S. Herman who continued as Associate Director for Extramural Programs, NEI, for the first 7 months of the fiscal year, was awarded the Public Health Service Meritorious Service Medal for his sustained high quality work performance in developing the extramural program of the Institute. The award was presented by Dr. Marston on March 3, 1971, in a ceremony at the National Eye Institute.

Dr. A. Edward Maumenee of Johns Hopkins University, a former member of the National Advisory Eye Council, received the first silver membership plaque of Research to Prevent Blindness, Inc., from President Richard M. Nixon. The plaque was presented in a White House ceremony launching a nationwide membership campaign in an intensified scientific effort to halt the rising incidents of blinding eye disease.

Two members of the NEI Vision Research and Training Committee have received Guggenheim Fellowships to study abroad. Dr. Lorrin A. Riggs, Professor of Psychology, Brown University, will spend a year in England at the Physiological Laboratory, Cambridge University. Dr. Abraham Spector, Associate Professor of Ophthalmology, Columbia University, will travel to Paris where he will spend a year at the Institute de Biologie-Physico-Chemique, Foundation Edmund Rothschild.

Program

Although the new National Eye Institute is still taking its first steps toward program analysis and planning, it is already evident that research on rehabilitation and on aids for the blind and the partially sighted is minimal. The Institute's mission encompasses such endeavors, and a few applications in this area are received for almost every Council. As a

rule the applications are relatively unsophisticated; and as a result, the Institute supports very few such projects. Most projects aimed at developing motility and reading aids for the blind and the partially sighted are largely bioengineering efforts, and they are usually very expensive. The costly nature of such research in the face of current budgetary limitations restrains the Institute's inclination to actively promote applications in this area of research.

Through inquiries from investigators and educators in the field, the National Eye Institute has become aware of deficiencies in the support available for certain activities ordinarily considered beyond the mission of the research Institutes of NIH. These inquiries relate to such activities as demonstrating the applicability of research results for clinical practice, curriculum development aimed at reducing the number of years required for specialization in ophthalmology, the development of computerized teaching aids in ophthalmology, and the continuing education of optometrists to improve recognition of ocular pathology. The inability of interested persons to find programs in other components of the Public Health Service or the Department of Health, Education,and Welfare that support such categorical activities suggests that the interests and the responsibilities of the categorical Institutes of NIH would be furthered by an expanded NIH mission concept.

Administration

During the year, Dr. George T. Brooks replaced Dr. Samuel S. Herman as Associate Director for Extramural Programs. There were no organizational changes in the Extramural Program. However, because of pressing needs for developing other segments of the National Eye Institute, the Extramural Program lost three positions. This is a reduction in staff below a level that was already marginal for the sound administration of the program.