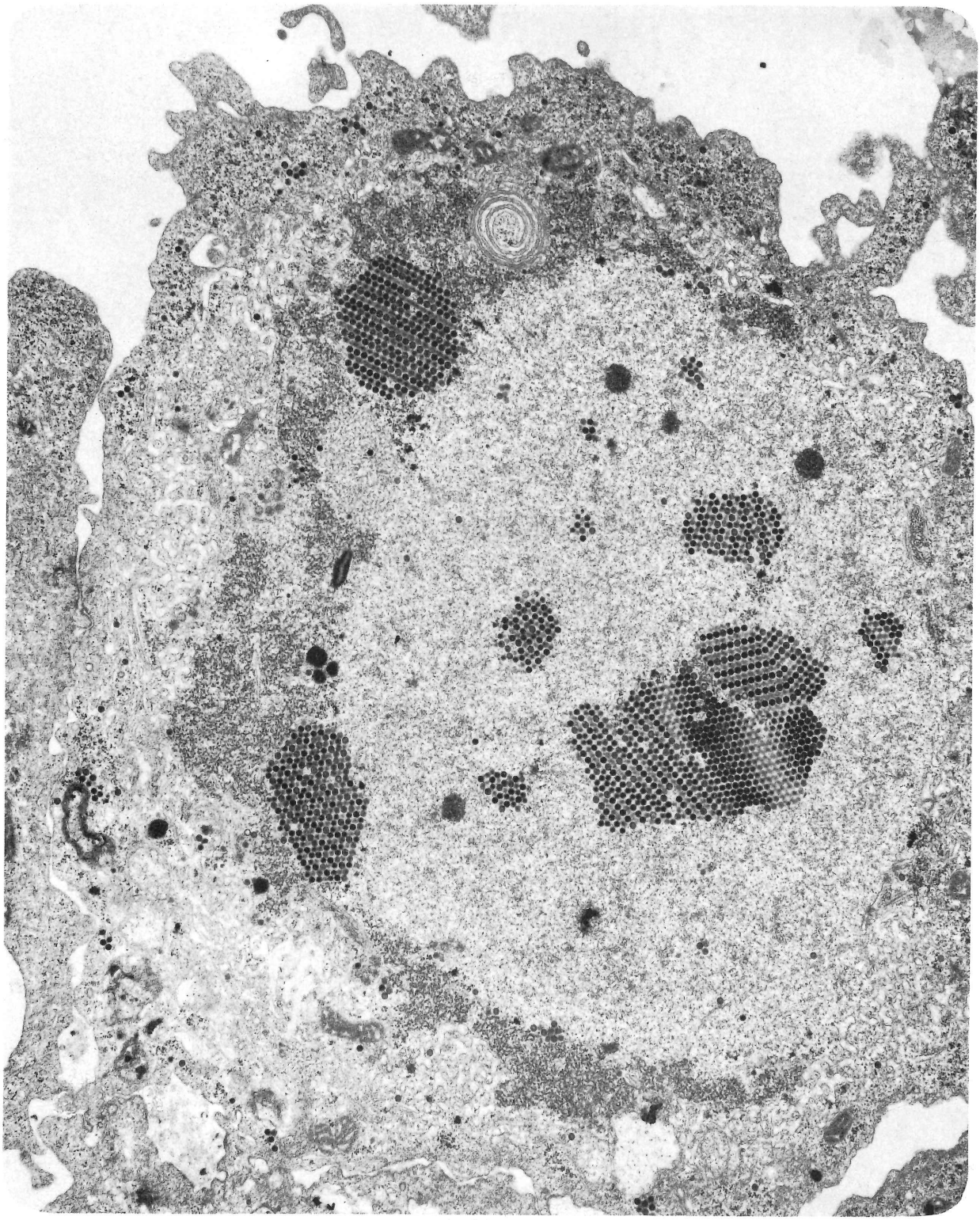
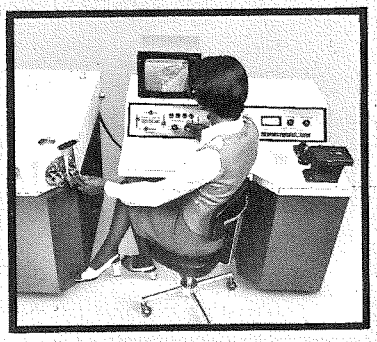
TSEM Texas Society for Electron Microscopy e⁻ NEWSLETTER

Winter 1977



Even a beginner gets expert results



. . . the affordable*, no-nonsense SEM for quality control and material analysis.

The Balscan Scanning Electron Microscope puts the emphasis on simple operation, reliable performance and high quality photography. Many exterior controls have been replaced by internal automation. No previous technical background or extensive training is needed to operate the Balscan SEM. The result is an economically priced SEM geared for quick, uncomplicated performance—ideal for surface studies and direct specimen examination of metallic and non-metallic materials.

And Balscan offers as standard equipment many features which are found on much more expensive SEMs:

Integrated High Resolution Photography System—There is no delay for set up. Changeover to slow scan is fully automatic—just press the photo button and the SEM is switched into slow scan mode. After 30 seconds, the photograph is completed and the SEM is switched back to TV scan image.

Waveform Display in the photographic mode on the visual monitor insures no-miss photographs every time—you get an evaluation of contrast and brightness before you photograph the specimen.

Specific Feature Selector, another Balscan exclusive. Now you can select a point of interest for viewing and photographing at continuously increasing magnification, without mechanically moving the specimen. A lower magnification photograph with the "bright up" selector square can be used as a reference for the area to be photographed at higher magnifications.

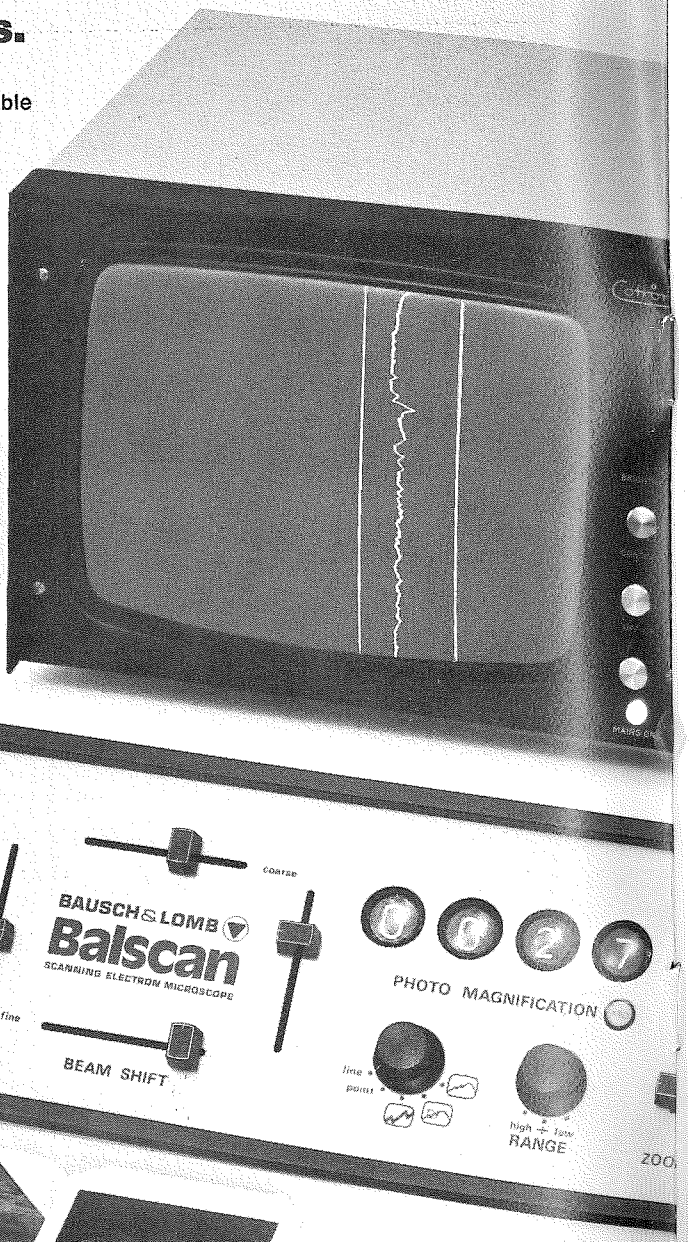
Continuous Zoom Magnification provides almost 50:1 zoom capability allowing you to select just the right degree of magnification for a particular specimen. Visual magnification ranges from 10X-60,000X while photographic magnification is from 5X to 30,000X.

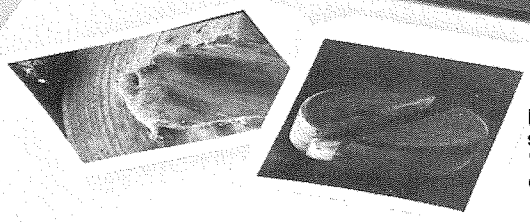
TV-rate Scanning gives you the "no Flicker" green phosphor image which prevents eye strain during long periods of viewing. The big 11" green phosphor display allows several people to view the specimen simultaneously under normal lighting conditions. You can also make video tape recordings and hook up additional TV display units in any area.

Variable Accelerating Voltage from 1kv to 25kv in 1kv steps. You get greater flexibility in examining specimens without coating. With a 1kv step capability 400 Angstroms is all the resolution you need for most applications.

Get the full story of the Bausch & Lomb Balscan SEM. Write for a free demonstration and Catalog No. 42-2416.

*Base Price with photographic unit $23,961.
The Balscan SEM is manufactured by Cambridge Scanning Company, Ltd., Cambridge, England and is distributed and serviced worldwide by Bausch & Lomb, Inc.





Balscan's waveform display shown above gives you "no-miss" photographs every time.

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Write to Bausch & Lomb, Scientific Optical Products Division, DEPT. 6602, 1400 North Goodman St., Rochester, N.Y. 14602.

Contents

Volume 9, Number 1
Winter, 1977

Texas Society for Electron Microscopy
"For the purpose of dissemination of research with the electron microscope"

ON THE COVER

Crystalline arrays of ad enovirus type 19 shown at 20,000x from the Section of Virology, Department of Microbiology, Scott & White Clinic, Temple, Texas.

The new generation X-ray analysis system from TracorNorthern.

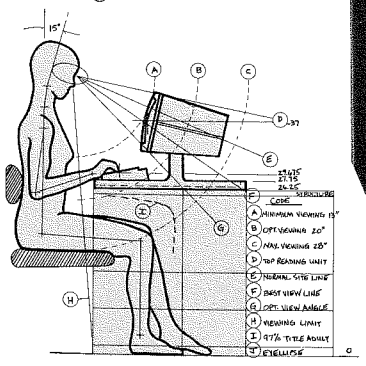
TN-2000

At Tracor Northern we've established a reputation for leadership in developing innovative analytical instrumentation.

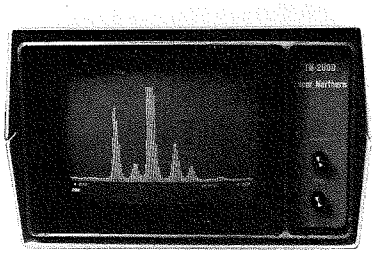
Five years ago we introduced the NS-880, the first truly computer-based X-ray elemental analysis system. The NS-880 has gained world-wide acceptance and is today the premier quantitative analysis system.

Tracor Northern now proudly introduces System TN-2000, a new microcomputer-based system with the exceptional performance you've come to expect from our instruments. System TN-2000 offers human-engineered convenience and simplified operation at a modest price, while maintaining our tradition of uncompromising quality and unmatched versatility.

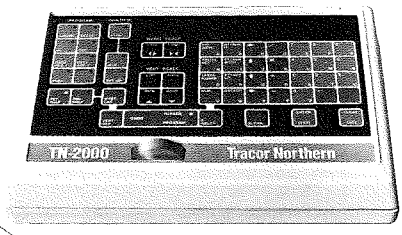
System TN-2000 is totally human-engineered for convenience and ease of operation. The modular backlighted control keypad and large-screen high-resolution display can be individually placed to optimally suit your working environment.



System TN-2000's superior display is controlled by its own microprocessor to provide comprehensive visual information with unparalleled clarity and versatility. Labeled keypad functions and unique OMNI control provide instant interaction with all display features.



A uniquely new, elegantly simplified approach to operation setup and control allows you to perform complete analyses with unprecedented ease. You simply select desired operations from displayed choices, with current selections identified by complete and lucid text.



Efficient operation is ensured by permitting operating parameters to be individually modified.

System TN-2000 is more than an innovative instrument. It is a state-of-the-art computer-based system whose capabilities can grow with your needs.

If you are interested in rapid and reliable X-ray elemental analysis, you must see System TN-2000 in action. Contact us now.

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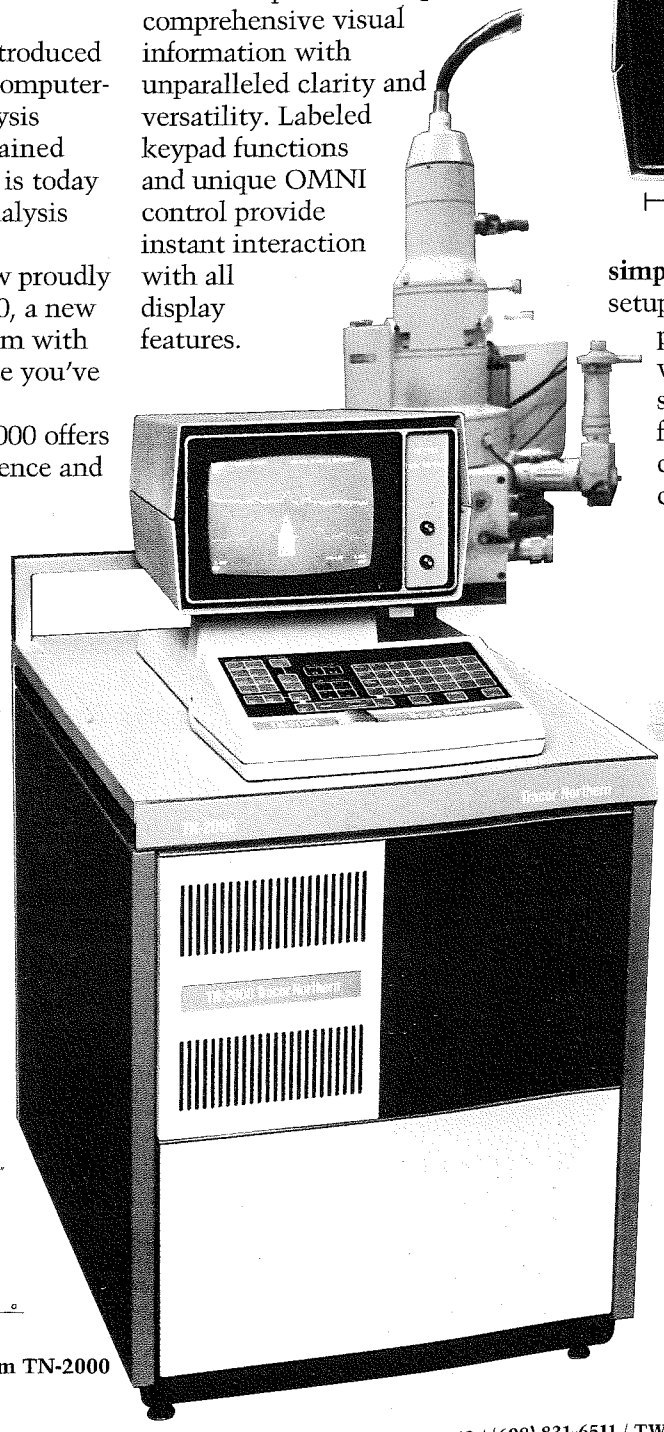

System TN-2000

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Tracor Europa B.V. / Schiphol Airport Amsterdam / Building 106, The Netherlands / Telephone (020) 41 1865 / Telex 13695

President's Message

I look forward to February 9-11, 1978 and the Seventh Annual LSEM-TSEM Joint Symposium and take this opportunity to welcome our visitors from Louisiana. We hope you enjoy the meetings and your stay in San Antonio as much as we enjoyed your endeavors in New Orleans last year. I think we all agree that our decision seven years ago to initiate these Joint Symposia was a wise one.

The spring meeting of TSEM will be held May 4-6, 1978 at the Southpark Inn, Lubbock, Texas. Randy Brackeen, Department of Anatomy, Texas Tech University School of Medicine is the local arrangements chairman for this meeting and if you have questions, call him at (806) 743-2705. Incidently, we are investigating the possibility of group rates on airlines and will have an announcement about this at the business meeting. Suffice it to say here that there is an airline that will transport you roundtrip to Lubbock from Dallas for $30 and from Houston for $50.

I think it noteworthy to mention that Ann Goldstein has been appointed Associate Newsletter Editor and will become our Editor in the fall of 1978. Also, Leon McGraw has volunteered to be the TSEM Historian and will be lurking in the shadows with a new camera to record our historic events. I suggest you be good or at least beware.

JERRY D. BERLIN
President

TSEM FINANCIAL REPORT
Period Ending December 1, 1977

Total Assets of August 1, 1977		$ 7,510.84
Certificate of Deposit (University Bank #4470)		1,252.22
Certificate of Deposit (Fannin Bank #17864)		1,000.00
Savings Account (University National Bank		4,000.00
Balance in Checking Account August 1, 1977		1,258.62
RECEIPTS		
Corporate dues	150.00	
Regular dues	245.00	
Student dues	21.50	
Corporate donations (Arlington Mtg)		
Registration Receipts (Arlington Mtg)	1,591.00	
Savings account transfer (11/29/77)	500.00	
Interest on C Dep. 17864	18.27	
Total Income	$ 2,525.77	$ 2,525.77
		$ 3,784.39
DISBURSEMENTS		
Travel (President TSEM to EMSA-Boston)	196.00	
Scott and White — Secretarial fund	1,000.00	
Treasurers Expenses (Xerox & Mailing)	156.29	
Meeting expenses (Arlington)	1,392.27	
Student travel (Arlington)	100.00	
Preliminary expenses TSEM/LSEM Meeting	300.00	
Workshop refunds (Student over charges)	57.00	
American Indemnity Co. (Treasurer's Bond)	28.00	
Camera — Society's historian	163.75	
	3,393.31	
		3,393.31
Balance in checking account 12/1/77		391.08
SAVINGS ACCOUNTS		
Certificate of Deposit (University Bank #4470)		$ 1,271.14
Certificate of Deposit (Fannin Bank #17864)		1,000.00
Savings Account (University National Bank #01-7420-3)		3,500.00
TOTAL ASSETS December 1, 1977		**$ 6,162.22**

Zeiss offers a full line of

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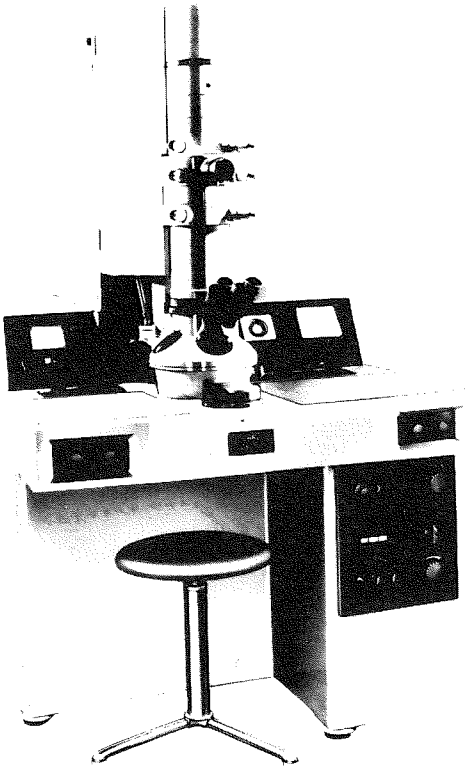
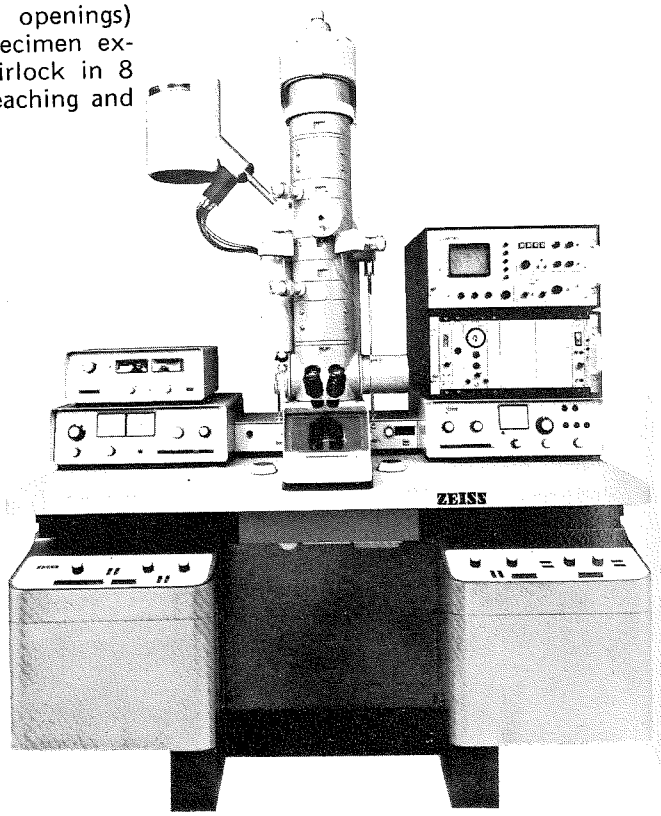


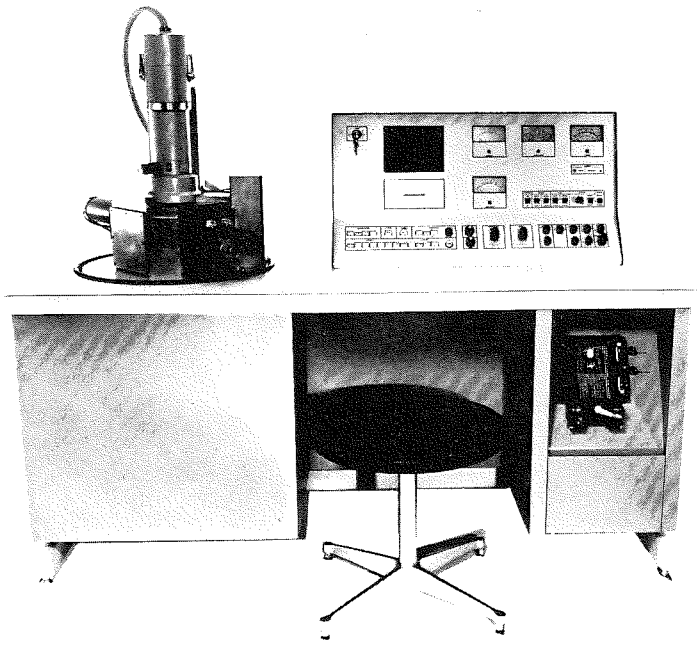
EM 9 S-2 7 Å p.t.p.
Transmission Electron Microscope

Fully automatic camera system includes identification of negatives. Extremely simple to operate. Small size. Big performance. Low price. Zoom or fixed-step magnifications from 30x to 60,000 — distortion-free with direct read-out. Focusing aid. Multiple (21 openings) thin-metal film aperture. Specimen exchange through foolproof airlock in 8 secs; stereo tilt. Ideal for teaching and heavy daily use.



EM 10 3.0 Å p.t.p.
High-Resolution Transmission Electron Microscope 1.4 Å lattice

Now two models: EM10A, 100x—200,000x; EM10B, 100x—500,000x. Easy, foolproof operation. Full X-ray protection. High-resolution goniometer, multiple specimen holder, cartridges for cooling, heating, tensile testing. Fully automatic 3¼ x 4", 70mm, and 35mm camera systems, with unique 5-character automatic data imprint. 45° take-off angle for X-ray element analyzer of your choice. Super-clean vacuum with ion-getter pump system.



NOVASCAN 100 Å (edge)
New Scanning Electron Microscope

7x — 150,000x magnification at all scan speeds. 15 and 30 kV, and 1 — 5 kV continuously variable. Large specimen chamber with 6 ports and unique top-plate lift for easy access to 5-axis goniometer stage and correct positioning of specimen. Push-button controlled systems for easy and efficient operation. Polaroid camera is standard, other cameras optional. Full range of accessories.

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The Zeiss EM-10 High-Resolution TEM: modify it in less than a minute.

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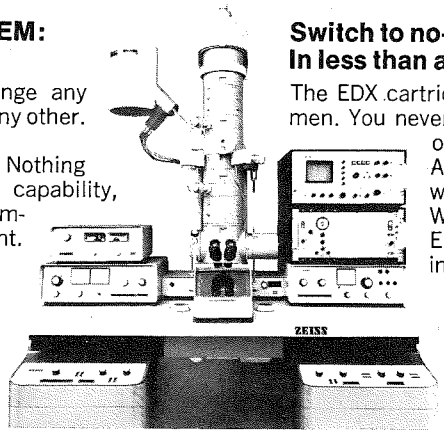
Even the Goniometer can be inserted in less than a minute. Without affecting the focal length, magnification, or resolution. Without restricting the microscope in any way for any other use.



Switch to no-tilt X-Ray Analysis. In less than a minute.

The EDX cartridge goes straight in, without tilting the specimen. You never lose sight of it. That's because any analyzer of your choice can be mounted at a 45° angle. And, like the Goniometer, it can be added without exchanging specimen stages or lenses. Without re-alignment. Without turning your EM-10 into an exclusively special-purpose instrument.

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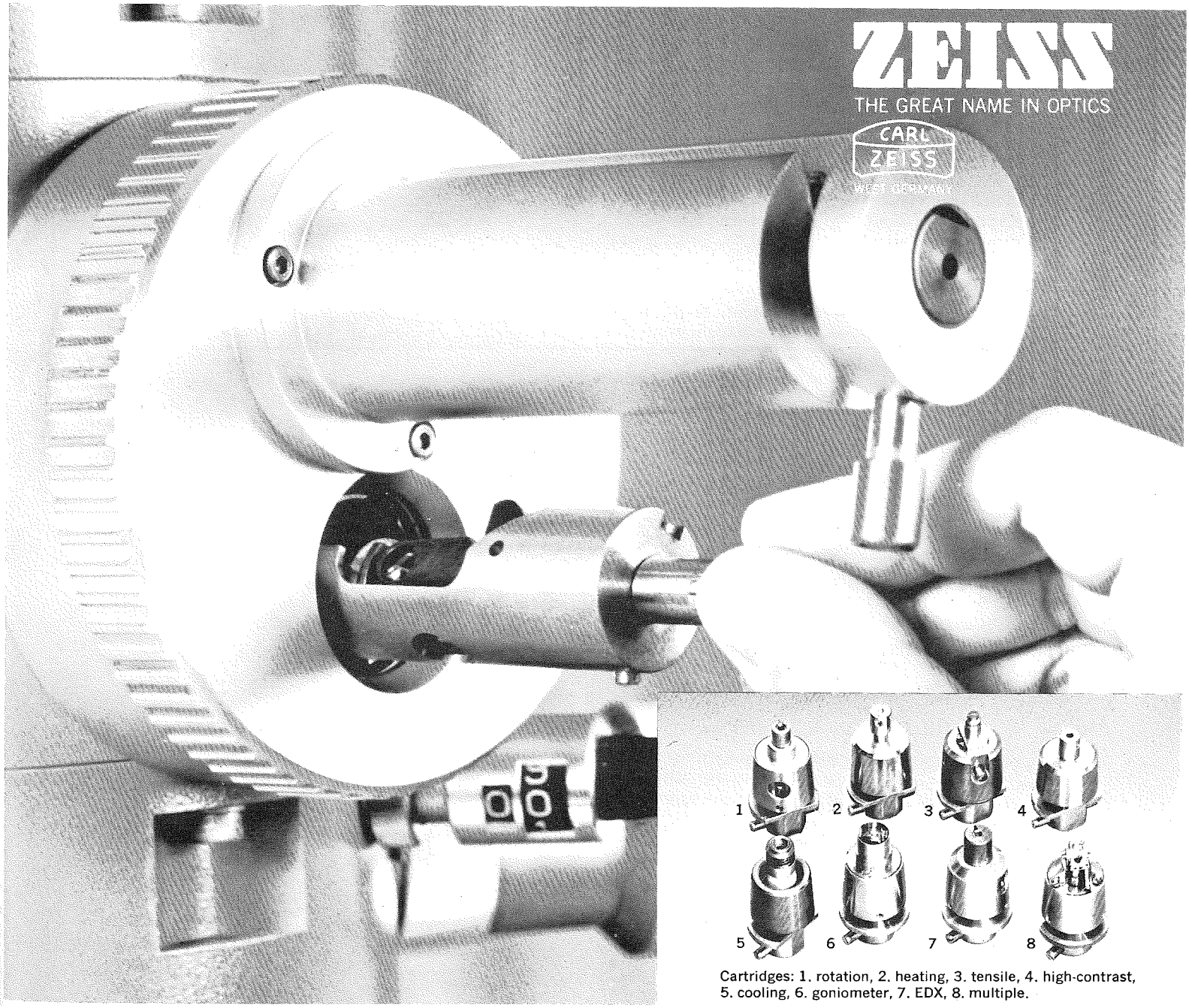

Cartridges: 1. rotation, 2. heating, 3. tensile, 4. high-contrast, 5. cooling, 6. goniometer, 7. EDX, 8. multiple.

Welcome LSEM

TSEM takes great pleasure in hosting the 7th Annual TSEM-LSEM Symposium in San Antonio, Texas.

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A word of special thanks and recognition to these members of the Local Arrangements Committee for a job well done for this highly successful TSEM-LSEM Symposium.

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DR. DAMON HERBERT ..... Socials
DR. FRANK WEAKER ..... Socials
DR. CURT GRAVIS ..... Program
MR. CON SMITH ..... Security and Ladies Hospitality

TENTATIVE PROGRAM
1978
TSEM-LSEM SYMPOSIUM

THURSDAY, FEBRUARY 9

12:00-5:00 p.m. Registration, Lobby
12:00-5:00 p.m. Ladies Hospitality, Room 350
1:00-3:00 p.m. Executive Committee Meeting, Room 361
7:30-10:00 p.m. Social, Anacacho Room

FRIDAY, FEBRUARY 10

All Day Ladies Hospitality, Room 350
All Day Exhibits, Peraux Room and Lobby
8:30-11:45 a.m. Session I, Anacacho Room
9:30-11:45 a.m. Session II, Navarro Room
7:00-11:30 p.m. Friday Night Mexican Buffet and Supper
6:30-6:45 —
11:30-11:45 p.m. Transportation Schedule, bus or barge depending on weather.

SATURDAY, FEBRUARY 11

8:00-Noon Ladies Hospitality, Room 350
8:00-Noon Exhibits, Peraux Room and Lobby
8:15-11:45 a.m. Session V, Cavalier Room
Session VI, Navarro Room

1:30-5:45 p.m. Session III — Anacacho Room
1:30 p.m. Invited Speaker — Dr. Jean-Paul Revel
2:30-5:45 p.m. Session IV — Navarro Room

SESSION I

8:30 **Invited Speaker — Dr. Myron C. Ledbetter, The Use of Anaglyphs in Stereo Electron Microscopy,** Biology Department, Brookhaven National Laboratory, Upton, N.Y. 11973.

9:30 **Non-Random Cell Death During Spermatogensis in the Squash Bug, Anasa tristis.** James K. Butler, Department of Biology, The University of Texas at Arlington, Arlington, Texas 76019.

9:45 **Ribosome-Like Particles Are Associated With the Acrosomal Membrane of Guinea Pig Spermatids.** Hilton H. Mollenhauer and D. James Morre, Veterinary Toxicology and Entomology Research Laboratory, ARS, USDA, P.O. Drawer GE, College Station, Texas.

10:00 **A Stereological Analysis of Early and Later Passage Embryonic Syrian Hamster Fibroblasts.** J. J. Brokaw[1], J. K. Butler[1], and J. R. Williams[2], Department of Biology[1], The University of Texas at Arlington, Arlington, Texas; and Department of Physiology[2], The Harvard University School of Public Health, Boston, Mass.

10:15 **An Ultrastructural Study of the Ventral Nerve Cord of Cylmenella Torquata.** Claire E. Hulsebosch, Department of Zoology, University of Texas at Austin, Austin, Texas 78712.

10:30 **BREAK**

10:45 **Types And Relative Numbers of Specialized Contacts Between Cell Processes in the Spinal Substantia Gelatinosa of the Cat.** Donald Duncan and Ricardo Morales, Department of Anatomy, The University of Texas Medical Branch at Galveston.

11:00 **Nerve-Mast Cell Relationships in the Armadillo Dermis.** Robert F. Dyer, Department of Anatomy, L.S.U. Medical Center, New Orleans, Louisiana.

11:15 **Ultrastructure of the Caudal Integument (Resting Stage) in the Juvenile Skink, Eumeces Faciatus.** M. Lynn Davis, Department of Biology, Stephen F. Austin State University, Nacogdoches, Texas 75962.

11:30 **Isolation of Chloride Cells from the Branchial Epithelium of a Euryhaline Teleost.** Seth R. Hootman, Natalia Raphael and Charles W. Philpott, Department of Biology, Rice University, Houston, Texas 77001.

11:45 **An Electron Microscopic Study of the Baroreceptor Nerve Endings in the Internal Carotid Artery of the Rat.** R. D. Yates and I-li Chen, Department of Anatomy, Tulane Medical School.

LUNCH

SESSION II

9:30 **Stereological Analysis of Mitochondrial Development in Germinating Phaseolus Vulgaris L. Axis.** Betty Hamilton and Glenn Todd, School of Biological Sciences, Oklahoma State University, Stillwater, Oklahoma 74074.

9:45 **Isolation and Characterization of Myxococcus Disciformis, A Peat Bog-Associated Myxobacter.** Leonard A. Hook, John M. Larkin, Department of Microbiology, L.S.U., Baton Rouge, LA, and Ellis R. Brockman, Department of Biology, Central Michigan University, Mt. Pleasant, MI.

10:00 **A Scanning Electron Microscope Study of the Number of Fibers on Cotton Seeds.** Jerry D. Berlin, Department of Biological Sciences, Texas Tech University, Lubbock, Texas 79409.

10:15 **Morphological and Leaching Characteristics of Thermophilic Microorganisms Isolated from an Experimental Ore Body.** V. K. Berry, Department of Anatomy, The University of Texas Health Science Center, San Antonio, Texas 78284.

10:30 **BREAK**

10:45 **Morphologic and Radioautographic Studies of the Digestive Gland Cells of Drosera Capensis and the Role of the Golgi Apparatus in Slime Production.** Robert L. Outenreath, The Cell Research Unstitute, The University of Texas at Austin, Austin, Texas 78712.

11:00 **Chemical Characterization of Particulates From Coal Gasification and Relevance to Inhalation Toxicology.** D. L. Davidson and E. M. Gause, Southwest Research Center, San Antonio, Texas.

11:15 **Specimen Disc-Perma Stub System for Scanning Electron Microscopy.** Michael T. Postek, Department of Botany, L.S.U., Baton Rouge, Louisiana 70803, and

William A. Ladd. Ladd Research Industries, Inc., P.O. Box 901, Burlington, Vermont 05402.

11:30 **Reinforcement of Image Detail by Integrating Two Differently Colored Micrographs.** Mel Watson, Department of Biology, Texas A&M University, College Station, Texas 77840.

11:45 **Inadequacies of Conventional Methods of Floating Thin Section Herding and A Solution.** Alan B. Weckerling, Weckerling's World, 6702 Cypress Lake, San Antonio, Texas 78244.

LUNCH

SESSION III

2:30 **Electron Microscopic Study of Anaplastic Thyroid Carcinoma.** James R. Newland and Bruce Mackay, Department of Pathology, M. D. Anderson Hospital and Tumor Institute.

2:45 **HLA Typing of Melanoma and Colon Tumor Cell Lines and Their Corresponding Ultrastructural Features.** Cameron E. McCoy, Nancy D. Mabry, Wm. B. McCombs and A. Leibovitz, Scott & White Clinic, Temple, Texas 76501.

3:00 **Fine Structure of R-3372 Transplantable Prostate Tumor of Fischer-Copenhagen Rats.** G. Seman, J. M. Bowen, and L. Dmochowski, Department of Molecular Carcinogenesis and Virology, The University of Texas System Cancer Center, M. D. Anderson Hospital and Tumor Institute, Houston, Texas 77030.

3:15 **Acute Effects of Morphine on Mouse Liver and Adrenal Medulla.** Asa Thureson-Klein, Jack McC. Baggett, Jen Wang-Yang and Mary Lou Percy, Department of Pharmacology and Toxicology, University Mississippi Medical Center, Jackson, Mississippi 39216.

3:30 **Ultrastructure of Extraadrenal Chromaffin Cells Following Insulin Injection.** Joe A. Mascorro, John W. Armstead[1] and Robert D. Yates, Department of Anatomy, Tulane Medical School, New Orleans, Louisiana, and Department of Biology, Wabash College, Crawfords.ville, Indiana.[1]

3:45 BREAK

4:00 **The Role of Electron Microscopy in the Diagnosis of Reticulo-Endothelial Disorders.** Bruce Mackay, Barbara M. Osborne, Richard J. Ford and James J. Butler, Department of Pathology, M. D. Anderson Hospital and Tumor Institute.

4:15 **Diagnostic Virology Capabilities Expanded by the Use of Electron Microscopy at a Major Medical Center.** Steven E. Alexander, Dermott Acton, George Kasai, Virology Section; and Alan B. Weckerling, Lucy Olalde, Electron Microscopy Laboratory, Department of Biology and Area Lab Services, Brooke Army Medical Center, Fort Sam Houston, Texas 78234.

4:30 **Adenovirus 19 Epidemic Keratoconjunctivitis.** Albert Leibovitz, Cameron E. McCoy and Peter G. Amaral, Scott and White Clinic, Temple, Texas 76501.

4:45 **Comparison of Mason-Pfizer Monkey Virus and Squirrel Monkey (Saimiri Sciureus) Retrovirus, Immunoelectron Microscopy.** G. C. Smith, R. L. Heberling, and S.!S. Kalter, Microbiology and Infectious Diseases, Southwest Foundation for Research and Education, San Antonio, Texas.

5:00 **Ultrastructure of the Host-Pathogen Relationship in the Dewberry Rust Disease.** Dale G. Glidewell and Charles W. Mims, Department of Biology, Stephen F. Austin State University, Nacogdoches, Texas 75962.

5:15 **Bacterial Distribution and Histological Changes of Rat Gingiva During Total Intravenous Feeding.** Allan G. Kaster and Ivan L. Cameron, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas 78284.

5:30 **Morphology of Type A and Type B Particles After Fixation In Picric Acid-Paraformaldehyde.** J. Phillips and G. Seman, The University of Texas System Cancer Center, M. D. Anderson Hospital and Tumor Institute, Houston, Texas 77030.

SESSION IV

2:30 **Molecular and Ultrastructural Events Associated with Plasma Membrane Turnover in the Avian Salt Gland.** Michael P. Sarras, Jr. and Fred E. Hossler, Department of Anatomy, L.S.U. Medical Center, New Orleans, LA 70119.

2:45 **The Effect of Inorganic Cations on the Membranes of Tetrahymena.** Sharon M. Mattox and Guy A. Thompson, Jr., Department of Botany, The University of Texas, Austin, Texas.

3:00 **Ultrastructure of the Cell Envelope and Sulfur Inclusions of Beggiatoa.** W. R. Strohl, J. M. Larkin, S. Seuferer and K. S. Howard, Department of Microbiology, L.S.U., Baton Rouge, LA.

3:15 **Lectin Induced Redistribution of Cell Surface Anionic Sites in Hepatoma Ascites Cells.** Peter C. Moller and Jeffrey P. Chang, Division of Cell Biology, Department of Human Biological Chemistry & Genetics, The University of Texas Medical Branch, Galveston, Texas 77550.

3:30 **Surface Features of AS30D In Vitro.** K. E. Savage and P. S. Baur, Department of Human Biological Chemistry, Genetics and Cell Biology, Galveston, Texas.

3:45 BREAK

4:00 **Morphogenesis of Membrane Ribosomal Lamellae in Hairy Cell Leukemia.** Milka M. Montiel, M.D., and Barbara Boehm, The Department of Pathology, The University of Texas Health Science Center, San Antonio, Texas.

4:15 **Scanning Electron Microscopic Observations on the Mechanism of Somatic Cell Fusion Using Polyethylene Glycol.** Mike A. Clark and Jerry W. Shay, The University of Texas Health Science Center, Dallas, Texas.

4:30 **Replicating Chromatin: Electron Microscopic Visualazation.** G. B. Howze, Department of Biology, Texas Southern University, Houston, Texas 77004.

4:45 **Morphologic & Histologic Features of Infectious Bursal Disease — An Immuno Suppresive Disease of Commercial Chickens.** Danny Millar, Texas A&M University, Department of Veterinary Microbiology.

5:00 **Bacterial Endosy-Biosis in the Caecum of the Southern Green Stink Bug (Nezara viridula L.).** Robert P. West and A. D. Larson, Department of Entomology and Microbiology, L.S.U., Baton Rouge, Louisiana 70803.

5:15 **A Scanning Electron Microscopic Study of Periphytic Microorganisms.** Thomas M. Dreier, Electron Microscopy Center, Department of Biology, Texas A&M University, College Station, Texas 77843.

SESSION V

8:30 **Studies on the Eyes of Bigeyes (Teleostei: Priacanthidae) With Special Reference to the Tapetum Lucidum.** R. T. Wang, J. A. C. Nicol, E. L. Thurston, and M. McCants, Department of Zoology, The University of Texas at Austin 78712 and Department of Biology, Electron Microscopy Center, Texas A&M University, College Station, Texas 77843.

8:45 **Surface Morphology of the Gill Filaments of the Mullet, Mugil Cehpalus.** Fred E. Hossler and John R. Ruby, Department of Anatomy, L.S.U., New Orleans, LA., and Thomas McIlwain, Gulf Coast Research Lab., Ocean Springs, Mississippi.

9:00 **Brain Stem Lesions in the Spontaneously Hypertensive Rat.** P. M. Klara, H. Kawamura, C. Knox and E. Frohlich, Tulane University, Department of Anatomy and the Ochsner Foundation.

9:15 **Permeability Changes in Arterioles, Venules and Capillaries Following Experimental Cerebral Angiography.** R. L. Casady, G. T. Kitten and P. R. Sterrett, Department of Anatomy, Texas Tech University School of Medicine, Lubbock, Texas 79409.

9:30 **Adenosine Triphosphatase and Alkaline Phosphate Localization in the Cerebral Vasculature of Spontaneously Hypertensive Rats. 1C. A. Knox[1]**, P. M. Klara[1], R. D. Yates[1], and E. Frohlich[2], Department of Anatomy, Tulane Medical School[1], and the Ochsner Foundation Hospital[2], New Orleans, Louisiana.

9:45 **Ultrastructural Changes in Myocardium of Dogs Following Suprathreshold Transchest Defibrillating Electric Shock.** Joyce S. Davis, M.D.; Hilton H. Mollenhauer, Ph.D., Texas A&M University, College of Medicine; J. T. Lie, M.D., Mayo Medical School; W. A. Tacker, M.D., Ph.D., Purdue University.

10:00 **BREAK**

10:15 **Ultrastructural Studies of Cardiac Mesenchyme Cultured on Collagen Substrates.** D.H. Bernanke, R.R. Markwald, J.M. Krooe, Department of Anatomy, Texas Tech University School of Medicine, Lubbock, Texas 79409.

10:30 **Cell: Matrix Interaction During Cushion Tissue Cell (CTC) In Vivo Migration.** R. R. Markwald, J. M. Krook, Department of Anatomy, Texas Tech University, School of Medicine, Lubbock, Texas 79409.

10:45 **A Comparison of Intracellular Elements in Normal and Tumor Cells as Dtermined by X-Ray Microanalysis.** Thomas B. Pool, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas.

11:00 **The ytology of Canine Pacreatic Acinar Cells Following Hypophysectomy.** A. H. Crenshaw, Jr., and W. A. Shannon, Jr., Veterans Administration Hospital and Department of Cell Biology, Southwestern Medical School, Dallas, Texas.

11:15 **Pacreatic Duct Ligation and Resulting Fine Structural Alterations in the Canine Pancreas.** A. H. Crenshaw, Jr., W J. Brown, G. Patton, R. E. Dobbs, and W. A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Physiology, Southwestern Medical School, Dallas, Texas.

11:30 **Abnormal Structures in Lymphocytes from Patients With Oculocraniosomatic Neuromuscular Disease.** W. J. Brown, J. D. Cook, and W. A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Neurology, Southwestern Medical School, Dallas, Texas.

11:45 **Mitochondriopathy in Oculocraniosomatic Neuromusuclar Disease.** W.J. Brown, J.D. Cook., and W.A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Neurology, Southwestern Medical School, Dallas, Texas.

SESSION VI

8:30 **Morphological and Ultrastructural Changes in Glandless Cottonseed (Gossypium Hirusutum L.) Induced In A Non-Extruded Texturized Vegetable Protein.** M. V. Taranto and K. C.Rhee, Food Protein R&D Center, Texas A&M University, College Station, Texas.

8:45 **An Anatomical Analysis of the Epidermal Surafec Elaborations of Carya Illinoensis.** Larry Grauke, Department of Horticulture, Texas A&M University.

9:00 **Ferromangnaese Microaccretions from the Mid Atlantic Ridge Famous Area.** L. A. Barnard, Department of Oceanography, Texas A&M University, College Station, Texas.

9:15 **SEM Used To Evaluate Formation Damage.** D. F. Feuerbacher and M. H. Cordova, Dresser Industries, Inc.

9:30 **Evaulation of the Thin Film Assumption in Quantitative X-Ray Analysis.** D. P. Skinner and N. C. Barbi, Princeton Gamma-Tech, Inc.

9:45 **Ultrastructural Observations on the Motile Cells of the Green Epiphytic Alga Phycopeltis Epiphyton (Chroolepidaceae.)** B.H. Good and R. L. Chapman, Department of Botany, L.S.U., Baton Rouge, Louisiana.

10:00 **BREAK**

10:15 **Ultrastructure of Sensory Sensilla on the Antennae of Some Parasites in the Chalcidoidea (Insecta: Hymenoptera).** Margaret R. Barlin, Department of Entomolgy, Texas A&M University.

POSTER SESSIONS

Intracellular Distribution of Elements in Mouse Pancreatic Acinar Cells by X-Ray Microanalysis. T. B. Pool, I. L. Cameron and Nancy Smith, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas.

Ultrastructure and Cytochemistry of Oogonial Development in Achlya Recurva. Ellzey, Joanne T., Elain Huizar and Michelle Baca, Ultrastructure Laboratory Biological Sciences, The University of Texas at El Paso.

Differences in Concentration of Elements in Brain Cells Due To: Opiates, Cell Type and Subcellular Location. An XpRay Microanalysis Study. Ivan Cameron, Peter Sheridan and Nancy Smith, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas.

Electron Microscopy, An Art Form. Ruth E. Lewis, Electron Microscopy Center, Texas A&M University, College Station, Texas 77843.

Abstracts received after the deadline of December 9, 1977 do not appear in this tentative program, but will appear in the final program distributed at the meeting on February 9-11, 1978.

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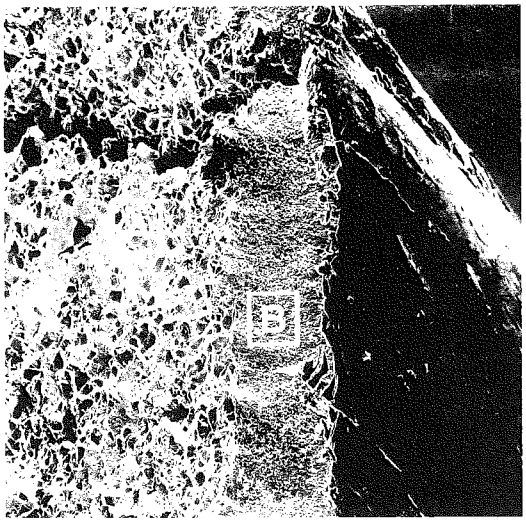
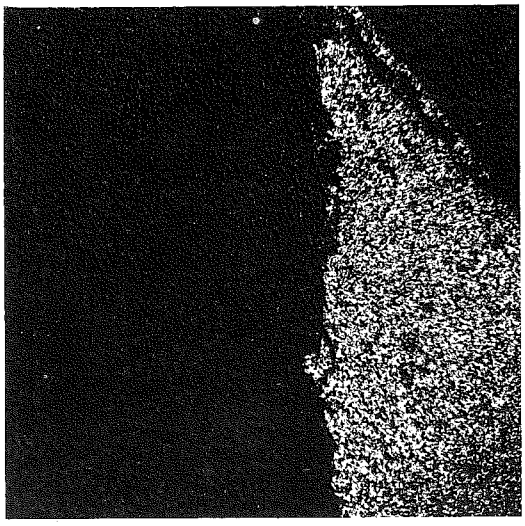
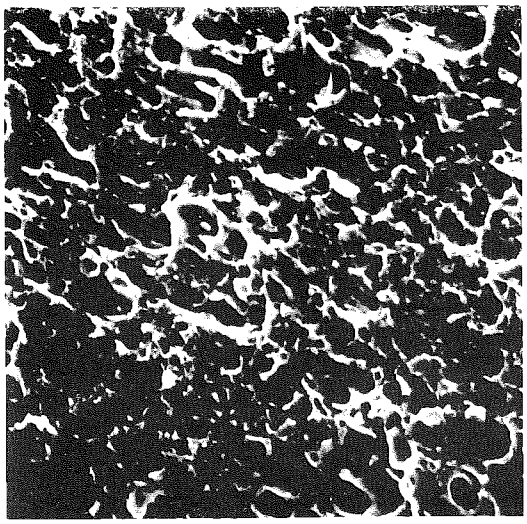


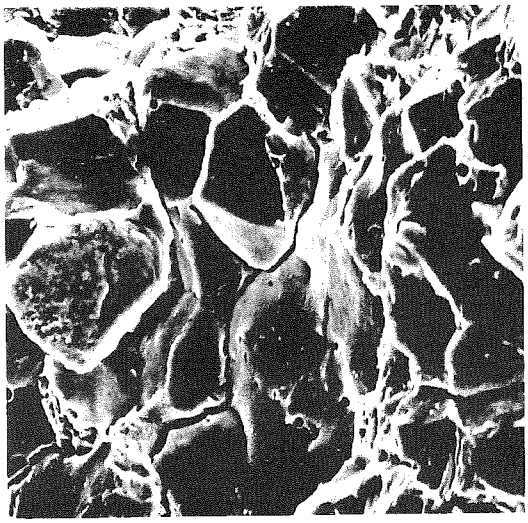
Fracture topography of plated steel component — area A failed in brittle fracture while area B failed in a ductile manner. 100X



Zinc x-ray distribution map of area corresponding to plating. 100X



Closeup of area B showing dimple rupture of plating. Note spherical second phase particles located within some dimples. 2000X



Closeup of area A shows intergranular cracking due to hydrogen embrittlement caused by improper bakeout after plating. 1600X

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High-Voltage Electron Microscopy In Biomedical Research

Jerry W. Shay
Department of Cell Biology, The University of Texas Health Science Center at Dallas. 5323 Harry Hines Boulevard, Dallas, Texas 75235, (214) 688-2623.

A. Introduction

Even though the standard transmission electron microscope, usually operated in the 40-100 KV range, is capable of resolving less than 10 angstroms (1 nanometer) and has a wide range of applications in both the biological and material sciences, there are certain areas of research where electron microscopes in the megavolt (1,000 KV) range might be desirable. In the mid-1940s when the standard transmission electron microscope was become generally available for biomedical research, a technical problem developed in that most biological specimens were too thick to be penetrated by the relatively low accelerating voltages available. In order to solve this technical problem, two possibilities existed: (1) make the specimens thinner, or (2) increase the accelerating voltage in order to penetrate thicker specimens. Basically, with the invention of the ultramicrotome by Keith Porter and Joseph Blum, attention was temporarily diverted from development of high-voltage electron microscopy as thinner sections were easily penetrated by the 40-100 KV standard transmission electron microscopes. During the last decade, however, microscopes containing accelerating voltages as high as 3 megavolts have been built in France, Great Britain, Japan, and more recently the United Staes. These high-voltage electron microscopes were originally used primarily as metallurgical instruments, but in recent years several microscopes have been built in order to determine if high-voltage electron microscopy can be applied to biological research.

B. Advantages of High-Voltage Electron Microscopes

Almost every article written about high-voltage electron microscopy lists three important properties of high-voltage operation (1, 2, 3). These are: (1) increased specimen penetration which allows the investigator to obtain photographs of relatively thick sections (0.5 - 1.0 μm or more); (2) reduced specimen damage under beam exposure; and (3) improved resolution due to a decrease in the spherical and chromatic aberrations of the lens system. Other advantages frequently mentioned include more efficient dark-field operation and more vesatility in that with a larger column, there is more space for special attachments such as experimental devices for examining living cells.

FIGURE 1 — *JEM 1000 high-voltage electron microscope facility at the University of Colorado in Boulder. (Photograph by Mr. Dick Carter.)*

C. High-Voltage Electron Microscopy National Research Resource Centers.

In addition to being complicated and costly (approximately one dollar per volt), high-voltage electron microscopes are several stories high and weigh tens of tons (Fig. 1). Therefore, experimental activities have been carried out at only a few existing installations. About two dozen high-voltage electron microscopes exist throughout the world with three installations available for biological researchers in the United States. These three 1 MeV (1,000 KV) electron microscopes are located at the U.S. Steel Research Center in Monroeville, Pennsylvania, University of Colorado in Boulder, and University of Wisconsin in Madison. These latter two installations are considered national facilities and were established by the National Institutes of Health in 1972 as centers for biological research. The installation at U.S. Steel was under contract with N.I.H. Division of Research Resources to explore possibilities for making these expensive facilities available. The use of the U.S. Steel facility helped to define the biological applicability of high-voltage electron microscopy, and now as this contract is being phased out the centers in Boulder and Madison are the principal areas where biological research is being conducted. In addition, there are several other high-voltage electron microscopes operating in the United States, but most of these are less than 1 MeV and are readily accessible to outside investigators.

The N.I.H. Division of Research Resources centers at the University of Colorado under the direction of Keith Porter and at the University of Wisconsin under the direction of Hans Ris have been in operation for several years with few technical problems. Both installations are used frequently by investigators residing outside the host institutions, and financial support from N.I.H. is possible. Individuals wishing to use the microscopes are required to submit a 3-4 page proposal which supplies such information as a description of the proposed project and the reasons why the use of high-voltage electron microscopy would be useful. In addition, there is required such standard information as curriculum vitae, current grant support, specific preparative methods, and an estimate of the microscope time required for completion of the project. Approved proposals are awarded microscope time, transportation expenses, and a per diem allowance. Funds for other aspects of the research project are not available through this agency, and if one wishes to get further information or to request a proposal application one should write:

Program Director
Biotechnology Resources Branch
Division of Research Resources
National Institutes of Health
Building 31, Room 5B-19
Bethesda, Maryland 20014

D. Biological Applications of High-Voltage Electron Microscopes

1. Thick Sections

Sections in the thickness range of 0.25 to 1.0 μm are useful for high-voltage electron microscopy where fine structural information is required. Even though the high-voltage electron can penetrate even thicker specimens, usually the ultimate specimen thickness limiting factor is related to the complexity of the structures being studied. Since micrographs of thicker sections represent superimposed two-dimensional images of three-dimensional components, the use of stereoscopic microscopy allows one to interpret biological information from relatively thicker sections.

2. Serial Sections

Reconstruction of three-dimensional structures by thin-serial sectioning in both tedious and subject to

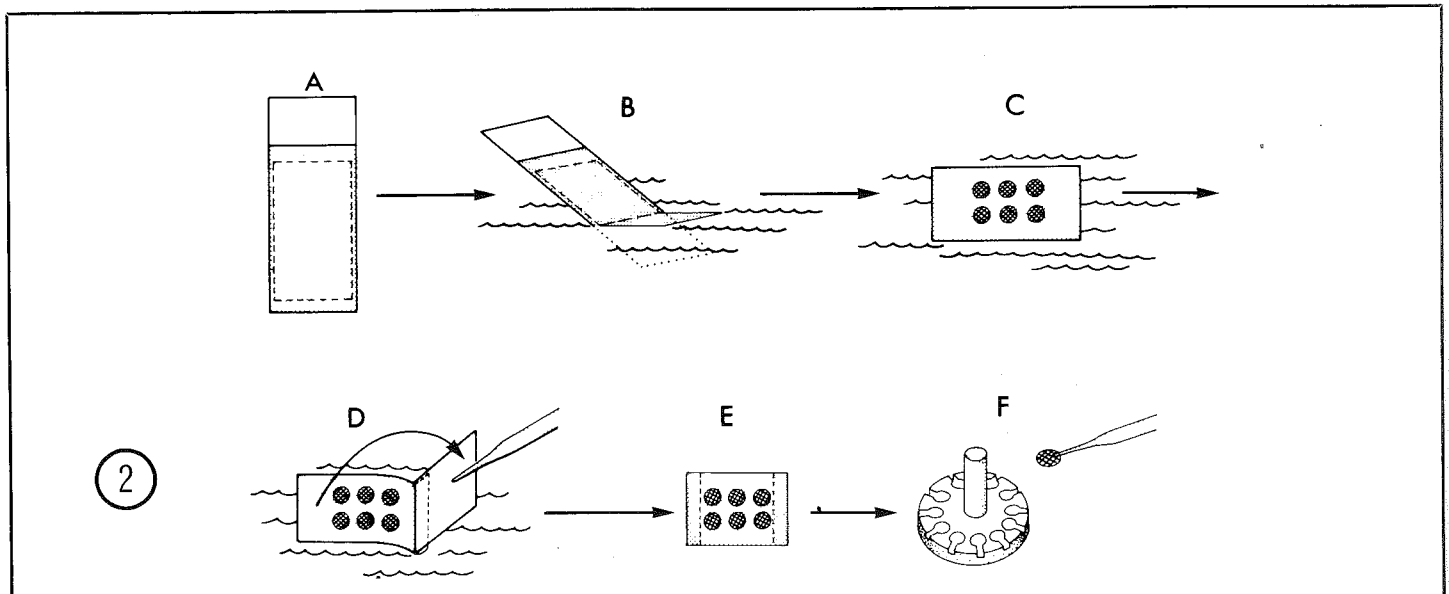


FIGURE 2 — *Technique for preparing Formvar-coated grids. (A) Glass slides are dipped in 0.7% Formvar and floated on distilled water (B). Gold grids are placed on the floating Formvar film (C) and picked up with standard glass coverslips (D). After U-V sterilization, cells are grown on these grids (E) and subsequently fixed and critical point dried in a specially prepared holder (F).*

numerous pitfalls. The use of thick-serial sections reduces the overall effort 10-20 times and does not result in the distortion usually encountered by cutting thin sections. Not only is this process easier to manage and more reliable, but one can now morphologically study larger systems that would not be feasible using thin-serial sections. The high-voltage electron microscope facility at Boulder is equipped with a videotape recorder which allows easier orientations of each succeeding section. In addition, several investigators are using digital computers not only for storing selected structures, but also for retrieval and three-dimensional image display and rotation.

3. Autoradiography

The use of standard transmission electron microscopy for examining autoradiographs requires that both the section and photographic emulsion be very thin. Usually this requires exposure times of 2-3 months to obtain a useful concentration of silver grains. With high-voltage electron microscopy, images can be obtained with 0.5 μm thick sections covered with emulsions at least 2-3 times the usual thickness. Using these conditions, one can obtain reasonable resolution with only 2-3 days' exposure, which results in a considerable savings in time by using high-voltage electron microscopy.

4. Isolated Cell Components

Fractionation of cell components and their isolation for biochemical studies have greatly increased the need for microscopy of these isolated cell components that are whole mount prepared. Isolated structures such as chromosomes, mitochondria, centrioles, and the mitotic apparatus are being investigated with high-voltage electron microscopy.

5. Selective Staining

By selectively staining only certain components such as the sarcoplasmic reticulum, lysosomes, or neuromuscular junctions, one can eliminate extraneous details often encountered in thicker sections. The use of tannic acid, acid phosphatase, and bismuth staining are being investigated in several laboratories.

6. Unprepared and Wet Specimens

Until the recent availability of high-voltage electron microscopy, there was little hope of observing biological specimens unaltered by preparation artifacts and in a natural environment instead of the usual microscope vacuum. The use of hydration chambers operating at high energies has enabled investigators to study such structures as human red blood cells and bacteria in an unfixed and wet buffer environment. Even though there are still some difficulties associated with these chambers, the technology is available to eliminate these problems which will open a new spectrum in many areas of biological research.

7. Whole Cells

The best way to study the organization and interrelationship of components within cells is to study the entire cell instead of thin or thick sections. With cells growing in monolayer culture, it is possible to prepare specimens for examination in the high-voltage electron microscope and to study such diverse subjects as virus-cell interactions, correlation of surface topography by scanning electron microscopy with internal morphology of the same cell by high-voltage electron microscopy, the distribution and interrelationship of fibrous proteins and other organelles within the cell, and recently the discovery of structures in the cytoplasmic ground substance not

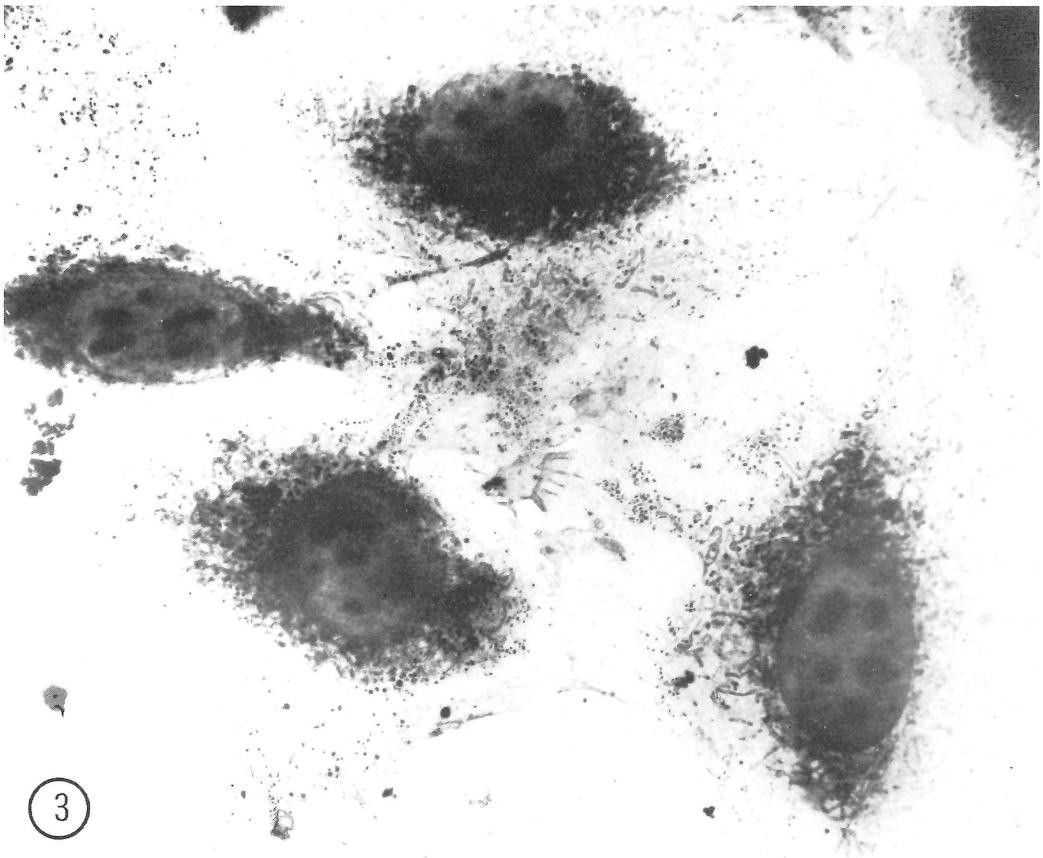


FIGURE 3 —
Low magnification of four Balb/3T3 cells prepared by standard fixation procedures and dried by the critical point method. These cells are strongly contact-inhibited, and high-voltage electron microscopy may prove useful in studying cell-cell interactions.

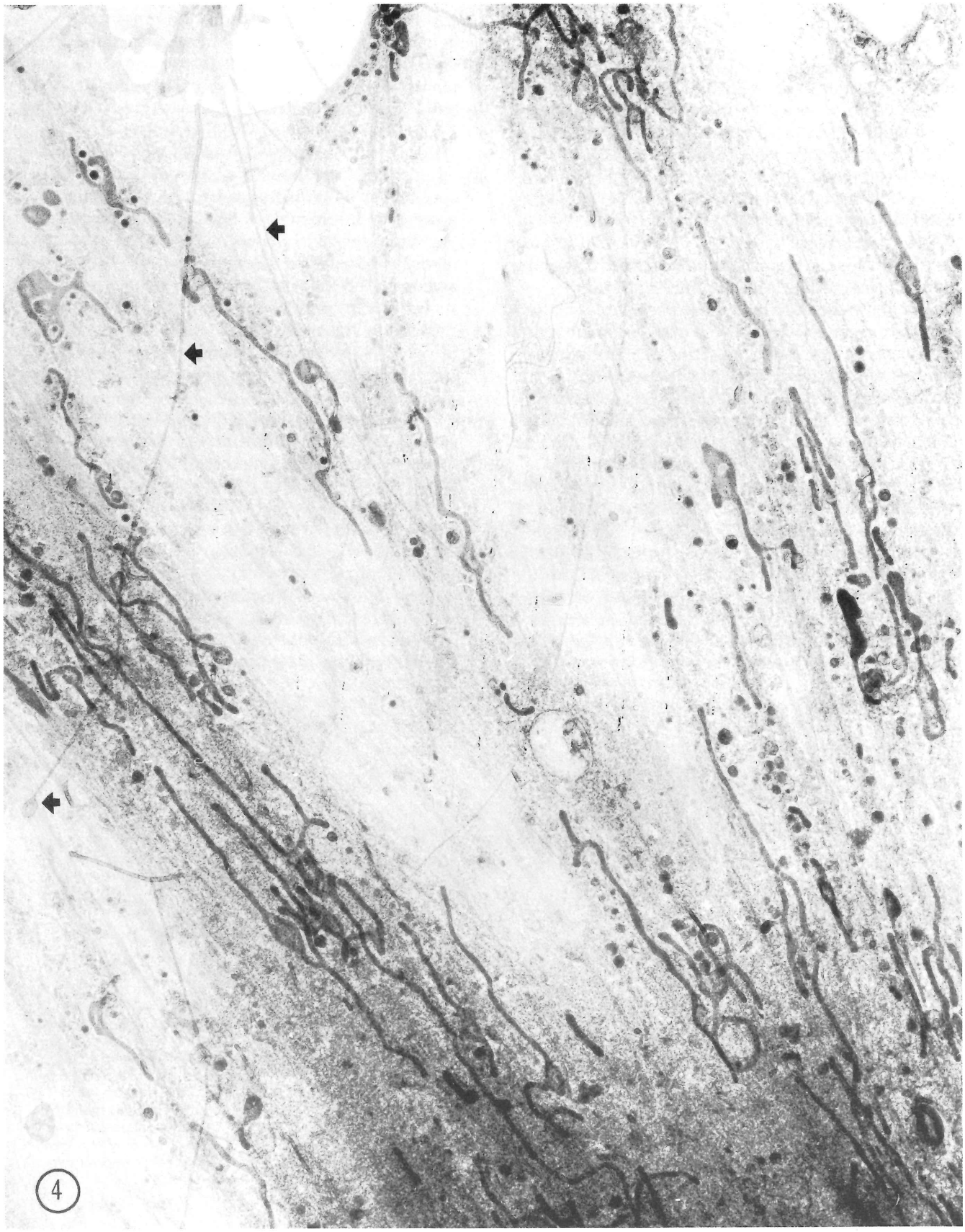


FIGURE 4 — *A portion of an L_6 myoblast as observed in the high-voltage electron microscope. In addition to numerous microfilaments and mitochondria, the arrows indicate where adjacent cells make contacts which ultimately result in membrane fusions.*

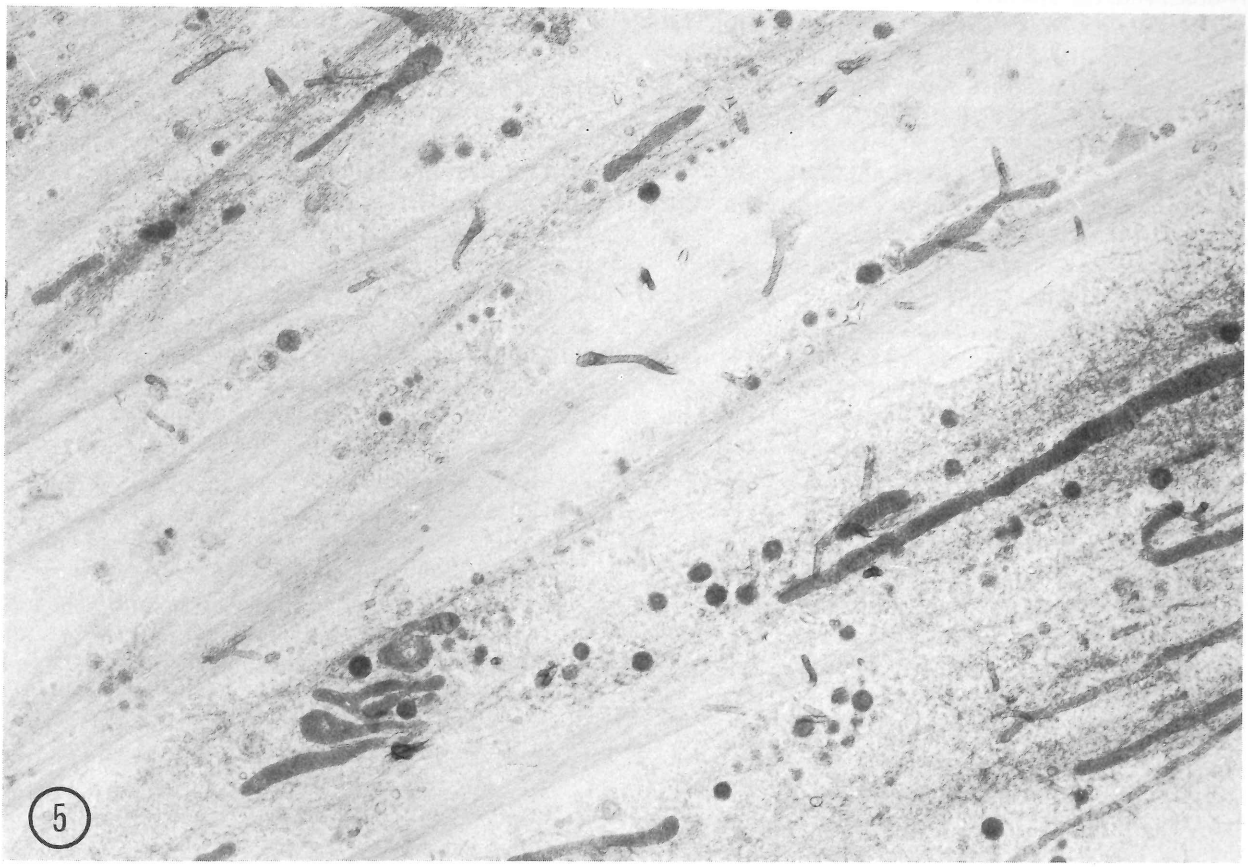


FIGURE 5 — *A portion of an L$_6$ myoblast revealing numerous bundles of microfilaments often referred to as stress fibers.*

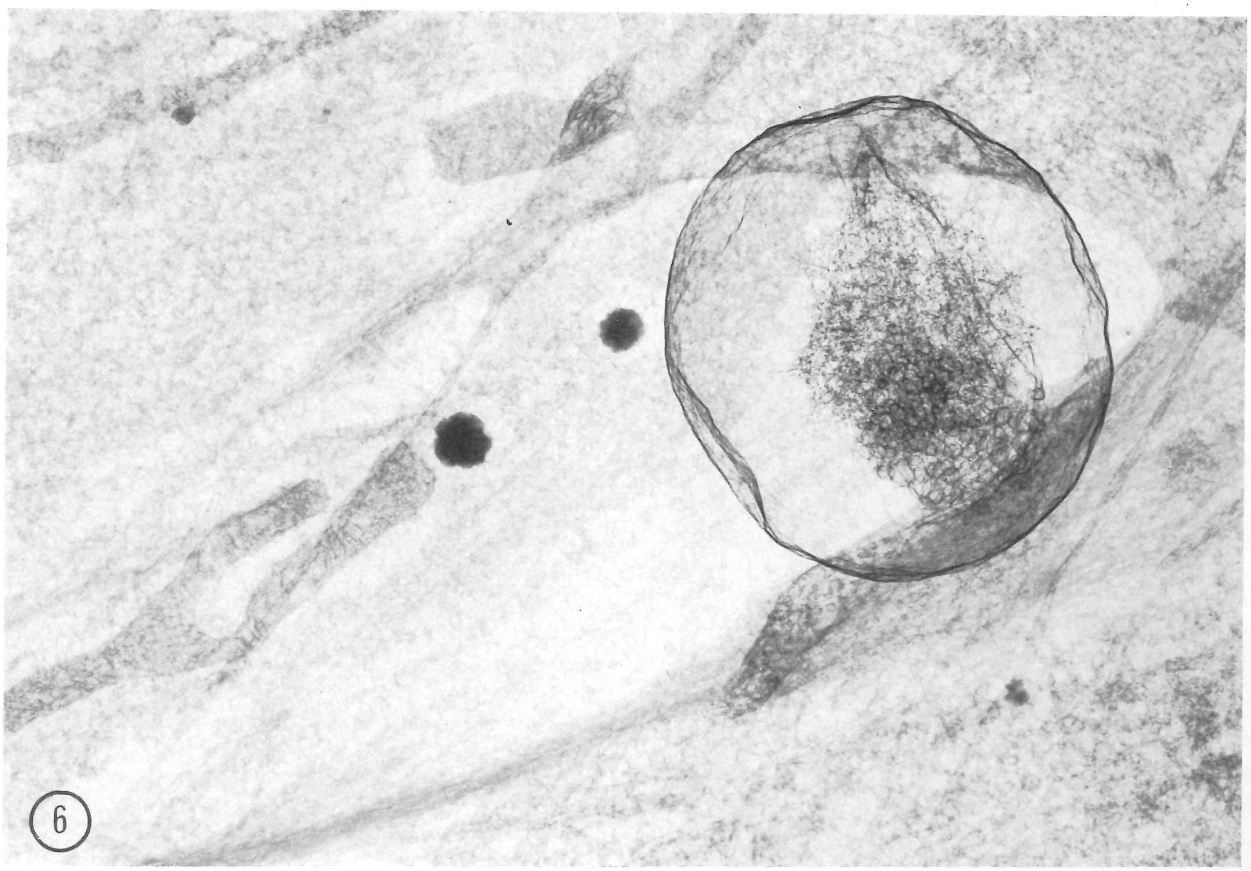


FIGURE 6 — *A higher magnification of an L$_6$ myoblast illustrating a relatively large surface bleb.*

previously observed. As this area of studying whole cells in the high-voltage electron microscope is one in which I have had some personal experience, I would like to present in somewhat greater detail a few of my observations.

E. Observations On Whole Cells in the High-Voltage Electron Microscope

As early as 1945, investigators were studying tissue culture cell whole mounts in the electron microscope (4). Even though fixation techniques were far from perfected and high-voltage electron microscopes were not available, these early studies proved to be quite informative and established the precedent for observations on whole cells when the improved technologies became available. So until recently, the main obstacles to transmission observations on whole cells have been the lack of adequate preparation techniques and the limited penetration of the low energy electron through the whole cells. To a large extent, these obstacles have been overcome by the availability of the high-voltage electron microscope and improved fixation procedures combined with drying by the critical point method. The remainder of this article will describe some of the preparation techniques and will illustrate some examples of whole cells as observed in the electron microscope.

1. Preparation Techniques

Cells for high-voltage electron microscopy were prepared essentially as originally described by Gershenbaum **et al.** (5) and more recently modified by Wolosewick and Porter (6). The cells were grown on gold grids, to avoid any toxicity, which were presented from E.F. Fullman, Inc. (Schenectady, New York). The London finder H-2 or honeycomb H-6 or H-9 grids were cleaned in nitric acid, rinsed in distilled water, and coated with a 0.7% Formvar film (Ladd Research Industries, Burlington, Vermont). The Formvar film was released from a cleaned glass microscope slide (Figs. 2a, 2b) onto distilled water. The grids were then placed on the floating Formvar film (Fig. 2c) and the film and grids were then picked up on standard 18 x 18 mm glass coverslips (Fig. 2d). After drying, the Formvar coated grids (Fig. 2e) were then placed in standard 35 x 10 mm plastic petri dishes, and various cell types could be grown directly on the coverslips in culture medium specific for the cell type being investigated.

When ready for fixation, the Formvar-coated grids, now containing cells, were removed from the growth medium, rinsed in prewarmed (37°C) Puck's Saline-G (pH 7.2-7.4), and then immersed in 2-3% glutaraldehyde in 0.1 M sodium cacodylate (pH 7.2-7.4) at 37°C for 20-45 minutes. The preparations were then rinsed in 0.2 M sodium cacodylate and then postfixed in 1% osmium tetroxide in sodium cacodylate for approximately 10-20 minutes. The preparations were then washed in distilled water and rapidly dehydrated in an acetone series. The cells were simultaneously stained and dehydrated by first removing the coated grids containing the cells, placing them in a special brass multiple-grid holder (Fig. 2f), and dehydrating in an acetone series containing 1% uranyl acetate up to a concentration of 50% acetone. The remainder of the dehydration series was completed without uranyl acetate. Finally, the cells were critical-point dried through CO_2 as originally described by Anderson (7) using equipment manufactured by Sorvall-Dupont Instruments. The dried grids were then coated with carbon and stored in a desiccator until exmained in the JEM-1000 electron microscope operated at an accelerating voltage of 1,000 KV. The observations reported here were all undertaken at the University of Colorado facility, and I would like to acknowledge Drs. Keith Porter and Mircea Fotino as well as Mr. George Wray for their help throughout these studies.

2. Observations

Figure 3 is a low magnification of four Balb/3T3 whole cells as seen in the high-voltage electron microscope. The thinner margins of the cells are probably less than 0.5 μm in thickness, whereas the juxtanuclear areas are much thicker. Even using 1,000 KV, very little useful information can be obtained in these thicker areas. Microscopes in the 3,000-10,000 KV range, which are being built, may be able to adequately penetrate such thick areas, but the complexities of the structures in these areas may be difficult to interpret, even using high-voltage electron microscopy in conjunction with stereomicroscopy. The cell margins, however, are much thinner, and information concerning the organization and interrlationships of various inclusions and organelles can be easily observed. Figure 4 is a relatively low magnification of the marginal zone of a rat L_4 myoblast cell. One of the interesting aspects of these cells is that they grow as single cells (myoblasts) until confluency is reached and then the cells stop dividing and fuse to from myotubes. The myotubes then form myofibers and frequently contract spontaneously in cell culture. The actual processes of fusion are not well understood, and hopefully studies combining both scanning and high voltage electron microscopy may help elucidate certain aspects of these processes (8). In the photomicrograph, one can easily observe numerous long, slender mitochondria. In addition, the lighter areas of the cytoplasm are filled with numerous microfilaments. The lower right of the photomicrograph appears much more dense as this area is much thicker and is very close to the nucleus. Notice the filopodia extending from adjacent cells (arrows). These filopodia make initial contact between adjacent cells, thus allowing some form of communication

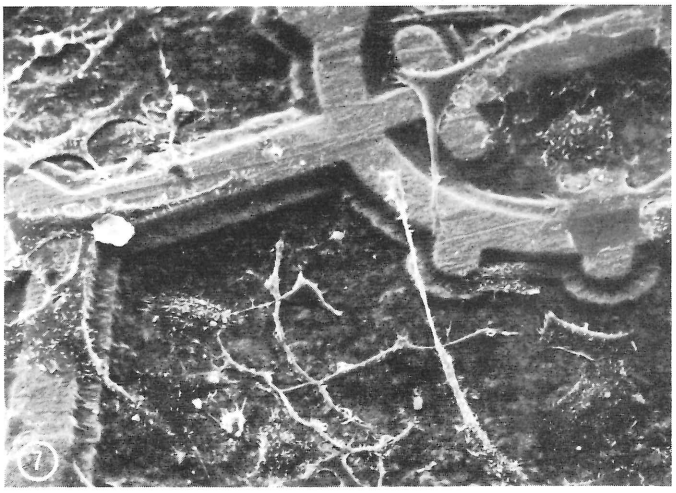


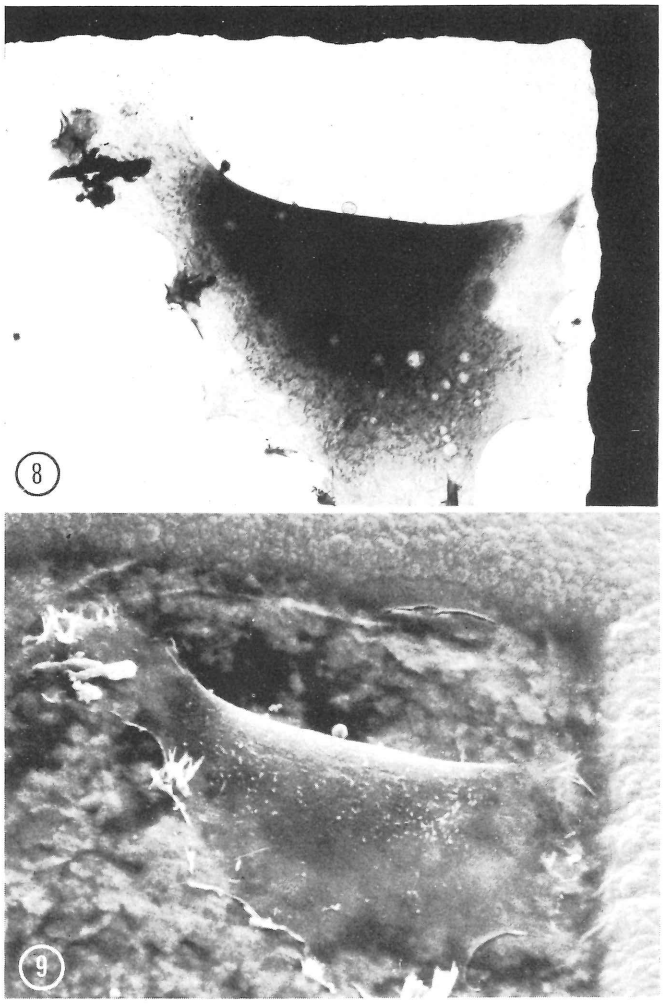
FIGURE 7 — *A scanning electron micrograph of a number of CHO cells grown on a gold honeycomb marker grid.*

between cells, which results in very little cellular overlapping and eventually in membrane fusion.

Figure 5 is also from an L_6 myoblast. In addition to individual microfilaments which measure approximately 50-70 A in diameter, there are also bundles of microfilaments which have been termed stress fibers (9, 10). At higher magnification (Fig. 6), occasional surface blebs can be seen with the high-voltage electron microscope. As revealed in thin sections by standard transmission electron microscopy, these blebs frequently contain monosomic ribosomes, but their functional significance is far from clear.

If one is interested in first looking at a particular cell in the high-voltage electron microscope and then correlating the internal structures of that cell with the surface topography of the same cell in the scanning microscope, the procedures are relatively simple. Figure 7 illustrates several Chinese hamster ovary cells grown on a Formvar-coated gold honeycomb marker grid and observed in the scanning electron microscope. Figure 8 illustrates a CHO cell in the high-voltage electron microscope, and Figure 9 illustrates the identical cell in the scanning electron microscope. Figures 10 and 11 illustrate the same concept at somewhat higher magnification. Basically, one first studies and photographs the cells of interest in the high-voltage electron microscope and then removes the grids, and after mounting on scanning electron microscope specimen studs, the samples are coated with a thin layer of carbon and gold or gold-palladium and viewed in the scanning electron microscope. As illustrated in Figure 7, the marker grids aid in relocating the identical cells.

In conclusion, looking at whole cells in the electron microscope provides an image of three-dimensional fine structure and, when combined with stereoviewing, produces an image that is not readily available by any other procedure. Instead of looking at two-dimensional thin slices through a cell, one has the opportunity to look at entire structures with the components in their normal functional relationships. One final note should be mentioned is that most electron microscopists do not realize that the procedures presented here for observing whole cells in the high-voltage electron microscope can also be applied to the standard transmission electron microscope operated at 100-125 KV. Even though beam damage is somewhat greater, it is surprising the amount of useful information one can acquire by adapting these techniques to the instruments that are more readily available.



FIGURES 8 & 9 — *High-voltage and scanning electron micrographs of the same Balb/3T3 cells.*

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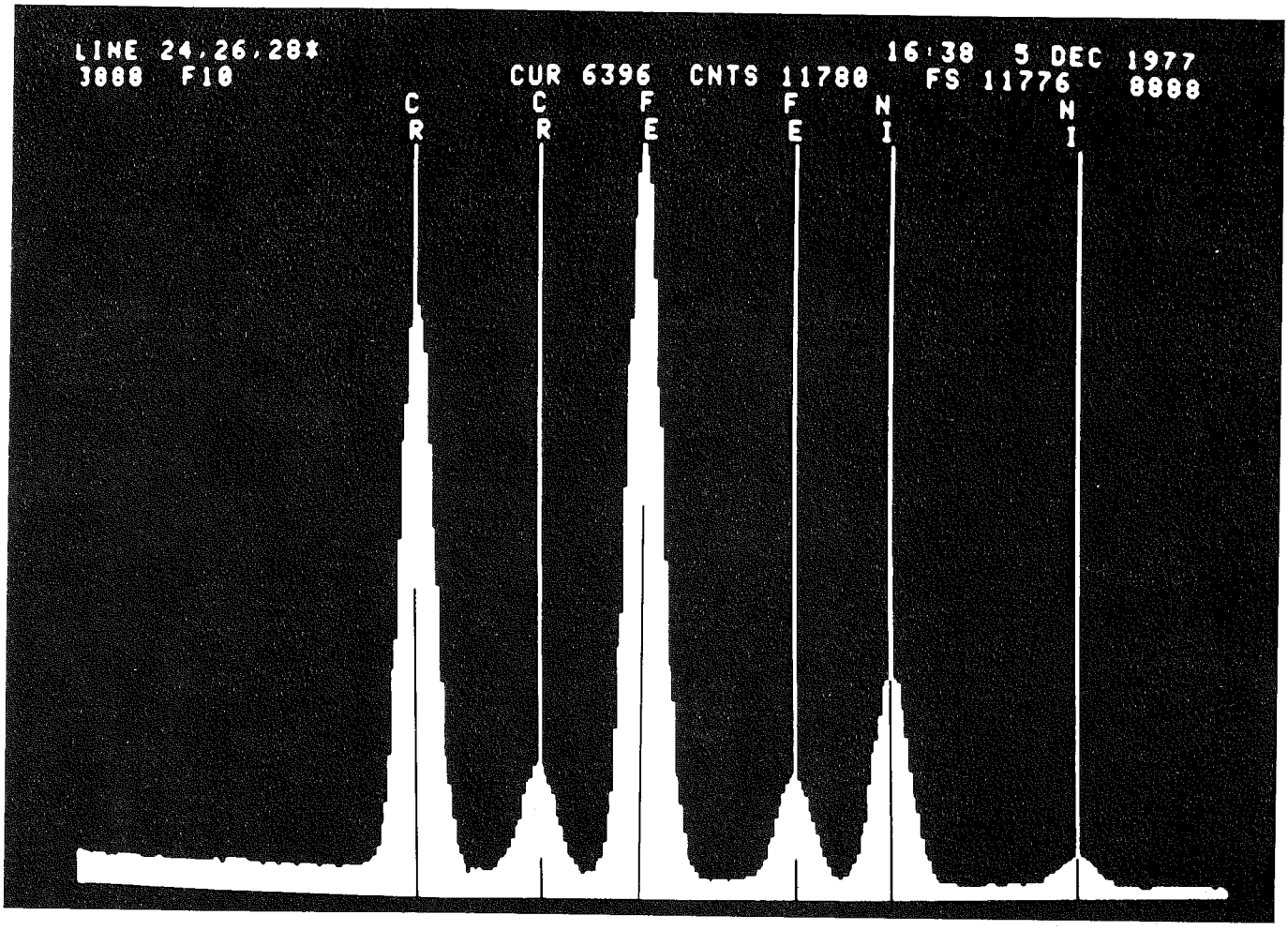
Grant support from N. I. H. Division of Research Resources is gratefully acknowledged in partial support of these investigations. In addition, support from the Muscular Dystrophy Associations, Inc., American &Heart Association, Texas Affiliate, and N. I. H. grant HL-20643 are acknowledged.

FIGURES 10 & 11 — *High-voltage and scanning electron micrographs of the same Balb/3T3 cells. Note the stress fibers in Figure 10.*

Abstracts

(Continued from page 45)

culture media of intact beating hearts from 19- to 20-day fetal mice. After 3 hours, autophagic vacuoles developed within both myocytes and interstitial cells and continued to enlarge with time. After 1 or 2 days exposure to the drug, large autophagic vacuoles with myeloid figures and inclusion bodies developed and, in addition, filamentous elements which resembled cardiac myofilaments were seen within many of the interstital cell secondary lysosomes. After 2 days, the total activities of both cathepsin D and glucosaminidase declined by 20% while acid phosphatase decreased by only 3%. The distribution of lysosomal enzyme activities was also altered by chloroquine treatment. One day of exposure led to a slight increase in non-sedimentable activity, whereas after two days, there was a siginificant decrease in the proportion of total activity that was non-sedimentable. The morphological changes induced by chloroquine treatment in fetal mouse hearts in organ culture are similar to those observed in other tissues, suggesting that this easily controlled system should serve as a useful model for studying chloroquine's actions on lysosomal function **in vitro.** The appearance of vacuoles within interstitial cells containing filaments suggests that proteins of the contractile apparatus may eventually enter secondary lysosomes where degradation may occur. (Supported by grants HLB 14706 and 17669)



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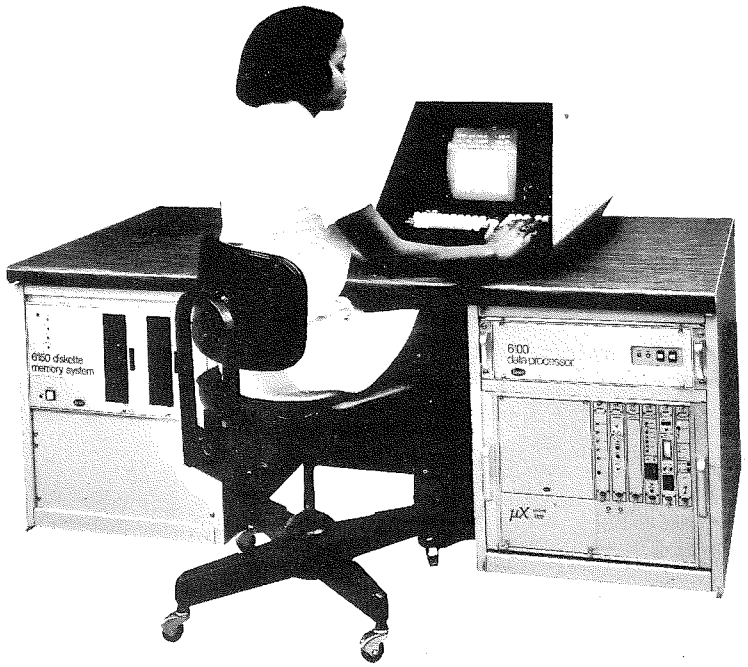

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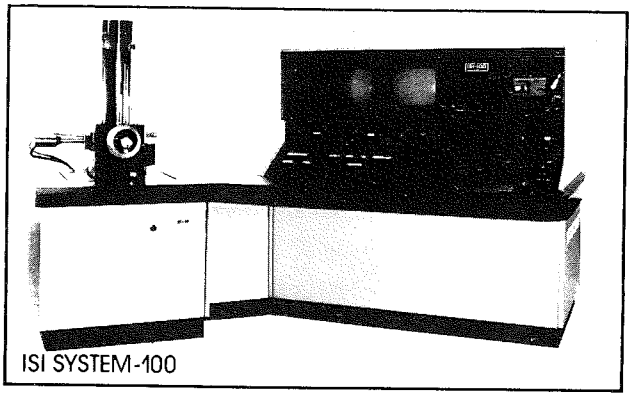

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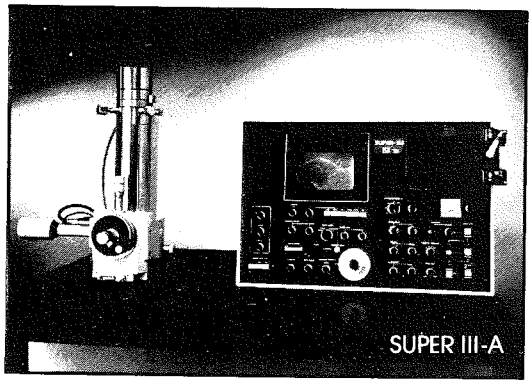

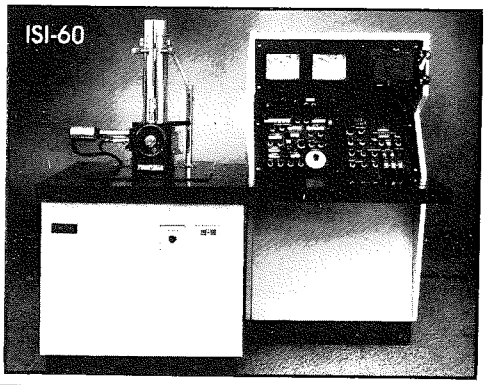

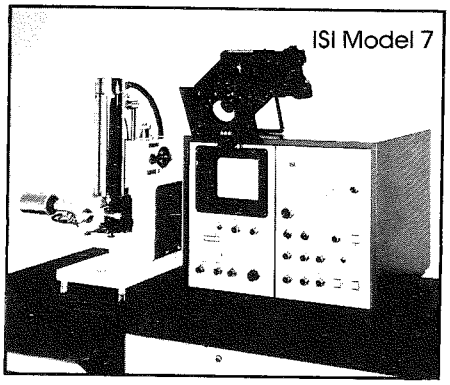

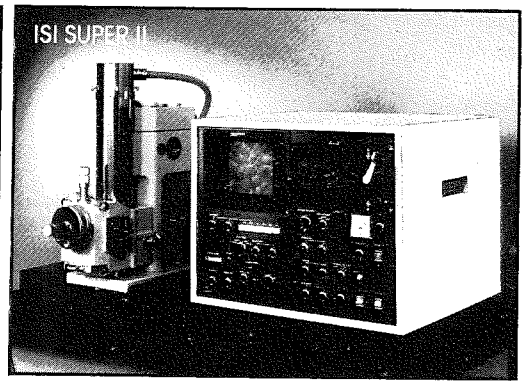

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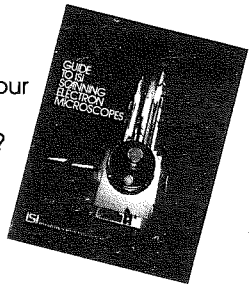

Abstracts

AN SEM-MICROPROBE STUDY OF INCLUSIONS IN CORES. D. G. Feuerbacher and M. H. Cordova, Dresser Industries Inc.

Combined scanning electron microscopy and x-ray microprobe analysis have proved to provide valuable information in studies of particulate inclusions in core samples. In particular, core samples of potential hydrocarbon-bearing formations have been examined, looking specifically for characteristics of included particles or cemented zones of particles which would aid in giving information on the formation related to oil well drilling, well completion, or mechanisms of hydrocarbon recovery. Both polished sections and fractured surfaced were examined, according to the type of information desired.

The examination of core samples was conducted along three separate lines: (1) Studies of fractured or polished surfaces to determine relative particle or grain size, cementation, and pore size and structure; (2) SEM-Microprobe identification, by both crystalline structure and x-ray spectra, of separate particulates in the core surface, and (3) Analysis of zones of cemented particulates by back-scattered, secondary electron and x-ray images.

Studies of both limestone and sandstone cores indicate that impurities in the cores are particular inclusions, sometimes cemented into specific zones. The types of mineral inclusions give an idea of the age or other geological characteristics of the formation, and grain and pore size studies correlate well with permeability measurements of individual sections of core.

AN ELECTRON MICROSCOPIC STUDY OF CYSTOLITHS IN THE LEAF OF THE MULBERRY. H. J. Arnott and L. G. Diana. Department of Biology, The University of Texas at Arlington, Arlington, Texas 76019.

Using isolation techniques developed for this purpose we have been studying the cystoliths of **Morus alba** L., the mulberry, via scanning and transmission electron microscopy and x-ray diffraction. The cystoliths are intracellular deposits of calcium carbonate having vaterite as the major crystalline species. Histochemical studies of demineralized cystoliths show that carbohydrates and proteins are the major organic components of the bodies but that the distribution of these major components is unequal. The structure of whole, demineralized and ashed specimens will be presented and compared. The nature of mineralization in these structures will be compared with that found in other plant calcification systems. Cystoliths from several other sources will also be discussed and illustrated.

A COMPARISON OF THREE METHODS FOR OBTAINING AREAL INFORMATION IN STEREOLOGICAL ANALYSIS. W. R. Fagerberg and H. J. Arnott. Department of Biology, The University of Texas at Arlington, Arlington, Texas 76019.

Several methods for gathering stereological data are currently in use today. The choice as to the best methods to use in a specific instance is often difficult to make. In order to provide some way of dealing with that decision we have completed this study.

Point counting, planimetry and digitation data acquiring methods were compared as to their accuracy, reliability and ease of application. These analysis were carried out separately by both authors on geometric shapes of known dimension. Additionally, the area of a complicated irregular figures was also determined by these methods. Such studies provide a useful guide to the selection of the most accurate and easy to use method for gathering stereological information under several circumstances.

THE EFFECT OF OXAURACIL ON HERPESVIRUS REPLICATION. ULTRASTRUCTURAL STUDIES. Richard M. Jamison, Ph.D. and Lawton A. Seal, M. S. Department of Microbiology and Immunology, Louisiana State University Medical Center, Shreveport, Louisiana.

Treatment of rabbit kidney cells **in vitro** with $^{-3}$M oxauracil (2H-1, 3-oxazine-2, 6 (3H)-dione) prior to infection inhibits the replication of Herpes simplex, Type 2, as measured by assay of infectious virus. The production of infectious Type 1 virus is not inhibited by this treatment. The morphology of control uninfected cells incubated in oxauracil-containing medium was unaffected relatively by the drug treatment. Occasional swollen mitochondria were seen, but the gross ultrastructural appearance of the cells was normal. The appearance of cells infected with Type 2 virus and treated with oxauracil was similar to that of Type 2-infected untreated cells. However, many fewer cells with mature intranuclear viral inclusions were seen. Granular matricies typical of the very early stages of Type 2 infection were seen commonly suggesting early events in the replication cycle had occurred. The inclusions in the control infected cells were more advanced; they seemed to be more dense, stained more intensely and appeared to contain more virus particles. In addition, the number of "empty" particles appearing in the nucleus and cytoplasm was elevated in cultures treated with the drug. Occasional particles were seen in the cytoplasmic vesicles without concomitant nuclear pathology.

EFFECTS OF COLD INDUCED MICROTUBULE LOSS ON KIDNEY GLOMERULAR EPITHELIAL CELLS. Peter M. Andrews, Department of Cell Biology, The University of Texas Health Science Center at Dallas, 5323 Harry Hines Boulevard, Dallas, Texas 75235.

In a recent investigation it was reported that fully differentiated kidney glomerular epithelial cells (podocytes) can be incubated at 37°C for up to 12 hours in oxygenated balanced salt solution or culture media without resultant major changes in their fine structure (Andrews, P. M., and Stauver, M., in press). In the present investigation, this **in vitro** system has been used to evaluate the effects of cold induced microtubule loss on the glomerular epithelium. When incubated at 4°C, podocytes lose their cytoplasmic microtubules within 10 minutes. As a result of microtubule loss, cytoplasm flows from the large podocyte major processes into the nucleated cell bodies. Within one hour of incubation at 4°C, major processes have become as thin as pedicles along most of their lengths and podocyte cell bodies have swollen considerably. These observations parallel changes reported in response to vinblastine induced microtubule loss at normothermic temperatures **in vivo** (Andrews, P. M., Am. J. Anat. 150:53, 1977). Concomitant with these changes in cell shape, podocyte cytoplasm decreases in electron density and cytoplasmic organelles decrease in number and size. By six hours of incubation at 4°C, podocytes exhibit a considerable degree of degeneration (i.e., plasmalemmal pitting, degeneration of cytoplasmic organelles, etc.).

THE IN VITRO INCUBATION AND STUDY OF KIDNEY GLOMERULAR EPITHELIAL CELLS. Peter M. Andrews, Marguerite Stauver, Department of Cell Biology, The University of Texas Southwestern Medical School at Dallas, Dallas, Texas 75235.

Glomeruli exposed on thin (1mm) slices of rat kidney cortex were incubated at 37°C in a variety of culture media and conditions. We found that when maintained in a continuously oxygenated culture media, glomeruli remained viable for at least four days. During this time, the fine structure of the glomerular epithelium (podocytes) exposed to the incubating solution was studied by combined scanning and transmission electron microscopy. Within the first twelve hours of incubation, podocytes exhibit an increase in number and length of free surface microprojections and an increase in number of cytoplasmic lysosome-like bodies. After twenty-four hours of incubation, most podoctyes pedicels are lost and tight junctional complexes form between podocyte processes. During the second and third days of incubation, podocyte cell bodies gradually swell and major processes retract toward their cell bodies. By the fourth day, the glomerular epithelium appears not unlike the undifferentiated glomerular epithelium of newborn rats. Coincident with the above changes, the underlying capillary loops decrease in size and glomerular endothelial walls thicken. However, the glomerular endothelium, like the glomerular epithelium, remains viable throughout the four days of incubation. The glomerular basal lamina remains for the most part morphologically unaltered throughout the incubation period. The uriniferous tubular epithelium surrounding surface glomeruli at first undergoes rapid degeneration and then, by twelve hours, becomes modified to form a thin sheet (one to two cells thick) of viable surface epithelial cells. These cells exhibit some morphological features characteristic of uriniferous tubule cells.

AN IN SITU PROCEDURE FOR AND THE RESULTS OF BIOPSIES OF PRESSURE WRAPPED HYPERTROPHIC SCARS. P. S. Baur, B. F. Barratt, and G. M. Brown, Department Human Biological Chemistry and Genetics, University of Texas Medical Branch, Galveston, Texas.

A plexiglass device was developed in our laboratory which can be incorporated in the therapeutic elastic wrappings of hypertrophic scars. The removal of a small access port on this device permits the exposure of the underlying tissue for full thickness biopsies without the necessity of first removing the pressure wrapping. The ultrastructural detail of tissues biopsied in this fashion more readily reflect the **in situ** microarchitecture of the pressure wrapped skin and/or scars. In scars sampled while under pressure wraps there was a marked reduction in the interstitial spaces with the collagen fibers and/or cells closely pressed to one another. This was in contrast to the configurations observed in pressure treated scars biopsied after pressure wraps had been removed, which is the normal clinical procedure. In these biopsies the interstitial spaces were very obvious and appeared to reflect the swollen nature of hypertrophic tissue which is a reflection of its high water content. There were no other apparent ultrastructural differences in cells from "under pressure" scars when compared to their "after pressure" counterparts.

This work was supported in toto by the Shriner's Burn Institute.

THE USE OF ANAGLPHS IN STEREO ELECTRON MICROSCOPY. Myron C. Ledbetter, Biology Department, Brookhaven National Laboratory, Upton, New York 11973.

There is an increasing need to present the three-dimensional information available from stereo pairs of electron micrographs to audiences by projection and through publication. In the anaglyph system a composite is prepared in two colors (usually red and green) and viewed through spectacles having filters of corresponding colors. Each filter then channels the appropriated member of the stereo pair to the proper eye to create the three-dimensional illusion. Anaglyphs may be projected onto ordinary beaded screens using either a double or single projector; they can be published as red/green overprints of any convenient size; the display prints can be made with little special photographic equipment. The means of taking the stereo pair in transmission and scanning electron microscopes and the preparation of the micrographs as analyphs will be discussed and demonstrated.

NON-RANDOM CELL DEATH DURING SPERMATOGENESIS IN THE SQUASH BUG, Anasa tristis. James K. Butler, Department of Biology, The University of Texas at Arlington, Arlington, Texas 76019.

Non-random cell death is widely recognized as playing an important role in embryonic morphogenesis. The frequent occurrence of non-random cell death during specific stages of metazoan spermatogenesis, while well documented, appears to be much less well recognized.

Sections of **Anasa** testes regularly show transverse rows of "pycnotic" cells. Careful examination shows that these cells consist of all of the cells within several cysts, therefore, they are several clones of cells. Based on the position of the pycnotic cysts among the others within the testis follicles, the cells appear to be of secondary gonial or primary spermatocyte origin. At the ultrastructural level, the degenerative process involves development of a large electron-dense granular mass in the nucleus, a prominent e^--dense cytoplasmic granule, mitochondrial swelling, and alteration of the golgi remnant causing it to develop a greatly increased affinity for the stains used. Significantly, there is little evidence of the presence of structures with the morpholocial characteristics of secondary lysosomes. A correlation of ultrastructural and light cytochemical data will be undertaken as well as a consideration of the possible functional significance of this phenomenon.

RIBOSOME-LIKE PARTICLES ARE ASSOCIATED WITH THE ACROSOMAL MEMBRANE OF GUINEA PIG SPERMATIDS. Hilton H. Mollenhauer and D. James Morre, Veterinary Toxicology and Entomology Research Laboratory, ARS, USDA, P. O. Drawer GE, College Station, Texas 77840. Departments of Biological Sciences and Medical Chemistry, Lilly Hall of Life Sciences, Purdue University, Lafayette, Indiana 47907.

Particles resembling ribosomes are associated with parts of the acrosomal membrane of guinea pig spermatids. The ribosome-like particles (RLP) are confined to that part of the acrosomal membrane covering the acrosomal vesicle. No RLP are found on any part of the acrosomal membrane in contact with the acrosomal granule. Some RLP are aggregated into polyribosome-like configurations. The RLP react to fixatives like ribosomes. For example, RLP are preserved by gluteraldehyde and osmium tetroxide but lost after permangante fixation. The RLP differ from endoplasmic reticulum ribosomes in that they are not as closely bound to the membrane surface. To our knowledge, this is the first report of membrane-associated ribosomes with any structure other than nuclear envelope or endoplasmic reticulum. The presence of ribosomes on the acrosomal membrane would mean that protein biosynthesis could be a function of the acrosome itself.

A STEREOLOGI AL ANALYSIS OF EARLY AND LATE PASSAGE EMBRYONIC SYRIAN HAMSTER

FIBROBLASTS. J. J. Brokaw[1], J. K. Butler[1], and J. R. Williams[2], Department of Biology[1], The University of Texas at Arlington, Arlington, Texas 76019 and Department of Physiology[2], The Harvard University School of Public Health, Boston, Massachusetts 02115.

A culture of ESH fibroblasts in the early portion of its **in vitro** lifespan (passage 2), and a rapidly senescing culture (passage 6), were processed for electron microscopy. Point-counting volumetry (stereology) was then employed in order to determine the relative volumes occupied by nuclei, rough endoplasmic reticulum, mitochondria, and autophagic vacuoles. All of these components are known to exhibit age-related alterations in other senescing systems. The aim of this study was to determine whether or not the relative volumes occupied by these components change as a function of age in ESH cultures.

The most consistently reported change in senescing cultures has been the extensive accumulation of autophagic vacuoles with age. In addition, the RER, mitochondria, and nuclei all show morphological changes in late passage cultures. No such changes were noted in the ESH system. Not only did the fibroblasts not exhibit an increase in the relative volume of autophagic vacuoles, they actually showed a substantial decrease. This latter observation casts doubt upon one popular theory of **in vitro** cellular aging, in which the progressive accumulation of autophagic vacuoles is thought to account, at least in part, for senescence and death of the culture.

AN ULTRASTRUCTURAL STUDY OF THE VENTRAL NERVE CORD OF CLYMENELLA TORQUATA. Claire E. Hulsebosch, Department of Zoology, University of Texas at Austin, Austin, Texas 78712.

The annelid worm, **Clymenella torquata**, has a constant number of body segments or metameres. Several investigators suggest that metameric constancy in an annelid corresponds to a constancy of neuron numbers in the central nervous sytem (CNS). It is of interest to my investigations to determine if **Clymenella** adds CNS neurons during ontogeny (as do several other annelid species) or has a constant number of neurons.

One of the difficulties in CNS neuron counts is the error introduced by including non-neuronal cells which oftentimes are indistinguishable from neurons when viewed only at the level of the light microscope. By using electron microscopy, it is possible to identify the various CNS cell types based on ultrastructural differences and to correlate these observations with the light level histology.

The results of the electron microscopic observations indicate that non-neuronal cells (supportive glial cells, migratory glial cells) have elongated, small nuclei while the neurons have larger, round nuclei. This difference in nuclear shap and size allows easy identification of cell type at the level of the light microscope.

TYPES AND RELATIVE NUMBERS OF SPECIALIZED CONTACTS BETWEEN CELL PROCESSES IN THE SPINAL SUBSTANTIA GELATINOSA OF THE CAT. Donald Duncan and Ricardo Morales, Department of Anatomy, The University of Texas Medical Branch at Galveston.

Axodendritic synapses are the most numerous of all specialized contacts in the substantia gelatinsa. They outnumber all other types combined. Next in order of numbers are the gap junctions between astrocytes followed by puncta adhaerentia, mostly between neuronal processes, but also between nerve cells and glia cells. The axodentritic synapses are about equally divided between central terminals from dorsal root ganglion cells and those between spinal interneurons. About 3% of synaptic contacts are axoaxonal, dendrodendritic and dendroaxonic. Wide gap junctions both symmetric and asymmetric but lacking synaptic vesicles contribute a tiny fraction of the total. The various kinds of junctions will be illustrated and their significance, so far as known, will be presented.

NERVE-MAST CELL RELATIONSHIPS IN THE ARMADILLO DERMIS. Robert F. Dyer, Department of Anatomy, L. S. U. Medical Center, New Orleans, Louisiana.

Histochemical studies of the dermis of the armadillo **(Dasypus novemcinctus)** reveals large numbers of cells containing granules that stain metachromatically. This staining characteristic and the cytological detail of these cells indicate they are mast cells. Light microscopy also demonstrates a close anatomical relationship of some of these cells to cutaneous nerves of the dermis, in certain cases appearing to be within the confines of the nerve sheaths. Electron microscopy depicts granulated cells with the features of mast cells in the endoneurial space of the dermal nerves. This relationship has been seen in subepidermal cutaneous nerves consisting of a single Schwann cell and its associated unmyelinated nerve processes that is surrounded by a single perineurial cell. Studies using injected trypan blue demonstrate degranulation of dermal mast cells in a sequential exocytosis manner. The granulated cells of the endoneurium respond in a similar fashion, indicating that they are mast cells and that they are not protected from the degranulating agent by surrounding connective tissue elements. The role of mast cells withing the endoneurium cannot be determined from this study, but the results lend support to the concept that histamine may serve as a chemical mediator for cutaneous nerves, specifically those that are involved in pain recognition.

ULTRASTRUCTURE OF THE CAUDAL INTEGUMENT (RESTING STAGE) IN THE JUVENILE SKINK, EUMECES FACIATUS. M. Lynn Davis, Department of Biology, Stephen F. Austin State University, Nacogdoches, Texas 75962.

The five-lined skink, **Eumeces fasciatus**, is a lizard native to Eastern Texas. Juveniles possess a bright, blue tail which becomes brown in adults.

The integument includes two major subdivisions, the epidermis and the dermis. Subjacent to the dermis lies a hypodermal layer followed by striated skeletal muscle.

The stratum corneum of the epidermis at the resting stage of the sloughing cycle is characterized by outer and inner layers of flattened keratinized cells. Several layers of epithelial cells contain small to large nuclei and numerous mitochondria. A basement lamina and a layer of collagenous connective tissue separate the epidermis from the dermis.

The dermis consists of a ground matrix of collagen in which two types of chromatophores are found. The iridiophores are thought to contain reflective crystals of guanine and are the outermost chromatophores. Lying beneath them are chromatophores known as melanophores. These are filled with granules of melanin. The blue coloration of the tail in juvenile individuals is thought to result from a characteristic arrangement of iridiophores and melanophores.

ISOLATION OF CHLORIDE CELLS FROM THE BRANCHIAL EPITHELIUM OF A EURYHALINE TELEOST. Seth R. Hootman, Natalia Raphael and Charles W. Philpott, Department of Biology, Rice University, Houston, Texas 77001.

Several lines of evidence suggest that the chloride cells of the teleost gill are principally concerned with electrolyte regulation. The heterogeny of cell types of the branchial epithelium has, however, hampered studies of specific mechanisms by which chloride cells perform this vital function under changing environmental conditions. We have therefore

developed a procedure for the rapid isolation of enriched populations of chloride cells from the euryhaline pinfish, **Lagodon rhomboides**.

Gentle mechanical dissociation of the branchial epithelium in a Ca, Mg-free balanced salt solution containing 2 mM EDTA yielded a suspension of mixed cell types. This suspension could be separated into more homogenous cellular fractions having different densities by isopycnic centrifugation in a discontinuous density gradient of Ficoll (400.00 mol. wt.). In this manner a greatly enriched fraction containing 50 to 70 % chloride cells was routinely obtained from seawater-adapted pinfish. The activity of the transport associated Na+K-ATPase, an enzyme concentrated in chloride cells, was enriched three to four fold in this fraction over comparable activities in tissue homogenates, and the isolated cells exhibited excellent ultrastructural integrity. The isolation procedure thus facilitates examination of the underlying molecular mechanisms by which chloride cells effect electrolyte regulation. Supported by NSF grant P4B3634.

AN ELECTRON MICROSCOPIC STUDY OF THE BARORECEPTOR NERVE ENDINGS IN THE INTERNAL CAROTID ARTERY OF THE RAT. R. D. Yates and I-li Chen, Department of Anatomy, Tulane Medical School.

The region of the common carotid artery bifurcation into external and internal branches was removed from adult rats in order to locate and study the baroreceptor nerve endings. Anesthetized animals were sacrificed by intracardiac perfusion with 1% glutaraldehyde/1% paraformaldehyde in phosphate buffer (pH 7.1). Approximately 2 mm each of the common, external and internal carotid arteries were removed, postfixed in osmium tetroxide, dehydrated in ethanols and embedded in Epon 812. In each experiment the region dissected included the carotid body.

The portion of the carotid arterial bifurcation which exhibited baroreceptor nerve endings most frequently was located along the internal carotid artery on the side opposite the carotid body. The baroreceptor region did not display any noticeable enlargement as is characteristic of the carotid sinus of other species.

Close examination of the baroreceptor endings showed numerous mitochondria, small clear cored vesicles, abundant glycogen particles, and on occasion, dense cored vesicles in limited numbers. These nerve endings were situated mainly along the medio-adventitial border.

Our findings will be valuable in determining if morphological alterations occur in baroreceptor nerve endings and/or arterial wall structure in a hereditary strain (spontaneously hypertensive rats) which develop hypertension with increasing age.

STEREOLOGICAL ANALYSIS OF MITOCHONDRIAL DEVELOPMENT IN GERMINATING PHASEOLUS VULGARIS L. AXIS. Betty Hamilton and Glenn Todd, School of Biological Sciences, Oklahoma State University, Stillwater, Oklahoma 74074.

The early stages of seed germination are marked by rapid initiation of various metabolic systems as the tissue undergoes the transition from dormancy to active development. Respiratory uptake of O_2 is one of the first measurable metabolic activities, and is crucial to the initiation and continued function of other metabolic systems. Early respiratory activity parallels increased fresh weight of the tissue. Hydration occurs rapidly, within two hours in excised **Phaseolus vulgaris** L. axis, and is followed by a plateau phase in which neither fresh weight nor respiratory activity change rapidly. The plateau phase is terminated by steadily increasing respiratory rate and fresh weight. Once this transition occurs, the tissue is committed to the sequence of activities leading to development of the embryo into a mature plant.

The object of this study is to establish the relationship between changes in respiratory activity and changes in mitochondrial structure during the early stages of germination. Using the excised axis of **P. vulgaris** as a model system, the stereological parameters V_V, S_V, S/V, and n have been calculated for mitochondria at three points in the previously described sequence: the end of rapid hydration (two hours), the end of the end of the plateau phase (six hours) and the period of increasing respiratory activity (sixteen hours). The data provides insight into developmental processes occuring during the earliest stages of germination.

ISOLATION AND CHARACTERIZATION OF MYXOCOCCUS DISCIFORMIS, A PEAT BOG—ASSOCIATED MYXOBACTER. Leonard A. Hook, John M. Larkin, Department of Microbiology, LSU, Baton Rouge, Louisiana, and Ellis R. Brockman, Department of Biology, Central Michigan University, Mt. Pleasant, Michigan.

Myxococcus disciformis Thaxter 1904 was isolated from Vestaburg Bog (Davis Lake), Montcalm County, Michigan. This study, as well as previous ones, found **M. disciformis** to be associated with a peat bog dominated by **Sphagnum** sp. The myxobacter is microbiolytic, capable of utilizing living bacterial cells and autoclaved yeast cells as sources of carbon and energy. The isolate is a typical fruiting myxobacter that undergoes a dimorphic life cycle involving a morphogenesis from long, slender vegetative rods (that are motile by gliding) to shortened, spheroid myxospores. Fine structure analysis revealed a typical gram-negative cell envelope. The fruiting bodies are composed of sporangia that are often flattened, lobed structures, attached in clusters to the substrate either directly, or by individual sporangiophores. The sporangium which contains the myxospores, is enclosed by a thin wall, an important taxonomic criterion which may exclude the bacterium from its present taxon, the family **Myxococcaceae.**

A SCANNING ELECTRON MICROSCOPIC STUDY OF THE NUMBER OF FIBERS ON COTTON SEEDS. Jerry D. Berlin, Department of Biological Sciences, Texas Tech University, Lubbock, Texas 79409.

Cotton fibers are single cells that arise from the outer epidermal layer of cotton seeds. Some cells in this layer fail to initiate elongation and may be described as nonfiber cells. The ratio of fiber cells to nonfiber cells is, therefore, of considerable agronomic interest. We have undertaken a scanning electron microscope examination of different cotton species, varieties, and cultivars in an attempt to characterize fiber initiation and fiber:nonfiber ratios.

On the day that fiber elongation begins in cultivated cottons, i.e., the day of anthesis, it is possible to find some cotton seeds with fiber initials populating up to 80%, of the seed surface. In contrast, other seeds lack fiber initials on the day of anthesis. To optimize fiber quantity per seed may well require selecting genetic stocks with maximum surface areas of seeds devoted to fiber initiation on the day of anthesis.

The ratio of fiber to nonfiber cells in the outer epidermal layer of so-called naked seeds approximates 1:20. Commercial cultivars manifest nearly a 1:10 ratio. These results suggest that this ratio is genetically controlled. The possibility of reducing this ratio even further will be discussed.

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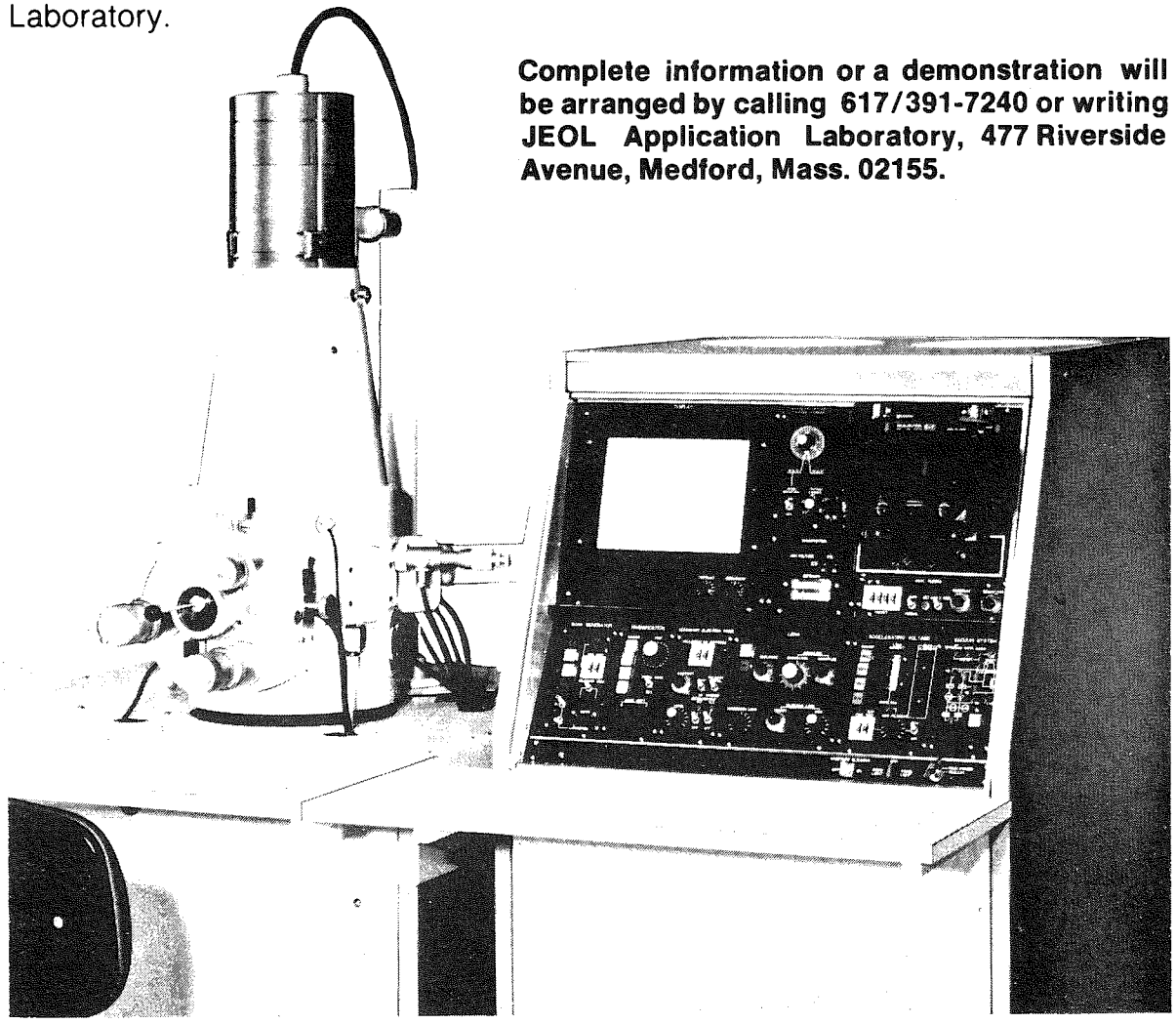

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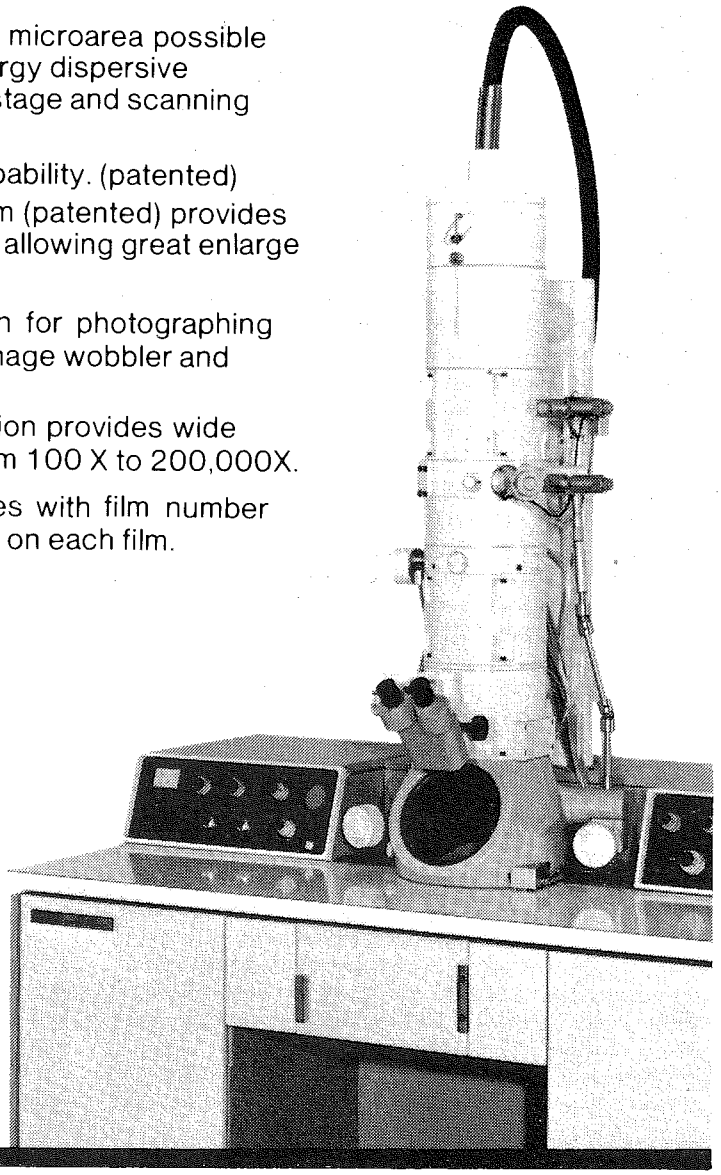

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MORPHOLOGICAL AND LEACHING CHARACTERISTICS OF THERMOPHILIC MICROORGANISMS ISOLATED FROM AN EXPERIMENTAL ORE BODY. V. K. Berry, Department of Anatomy, The University of Texas Health Science Center, San Antonio, Texas 78284.

Bacterial leaching of low-grade sulfide copper ore is now commercially practiced in many parts of the U.S.A. Iron and sulfur-oxidizing bacteria catalyze the oxidation of sulfides to water soluble sulfates and Fe^{2+} to Fe^{3+}. The mesophilic microorganism **Thiobacillus ferrooxidans** is known to take part in these reactions. These reactions are exothermic and in an in-situ or dump leaching operation, temperatures inside the dumps are known to reach as high as 80°C. The mesophilic microorganism does not survive at this temperature. In the study presented herein, a temperature profile of a 185 ton ore body was recorded at different distances along its height over a period of several months. The temperature of this ore body increased over a period of time and reached as high as 60°C. A nutrient medium was inoculated with samples of ore withdrawn from the parts recording the highest temperatures. Sulfur was used as the energy source and the culture medium incubated at 55°C. A very good growth of bacteria was noticed after a period of 10 to 14 days. Morphological studies of these microorganisms was done both by TEM and SEM. Two kinds of microorganisms were observed, one perfectly round in shape approximately 1μ in diameter and the second one rod shaped approximately 1.5μ in length and slightly less than 1μ in diameter. Leaching characteristics of these microorganisms were studied by stationary flask leaching at 55°C. These microorganisms are found to catalyze the oxidation reactions of Fe^{2+} to Fe^{3+} and sulfides to water soluble sulfates. Leaching of low-grade ores shown by the enhanced corrosion of pyrite and chalcopyrite surfaces by scanning micrographs is compared with the sterile control samples. The quantitative data on Cu solubilization and on Fe^{2+} and Fe^{3+} in solution in the control and inoculated specimens is also presented. The laboratory studies herein indicate the presence of microorganisms in large ore bodies active and continue the leaching process in the dumps at temperatures above 35°C, a limit for the active growth and survival of **T. ferrooxidans.**

MORPHOLOGIC AND RADIOAUTOGRAPHIC STUDIES OF THE DIGESTIVE GLAND CELLS OF DROSERA CAPENSIS AND THE ROLE OF THE GOLGI APPARATUS IN SLIME PRODUCTION. Robert L. Outenreath, The Cell Research Institute, The University of Texas at Austin, Austin, Texas 78712.

The digestive gland cells of the carnivorous plant, **Drosera capensis**, are being investigated with emphasis on the Golgi apparatus and its role in slime production. This system is ideally suited for studies of this type, since the individual digestive glands unfold from the leaf and begin secretion of slime at different times, thus permitting examination of the secretory process at various stages on the same leaf. In nonsecreting glands, the Golgi apparatus are rather small, with relatively few parallel stacks of cisternae. As the glands mature and begin slime secretion, the number of cisternae appears to increase, and commonly seen are stacks of a "stairstep" morphology, having many irregularly stacked cisternae. Also common in secreting glands are curled cisternae. Neither the stairstep appearance nor the curling of the cisternae has been observed in nonsecreting glands. While predominantly small vesicles are associated with the Golgi apparatus in the nonsecreting stage, during secretion there appear to be at least three morphologically different vesicle types which are Golgi associated. The relative number of one of these vesicle types appears to increase markedly during feeding of the glands, and radioautography using tritiated galactose has shown label to be associated with this vesicle type. Currently radioautographic techniques using other tritiated sugars and amino acids are being employed to further investigate the nature of these vesicles, and to determine what role, if any, they play in relation to the Golgi apparatus and/or in slime production.

CHEMICAL CHARACTERIZATION OF PARTICULATES FROM COAL GASIFICATION AND RELEVANCE TO INHALATION TOXICOLOGY. D. L. Davidson and E. M. Gause, Southwest Research Center, San Antonio, Texas.

Trace elements have been reported to be preferentially concentrated by condensation on the surfaces of combustion-derived particles, resulting in environmentally effective concentrations much higher than would be obtained from bulk particle analyses. Surface deposition also results in a greater specific concentration of trace element the smaller the particle diameter, indicating that particles in the respirable size range could have biologically significant concentrations of toxic elements which would not be detectable by conventional bulk analyses.

In this work, particulate matter from a coal gasification plant was chemically characterized using X-ray energy dispersive spectroscopy (Microprobe) for bulk analysis and X-ray photoelectron (ESCA) and Auger electron spectroscopies for surface analysis. Considerable concentrations of both lead and sulfur were detected on particle surfaces, which were not revealed by the bulk analysis. Analytical results are presented and the relationships of bulk and surface chemistry of such particles to their inhalation toxicological properties and their capacity for pulmonary clearance are discussed.

SPECIMEN DISC-PERMA STUB SYSTEM FOR SCANNING ELECTRON MICROSCOPY. Michael T. Postek, Department of Botany, Louisiana State University, Baton Rouge, Louisiana 70803, and William A. Ladd, Ladd Research Industries, Inc., P. O. Box 901, Burlington, Vermont 05402.

The safe storage of specimens has been a problem facing scanning electron microscopists who must retain their samples. Removal of the specimen from the stub for independent storage is in many cases impossible due to specimen fragility. Therefore, the general procedure has been to include the entire stub with its attached specimen in storage. Due to the size of some stubs this presents a space utilization problem and also increases operating costs, as stubs that are in storage cannot be used for new specimens. Presented in this paper will be an alternative to this inefficient specimen system for SEM. This system consists of a permanent stub designed to accept and firmly hold easily removable, replaceable discs upon which the specimens are mounted. The perma-stub is designed to fit each SEM model and can remain with the instrument. The specimens are mounted on removable discs and stored until viewing in a specially prepared storage box. This SEM disc-perma-stub system provides standardization in SEM specimen mounting and has the distinct advantage of safely storing a large number of specimens in a fraction of the space previously required.

REINFORCEMENT OF IMAGE DETAIL BY INTEGRATING TWO DIFFERENTLY COLORED MICROGRAPHS. Mel Watson, Department of Biology, Texas A&M University.

The application of photographic techniques to reinforce periodicities by amplitude contrast presents very substantial problems. The technique of linear integration, for example, obscures detail present in the individual superimposed images

and may yield spurious results, especially if multiple periodicities are present in a single micrograph. The peculiarities of individual regions of the micrograph are lost. Using a combination of color and amplitude contrast, it is possible to superimpose and laterally displace two images without obscuring the details of either the orginal images or the resultant composite image. This can be accomplished by printing each micrograph on a separate transparency in a contrasting primary color. The two transparencies are then superimposed and displaced as desired. If the displacement approximates a periodicity in the micrograph, the periodic structures will be differentially colored. When the displacement equals the periodicity exactly there are no primary-colored fringes around the periodic structures. Only periodic structures produce differentially colored periodic arrays and it is difficult to produce a spurious image with an incorrect periodicity. Because periodic structures can be differentially colored, it is possible to substract many unwanted components of an electron micrograph and also to superimpose several enhanced periodicities onto a single composite micrograph. The technique will be illustrated using micrographs of flagella and the results compared with those obtained by optical diffraction.

INADEQUACIES OF CONVENTIONAL METHODS OF FLOATING THIN SECTION HERDING AND A SOLUTION. Alan B. Weckerling, Weckerling's World, 6702 Cypress Lake, San Antonio, Texas 78244.

Since the advent of ultramicrotomy, the proper orientation and grouping of floating thin sections for grid pick-up has been a requirement. To meet this need, dedicated ultramicrotomists have butchered small paint brushes or tortured ourselves (or our associates) by tearing living eye lashes from very tender tissue. The problem with these sources is obvious: accidently cutting off the last hair on the paint brush; the refusal of the body to give up one of its beloved members; or the "donated" eyelash comes complete with mascara. After enduring all of these hardships, the brave scientist finds that the hair is too short to reach the sections or that it is too flacid to do the job. However there is hope. . .I have found the perfect solution: (1) A hair that is gracefully curved and smartly pointed for easy section herding; (2) Is long enough to reach the sections and be easily attached to a "handle"; (3) Provides proper contrast with any boat or lighting conditions. This marvelous discovery will be described, demonstrated, and it is inexpensive commercial availability announced.

ELECTRON MICROSCOPIC STUDY OF ANAPLASTIC THYROID CARCINOMA. James R. Newland and Bruce Mackay, Department of Pathology, M. D. Anderson Hospital and Tumor Institute.

Anaplastic thyroid carcinoma is an undifferentiated tumor composed of pleomorphic spindle and giant cells. It is a biologically aggressive, rapidly fatal neoplasm that is generally considered to arise from glandular epithelium of the thyroid follicle. Anaplastic thyroid carcinoma may be associated with differentiated thyroid carcinomas (follicular, papillary, mixed and Hurthle cell) and this association is helpful in making the diagnosis of anaplastic carcinoma by light microscopy. When differentiated elements are not present the diagnosis can be difficult and confusion with sarcomas is frequently a problem.

Fourteen anaplastic thyroid carcinomas have been studied with correlated light and transmission electron microscopy. In light microscopic sections, areas of differentiated thyroid carcinoma were found in ten cases. Ultrastructural examination of the anaplastic thyroid tumors revealed evidence of epithelial differentiation. The study illustrates the value of electron microscopy in establishing the diagnosis of anaplastic thyroid carcinoma and in differentiating between anaplastic thyroid carcinoma and sarcomas.

HLA TYPING OF MELANOMA AND COLON TUMOR CELL LINES AND THEIR CORRESPONDING ULTRASTRUCTURAL FEATURES. Cameron E. McCoy, Nancy D. Mabry, Wm. B. McCombs and A. Leibovitz, Scott and White Clinic, Temple, Texas 76501.

Melanoma and colon carcinoma cell lines established at Scott & White and tested for histocompatibility (HLA) antigens had readily identifiable A and B locus antigens on all the melanoma cells using the cytotoxicity test and unidentifiable antigens on all the colon tumor cells. All available alloantisera lysed the colon tumor cells upon addition of rabbit complement. The six melanoma cell lines tested had in common Aw30 or Aw31, and half had either B8 or Bw22.

Ultrastructurally, all the melanoma cell lines had similar features, but two of the six became amelanotic in vitro. All eight of the colon cell lines showed epithelial growth patterns, tonofibril bundles in the cytoplasm, a tendency to form microvilli on the free surfaces, and relatively dense glycocalyx formation.

Complement receptors on the membrane surfaces of the colon tumor cells could not definitely be located using fluorescent antibody techniques. Another possible explanation for cytolysis of the colon tumor cells is that glycoproteins making up the thick glycocalyx either mimmick or mask the glycoproteins composing the histocompatibility receptor sites.

FINE STRUCTURE OF R-3372 TRANSPLANTABLE PROSTATE TUMOR OF FISCHER-COPENHAGEN RATS. G. Seman, J. M. Bowen, and L. Dmochowski, Department of Molecular Carcinogenesis and Virology, The University of Texas System Cancer Center, M. D. Anderson Hospital and Tumor Institute, Houston, Texas 77030.

The rat prostate tumor R-3372 has been transplanted for many years in Fischer-Copenhagen F_1 hybrids. Histologically the tumor is a well-differentiated, encapsulated adenocarcinoma, often displaying papillary and cystic patterns. Microscopic structure of the tumor from rat to rat or from one transplant generation to the other appears very stable. Electron microscopy of transplants after several weeks of growth has shown the presence of a loosely organized alveolar epithelium, mostly columnar, but frequently flattened, bordered by myoepithelial cells and by a thin, often incomplete basement membrane. Irregular microvilli and cytoplasmic blebs were present at the apical surface of epithelial cells. The cytoplasm of the cells contained rounded mitochondria, dilated but short endoplasmic cavities, and numerous secretory vesicles. Secretory granules were very few. The lumen of the glands was filled with granulo-fibrillar material in which occasional bundles of 35 nm-wide microtubulee were embedded. The stroma consisted of loose connective tissue centered on small blood vessels, and contained in many places very large cells with fibroblastic characteristics. The large size of nuclei and cytoplasm of these cells, their well-developed endoplasmic reticulum and Golgi system reflected a high metabolic activity. Virus particles have not been observed in specimens so far examined. Apart from the presence of giant fibroblasts, R-3372 resembles in many respects the normal dorsal prostate tissue of rats. Supported in part by Grant CA-15438 from the National Cancer Institute, NIH, USPHS.

ACUTE EFFECTS OF MORPHINE ON MOUSE LIVER AND ADRENAL MEDULLA. Asa Thureson-Klein, Jack McC. Baggett, Jen Wang-Yang and Mary Lou Percy, Department of Pharmacology and Toxicology, University Mississippi Medical

Center, Jackson, Mississippi 39216.

Lipid accumulation begins in mouse hepatocytes within two hours after injection of morphine (40 mg per kg) and within five hours of morphine pellet implantation. Within these periods of time, the number of lipid droplets in the cytoplasm and lipoprotein particles in the endoplasmic reticulum are significantly increased. However, the most dramatic effects are observed eight hours after injection and 24 hours after pellet implantation. At these times numerous lipid droplets, often fused into large aggregates, are present. Many mitochondria are condensed and only a few stacks of the granular endoplasmic reticulum remain intact as many ribosomes are detached. Very low density lipoprotein particles are abundant in the hepatocytes and space of Disse. At the same time about 50 percent of the adrenaline in the adrenal medulla has been released while the noradrenaline content is unchanged. This is accompanied by ultrastructural changes in the chromaffin cells. Adrenaline augments fatty acid release from adipose tissue which is reflected by increased liver uptake of fatty acids and lipid formation. When the protein synthesis is suppressed, as indicated by the changes in endoplasmic reticulum and ribosomes, lipid droplets accumulate because the capacity for forming lipoproteins is compromised. Lipids are only transported and secreted after they have been coupled with proteins and carbohydrates. The steatosis may be augmented by a diminished food intake after morphine. Thus several mechanisms may be involved in the formation of a fatty liver in mice after morphine. Supported by USPHS Grants GM 15490 and 5 SO 7 RR05386-16.

ULTRASTRUCTURE OF EXTRAADRENAL CHROMAFFIN CELLS FOLLOWING INSULIN INJECTION. Joe A. Mascorro, John W. Armstead[1] and Robert D. Yates, Department of Anatomy, Tulane Medical School, New Orleans, Louisiana, and Department of Biology, Wabash College, Crawfordsville, Indiana[1].

Insulin induces a hypoglycemic state which neurogenically initiates adrenaline loss from adrenomedullary (intraadrenal) chromaffin cells. The loss is clearly manifested by morphological alterations in the electron-opaque cellular granules known to contain this catecholamine. However, the effects of this drug on the voluminous and catecholamine rich paraganglion (extraadrenal) chromaffin cells are not known. Four youg New Zealand white rabbits (350 grams) received 10 IU per 100 grams body weight of Regular Iletin Isulin and were sacrificed at 2, 4-1/2, and 5-1/2 hours following injection. Two additional rabbits served as controls. All animals were processed routinely for electron microscopic study. Untreated rabbits showed paraganglion cells which contained many catecholamine granules. The granules exhibited dense cores and randomly occupied large areas of the cytoplasm. Cells from treated animals showed striking changes in granule morphology at 4-1/2 and 5-1/2 hours following insulin intoxication. The granules displayed several degrees of degradation which included reduced core density, reticular core patterns or peripheral swelling. The changes were consistent and only very few granules remained totally unaffected. The catecholamine depleting action of insulin on adrenomedullary cells is well known. Thus, it is reasonable to suspect that altered granule morphology in paraganglion cells known to contain catecholamines may mirror a similar response. Further observations currently are in progress to determine the full effects of insulin induced hypoglycemia on the catecholamine content of the extraadrenal chromaffin system.

THE ROLE OF ELECTRON MICROSCOPY IN THE DIAGNOSIS OF RETICULO-ENDOTHELIAL DISORDERS. Bruce Mackay, Barbara M. Osborne, Richard J. Ford and James J. Butler, Department of Pathology, M. D. Anderson Hospital and Tumor Institute.

The modern approach to the diagnosis and classification of disorders of the reticulo-endothelial system involves the synthesis of morphologic findings with cytochemistry, marker studies, and clinical observations. Electron microscopy plays a limited but significant role as part of this battery of investigation. Examples of its contribution will be shown, including tumors of small lymphocytes in solid tissues and peripheral blood, and proliferative disorders involving transformed lymphocytes. The latter include B cell neoplasms containing admixtures of large cleaved and non-cleaved follicular center cells (histiocytic lymphomas), and immunoblastic lymphadenopathy and related conditions. It appears that non-stimulated B and T cells are morphologically identical, but there may be subtle differences between B and T immunoblasts. Cells of the T cell disorders, convoluted cell lymphoma and mycosis fungoides, have distinctive ultrastructural features, as do those of neoplasms of true histiocytes.

DIAGNOSTIC VIROLOGY CAPABILITIES EXPANDED BY THE USE OF ELECTRON MICROSCOPY AT A MAJOR MEDICAL CENTER. Steven K. Koester, Dermott Acton, George Kasai Virology Section; and Alan B. Weckerling, Lucy Olalde Electron Microscopy Laboratory, Department of Pathology and Area Lab Services, Brooke Army Medical Center, Fort Sam Houston, Texas 78234.

Electron microscopy can be a definite asset to the diagnostic virology laboratory for ascertaining the presence of a virus or viruses when tissue cultrue identification is difficult. This is especially true with double viral infections or with viruses which show minimal or nontypical cytopathic effect in tissue culture. Electron micrographs are a presumptive tool indicating viral size, gross morphology and location within the cell. This and micrographs of known viruses in tissue culture place the isolate in a general taxonomic category. The diagnosis is then accomplished utilizing standard tissue culture techniques. Methodology and results of typical cases will be discussed.

ADENOVIRUS 19 EPIDEMIC KERA KERATOCONJUNCTIVITIS.

Albert Leibovitz, Cameron E. McCoy and Peter G. Amaral, Scott and White Clinic, Temple, Texas 76501.

An outbreak of epidemic keratoconjunctivitis occurred in the central Texas area during the summer of 1977. Adenovirus (ADV) 19 was isolated from 9 of the 10 specimens submitted for virus studies. Ultrastructural studies in the various human tumor cell lines used for virus isolation showed ADV 19 to form inclusion bodies, virus factories and complete virions within 18 hours and rapid formation thereafter of crystalline arrays, destruction of the tissue cell's nucleur membrane, and the spread of complete virions and crystalline arrays throughout the cytoplasm. The crystalline arrays continued to enlarge in the cytoplasm and in some cases appeared to almost fill the entire cell. During the fall of 1977, other virus agents were isolated from sporadic cases of keratoconjunctivitis including types 3, 7, and 16. Ultrastructural studies of these agents and control prototypes ADV 8 and ADV 19 obtained from the NIH indicated a similar growth pattern ot the wild ADV 19 although these agents did not appear to disrupt the nuclear membrane as rapidly.

COMPARISON OF MASON-PFIZER MONKEY VIRUS AND SQUIRREL MONKEY (SAIMIRI SCIUREUS) RETROVIRUS, IMMUNOELECTRON MICROSCOPY. G. C. Smith, R. L. Heberling, and S. S. Kalter, Microbiology and

Infectious Diseases, Southwest Foundation for Research and Education, San Antonio, Texas.

Mason-Pfizer monkey virus (MPMV), isolated from "normal" and malignant rhesus monkey tissues, is the type species for the candidate oncornavirus D group, and has been found immunologically similar to type D oncornaviruses isolated from human cells in culture. Squirrel monkey retrovirus (SMRV), isolated from a variety of squirrel monkey tissues, has been shown to be morphologically and biochemically similar to MPMV. In this study, MPMV and SMRV, grown on a human rhabdomyosarcoma cell line (A204), have been compared by the indirect immunoperoxidase technique in order to define any antigenic relatedness. The results revealed the presence of a heavy, electron-dense reaction product encircling SMRV envelopes when exposed to goat ant-SMRV serum and peroxidase conjugate; however, similarly treated MPMV specimens were not labeled. Reciprocal studies utilizing goat anti-MPMV serum and conjugate displayed specific labeling of MPMV but no labeling of SMRV. Antiserum to a baboon endogenous type C virus (BaEV) did label BaEV, but no labeling of cell surfaces or envelopes of MPMV or SMRV was found when goat anti-BaEV or normal goat serum was employed. Similar results were obtained with uninfected cells. These data indicate that MPMV and SMRV do not share common envelope antigenic determinants.

ULTRASTRUCTURE OF THE HOST-PATHOGEN RELATIONSHIP IN THE DEWBERRY RUST DISEASE. Dale C. Glidewell and Charles W. Mims, Department of Biology, Stephen F. Austin State University, Nacogdoches, Texas 75962.

Transmission electron microscopy was used to examine the host-pathogen relationship in leaves of dewberry **(Rubus trivialis)** infected with the rust fungus **Gymnoconia peckinana**. Host cells are surrounded by fungal hyphae which grow between the walls of adjacent cells. Some intercellular spaces are almost completely filled with hyphae. The hyphae are septate and contain a typical complement of fungal organelles.

Host cells also contain numerous intracellular hyphae. These hyphae, which may be simple or branched, do not possess the morphological features characteristic of the intracellular absorbing organs (haustoria) produced by many rust fungi. The intracellular hyphae arise from intercellular hypae that are closely appressed to the host cell wall. Intracellular hyphae extend through the host cell wall and invaginate, but do not break the cell membrane. An intracellular hypha is separated from the host cell membrane by a sheath or encapsulation zone of unknown origin.

BACTERIAL DISTRIBUTION AND HISTOLOGICAL CHANGES OF RAT GINGIVA DURING TOTAL INTRAVENOUS FEEDING. Allan G. Kaster and Ivan L. Cameron, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas 78284.

The pattern of bacterial colonization as revealed by scanning electron microscopy and the histological changes which occur on the gingiva of rats fed solid food and of rats fed totally by vein (iv) for ten days are characterized. Bacterial habitation occurs selectively on the exposed edge of the exfoliating non-keratinized junctional epithelium and to a much lesser extent on the most coronal portion of the exfoliating hard keratinized free gingiva. No bacterial colonization is found on other hard keratinized areas of the gingiva. The selective habitation of bacterial populations on the exposed edge of the junctional epithelium is greater in the iv fed rats than in the rats fed solid food; there is also a greater abundance of plaque on the enamel surface of the molars of the iv fed rats in the area immediately adjacent to the exposed edge of the junctional epithelium. Histological observations show a widening of the junctional epithelium especially in the deeper regions of the ginigval sulcus in the iv fed rats. This increase in thickness of junctional epithelium is presumably due to the continued production of junctional epithelial cells at the bottom of the gingival sulcus with a decreased exfoliation of the cells at the top of the gingival sulcus. The decreased exfoliation is presumably due to a stasis or lack and "wear and tear" in the oral cavity of iv fed rats and is related to the increased number of bacteria found in the exfoliation areas. No sign of inflammation or bacterial infection was evident in the adjacent connective tissue of the gingival sulcus. Supported by NIH grant CA16831.

MORPHOLOGY OF TYPE A AND TYPE B PARTICLES AFTER FIXATION IN PICRIC ACID-PARAFORMALDEHYDE. J. Phillips and G. Seman, The University of Texas System Cancer Center, M. D. Anderson Hospital and Tumor Institute, Houston, Texas 77030.

After fixation in picric acid-paraformaldehyde (PAF) and staining with uranyl acetate and lead citrate, mitochondrial and cell membranes remain unstained and appear in "negative" in thin sections. PAF fixation was applied to mouse mammary tumors to determine whether new informations could be gained by this method on the fine structure of type A and type B particles. Specimens of spontaneous mammary tummors of C3H mice were fixed in PAF according to Moriarty and Tobin (J. Histochem. Cytochem. 24:1131, 1976) and embedded in epon-araldite. Results were compared with those obtained by conventional glutaraldehyde-osmic acid fixation of the same tissues. Intracytoplasmic and budding type A particles were well preserved and sharply outlined, in spite of some graininess of the background. Strands of electron-dense material, which is later to form the nucleoid of type B particles, were clearly seen in their core. Type B particles in intracytoplasmic vacuoles and in intercellular spaces appeared much more irregular in shape after PAF fixation than after conventional fixation. Their contents, in particular between core and envelope, and that of cytoplasmic fragments associated with the particles, were densely stained and extracellular viral material appeared easier to identify than in conventionally treated material. While type A particles had a similar diameter (0.084 to 0.087 nm) with both fixation methods, type B particles were on the average 20 to 30 percent smaller after PAF fixation (0.1 nm in diameter). Spikes on the envelope of type B particles were not visible. PAF fixation, which is widely used in immunocytochemistry, appears to be very useful to the study of type A particles and of non-core material of type B particles.

MOLECULAR AND ULTRASTRUCTURAL EVENTS ASSOCIATED WITH PLASMA MEMBRANE TURN-OVER IN THE AVIAN SALT GLAND. Michael P. Sarras, Jr., and Fred E. Hossler, Department of Anatomy, LSU Medical Center, New Orleans, Louisiana 70119.

We have previously established that the avian salt gland is an ideal system for studying plasma membrane turnover. Our findings indicate that by feeding ducks 1% NaCl (stressing) a de novo synthesis of Na^+K^+-ATPase results as well as a tremendous increase in the plasma membrane volume as observed ultrastructurally. Autoradiography using ^3H-ouabain demonstrates that the Na^+K^+-ATPase is restricted to the secretory tubules of the gland. Replacing the 1% NaCl diet of fully stressed ducks with a fresh water diet (de-stressing) causes a reversal of this process. Electron microscopy reveals that by day 7-9 of de-stressing, the secretory cells have lost much of their plasma membrane and have reverted in appearance to a

non-stressed state. The appearance of filamentous material and multivesicular bodies is associated with this process; however, the latter is also seen during stressing. Biochemically this loss of plasma membrane parallels a decrease in Na^+K^+-ATPase. In addition, there is a relative increase in the specific activity of the hydrolytic enzymes, acid phosphatase and peptidase. Acid phosphatase histochemistry demonstrates that this enzyme is associated with the secretory tubules. Preliminary cytochemical studies indicate that acid phosphatase activity is not associated with the multivesicular bodies (previously suspected as being lysosomal) but is rather seen dispersed through the cytoplasm of the secretory cells as well as being closely associated with the plasma membrane.

THE EFFECT OF INORGANIC CATIONS ON THE MEMBRANES OF TETRAHYMENA. Sharon M. Mattox and Guy A. Thompson, Jr., Department of Botany, The University of Texas, Austin, Texas.

Tetrahymena grown under high concentrations of cations exhibit several membrane effects. When cells are grown in media supplemented with either 300 mM NaCl or 100 mM $CaCl_2$, marked changes occur in both the fatty acid and headgroup composition of the phospholipids. Certain physiological functions are also altered. The rate of food vacuole formation is affected by Ca^{++} concentration. Some Ca^{++} is essential for food vacuole formation but higher levels inhibit this process. When cells are adapted to high Ca^{++} levels, some of the lost vacuole forming ability is regained. When monitored by the method of Kitajima and Thompson (J. Cell Biol. 72 (1977): 744), the freeze fracture patterns of most cell membranes of calcium acclimated cells are similar to those of control cells, but those of food vacuole membranes are altered. In these membranes, large particle-free domains appear at significantly higher temperatures than in control cells. The fluidity of surface membranes therefore appears to be affected by cations. The addition of K^+ (5 mM) to cells with vacuole formation partially inhibited with Ca^{++} (2, 5mM), increases the degree of inhibition. No inhibition of vacuole formation is seen with K^+ alone. This suggests that K^+ may be facilitating Ca^{++} entry into the cell by opening Ca^{++} passages located in the surface membrane. Thus, the regulation of vacuole formation in **Tetrahymena** appears to be similar to the mechanisms of cation regulation in **Paramecium**.

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ULTRASTRUCTURE OF THE CELL ENVELOPE AND SULFUR INCLUSIONS OF BEGGIATOA. W. R. Strohl, J. M. Larkin, S. Seuferer, and K. S. Howard, Department of Microbiology, Louisiana State University, Baton Rouge, Louisiana.

The cell envelope of the gliding bacterium, **Beggiatoa**, has been analyzed by thin section, freeze-etch, and scanning electron microscopy. The cell envelope contained a cytoplasmic membrane bearing numerous pores, a peptidoglycan layer, a bilayer external to the peptidoglycan, and three outer layers. Of the three outer layers, the first appeared to be a single dense layer, the second appeared to be a bilayer containing a linear array of particles, and the outermost layer was observable with ruthenium red staining. A ruthenium red-negative extracellular slime was observed to be chemically similar whether the organism was grown heterotrophically (0.05% nutrient broth + 0.05% acetate) or mixotrophically (0.03% H_2S + 0.0001% acetate). Sulfur granules, which were surrounded by an unusual trilinear membrane, were isolated from **Beggiatoa** grown in the presence of H_2S. The sulfur granule membranes were sensitive to treatment with phospholipase C, **Pseudomonas aeruginosa** hemolysin, protease, proteinase K, and lysozyme, but they were not sensitive to trypsin, SDS, EDTA, or Triton X-100. These results suggest that the membrane may contain a protein-phospholipid moiety with a muramic acid-like backbone. The development of the sulfur granules during conversion from heterotrophic growth to mixotrophic growth has also been observed. The sulfur granule membranes are present in a collapsed rudimentary form during heterotrophic growth. Upon conversion to mixotrophy, the H_2S is oxidized to elemental sulfur which is deposited within the expanded trilinear membranes. The trilinear sulfur granule membranes lie external to the cytoplasmic membrane under all growth conditions.

LECTIN INDUCED REDISTRIBUTION OF CELL SURFACE ANIONIC SITES IN HEPATOMA ASCITES CELLS. Peter C. Moller and Jeffrey P. Chang, Division of Cell Biology, Department of Human Biological Chemistry & Genetics, The University of Texas Medical Branch, Galveston, Texas 77550.

A polycationic derivative of ferritin, cationized ferritin (CF) has been developed for the visualization of negative charges on the plasma membrane. In the past, CF has been used to detect surface anionic sites on different cell types, but not hepatoma cells. Thus the effect of Concavalin A (Con A) on the distribution of anionic sites on the surface of Chang hepatoma ascites cells was examined. When these cells were treated at 25°C with Con A, washed, fixed in glutaraldehyde and then exposed to CF, the labeling on the surface of microvilli and plasma membrane was clustered and irregular. The areas of the cell surface between CF clusters were either lightly labeled or completely free of label. Cells labeled with CF prior to Con A treatment and fixation displayed extensive microvilli formation, randomly distributed clusters of CF on the cell surface and some internalization of ferritin particles. Cells fixed before Con A and CF treatment or cells incubated with Con A and α-methyl-D-mannoside were labeled with a continuous, even layer of ferritin particles. The clustered configuration of the CF particles in the Con A-CF treated hepatoma, is probably an indication of the greater relative mobility of Con A receptors in transformed cells and suggests that Con A and CF receptors may be on the same molecule. (Supported by NIH Research Grant CA-16663 from the USPHS).

SURFACE FEATURES OF AS30D IN VIVO AND IN VITRO. K. E. Savage and P. S. Baur, Department of Human Biological Chemistry, Genetics and Cell Biology, Galveston, Texas.

Cell surface alterations have been associated with the transformation of normal cells. Transformed cells may possess extensive surface elaborations such as microvilli, blebs, and ruffles. In addition, glycocalyx and membrane components have been reported to be altered in various tumor model systems. Many of these studied have compared transformed cells with their normal counterparts either **in vivo** or **in vitro**. We have studied the surface features and glycocalyx of clones of a chemically induced tumor in rats, AS30D. The clones were selected on the basis of their sensitivity to several cancer therapy drugs and were surveyed under **in vitro** and **in vivo** conditions. The cell surface features were determined via scanning electron microscopy. The glycocalyces were stained with ruthenium red (RR) and visualized via transmission electron microscopy.

Our results indicate great similarity in the topology and RR staining of the glycocalyx of all AS30D lines examined from **in vivo** ascites tumors. Cells were rounded with many microvilli. **In vitro** these cells exhibited clonal differences with respect to topology ranging from primarily microvilli to predominantely

bleb bearing and from rounded to irregularly shaped. The cell coats of these cells stained much more densely with RR than did those of the ascites cells. These results reflect differences in surface features and glycocalyces of AS30D tumor cells cultured **in vitro** with respect to their in vivo counterparts.

This finding is of particular interest because the cell surface components appear to be the medium by which cells communicate with each other and their environments. Additionally, these results may have impact on other on going tumor studies employing **in vitro** culture methods. Supported by American Cancer Society Institutional Research Grant IN 112A and National Research Service Award 07204 from General Medical Services Awarding Unit.

MORPHOGENESIS OF MEMBRANE RIBOSOMAL LAMELLAE IN HAIRY CELL LEUKEMIA. Milka M. Monteil, MD, and Barbara Boehm, The Department of Pathology, The University of Texas Health Science Center, San Antonio, Texas.

Intracytoplasmic membrane-ribosomal lamellae are seen characteristically in "hairy" cell leukemia but have also been observed in chronic lymphocytic leukemia, lymphosarcoma cell leukemia, monocytic leukemia and even in adrinal adenoma. While their ribosomal content has been proved by ribonuclease treatment, the origin of these inclusions is still obscure. This study consists of serial sections of conventional epon embedded particles of bone marrows and spleens from four patients manifesting clinical and morphologic picture of hairy cell leukemia. The sections were studied by transmission electron microscopy. The leukemic cells contained single and double layered ring-like inclusions of RER as well as classic multi-layered ribosomal inclusions. Transition of RER into these lamellar inclusions will be demonstrated as well as graphic illustration of the concept of their morphogenesis.

SCANNING ELECTRON MICROSCOPIC OBSERVATIONS ON THE MECHANISM OF SOMATIC CELL FUSION USING POLYETHYLENE GLYCOL. Mike A. Clark and Jerry W. Shay, Department of Cell Biology, The University of Texas Health Science Center, Dallas, Texas 75235.

The scanning electron microscope (SEM) was used to observe the morphological events of somatic cell fusion induced by exposure to various molecular weights of polyethylene glycol (PEG). In addition, the effects of dimethyl sulfoxide (DMSO), polyehtyline amine, colchicine, and cytochalasin B were investigated to help determine the mechanisms of PEG induced fusion. The surfaces of L-929 cells are covered by microvilli, blebs and marginal ruffles, but after treatment for one minute with either PEG 1,000 or 6,000, there is a dramatic reduction in the number of microvilli and an increase in the appearance of many small blebs. The addition of DMSO to the PEG solution immediately denudes the cell surface of microvilli and causes an increase in fusion efficiency. The slower rate of fusion produced by PEG 400 allows more detailed observation of the series of events associated with the process of fusion, and these experiments suggest that cell fusion is a three step process. The first step occurs after approximately five minutes of recovery and is characterized by the random appearance of small cytoplasmic bridges. Polyethylene amine, a fusogen analogue, did not induce cytoplasmic bridging or subsequent hybrid formation. The second step follows after 80 minutes of recovery and is characterized by blebs appearing on the cells as they actively move together. The second step is sensitive to cytochalasin B but is unaffected by colchicine. After approximately two hours of recovery from PEG treatment, the cell surfaces become repopulated by their usual distinguishing features with the cells now containing more than one nucleus. These studies can provide information on how cells fuse in the presence of PEG and may also be useful in our ultimate understanding of such phenomena as how myoblasts fuse **in vivo** to form myotubes.

REPLICATING CHROMATIN: ELECTRON MICROSCOPIC VISUALIZATION. G. B. Howze, Department of Biology, Texas Southern University, Houston, Texas 77004.

"S" phase nuclei of 8 cell stage **Arbacia** embryo and "S" phase CHO cell nuclei were studied. The nuclei were lysed, deposited on carbon coated grids, stained, and viewed at high magnification on either the Zeiss EM 10 or the Siemens 1A microscopes. The most significant finding is the presence of loop configurations in the otherwise linear unbranched chromatin fibers. The chromatin loops resemble the "eye forms" visualized in electron micrographs of replicating DNA. The arms of the chromatin loops are asymmetric. One arm is shorter and thicker than the other. The longer thinner arm usually has associated with it, two large electron dense particles. The asymmetry of the arms of a loop is tentatively interpreted as being due to early, prenucleosome, compacting of newly synthesized DNA. The chromatin fibers also contain nucleosomes. Nucleosomes were not, however, detected near the loops. In general, fewer nucleosomes have been detected in "S" phase chromatin. This scarcity of nucleosomes might be due to interaction of the preparative procedure with an altered state of the nucleosome which makes it less stable during "S", resulting in an apparent scarcity of nucleosomes.

MORPHOLOGIC AND HISTOLOGIC FEATURES OF INFECTIOUS BURSAL DISEASE — AN IMMUNO SUPPRESSIVE DISEASE OF COMMERCIAL CHICKENS. Danny Millar, Texas A&M University, Department of Veterinary Microbiology.

Infectious bursal disease (IBD) of chickens is a unique viral infection causing destruction of the bursa of Fabricius (BF) and probably other lymphoid organs. Bursa of Fabricius in avian species is an organ responsible for seeding competent antibody producing (B-type) lymphocytes into the system. Therefore, destruction of this organ by IBD virus results in immune suppression. In this preliminary study morphological and histopathological changes in the BF were studied on a sequential basis by histological examination and scanning electron microscopy following oral innoculation of day old SPAFAS chicks with the disease agent. Both histologic and SEM observations showed progressive destruction of the lymphoid follicles. The lymphocyte depletion, demonstrated by histological examination, coincides with the "inverting" effect on the follicles as seen with SEM. Ninety-six hours post innoculation obvious errosions are evident on the epithelial surface of the BF. Lymphocytes isolated from the effected bursa by trypsinization followed by purification by Ficoll-Hypaque gradient also showed noticable changes in surface morphology. These observations and other morphological features of IBD will be presented.

BACTERIAL ENDOSYMBIOSIS IN THE CAECUM OF THE SOUTHERN GREEN STINK BUG (NEZARA VIRIDULA L.). Robert P. West and A. D. Larson, Departments of Entomology and Microbiology, L.S.U., Baton Rouge, Louisana 70803.

To better understand the biology of the southern green stink bug, a major pest of soybeans in Louisiana, a study is being conducted to investigate possible endosymbiotic relationships within this species. Primary emphasis is being placed on the caecum, a region of the digestive tract previously associated with microbial endosymbiosis in insects. Caeca were removed from stink bugs, processed, and examined in the SEM. Initially, a

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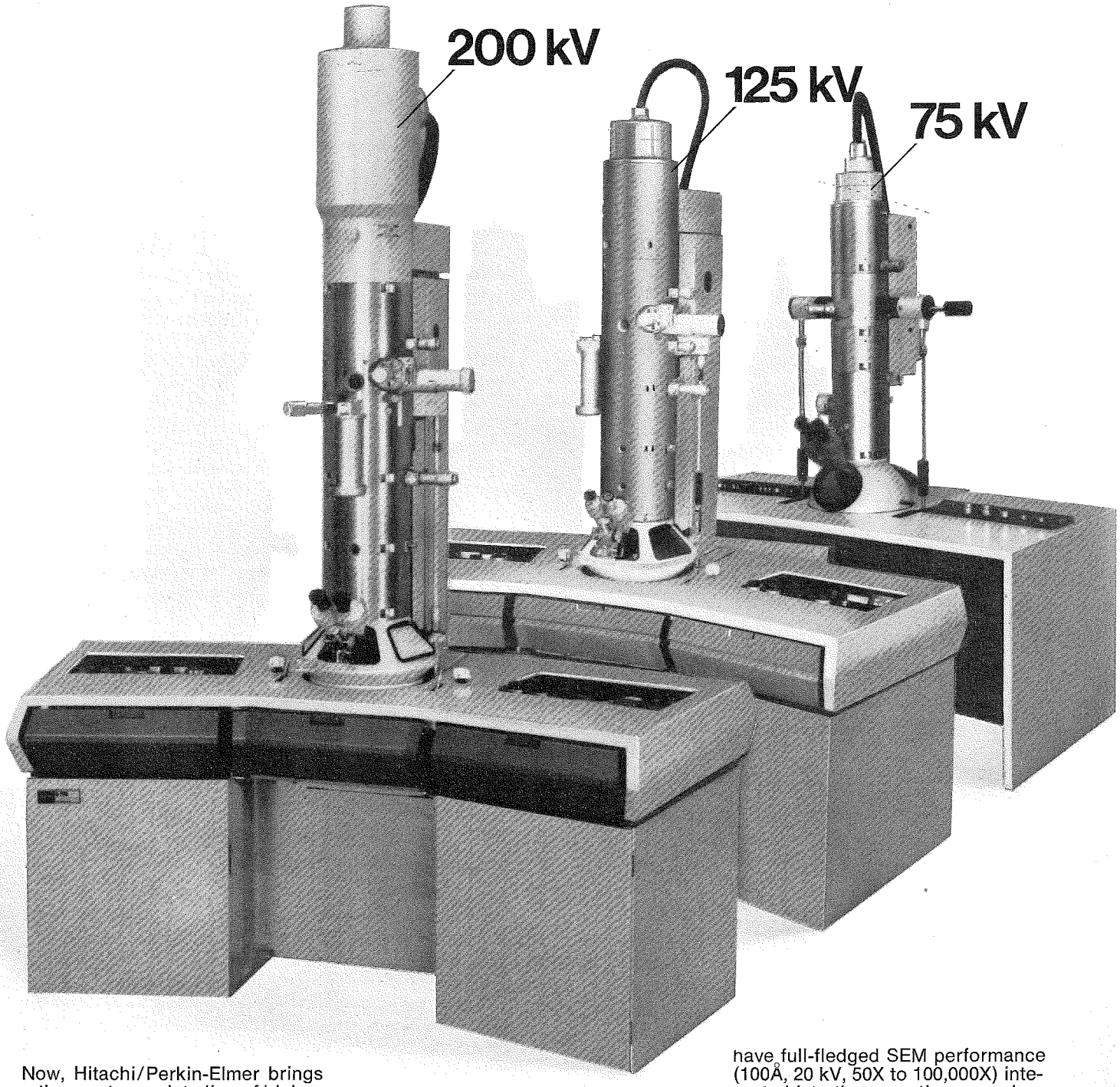

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monoculture of rod-like bacteria measuring 2.5μ x .5μ were observed adhering to the caecal crypt walls. Examination of additional specimens revealed various morphological variations, often found within the same specimen. Subsequently two distinct bacterial colonies were isolated from the caecum. These were injected into rabbits to produce antibodies which were labelled with polystyrene latex beads. The resulting immuno-latex complexes were used to confirm that the caecal bacteria had been isolated and to visually localize the two bacterial species within the caecum using the SEM.

A SCANNING ELECTRON MICROSCOPIC STUDY OF PERIPHYTIC MICROORGANISMS. Thomas M. Dreier, Electron Microscopy Center, Department of Biology, Texas A&M University, College Station, Texas 77843.

The investigation of bacterial populations in their natural environment has been hindered by their small dimensions. Scanning electron microscopy (SEM) was used to study the succession and role of periphytic microorganisms on plastic strips that were suspended vertically in a farm pond. These communities were viewed with SEM because sample preparation does not disturb the organisms attached to the artificial substrate. The initial pioneer community, one day, was composed of small colonies of rod shaped bacteria (2-4 cell/colony) and bacterial attachment was by mucoid holdfast material. After three days, colony size increased (9-12 cell/colony) and detritus commenced to accumulate in the bacterial colonies. The mucoid material responsible for holding the bacteria in place appeared to be associated with the formation of detritus platelets composed primarily of clay particles and broken diatom frustules. During the pioneer phase (1 to 9 days), the bacteria exhibit overdispersion on the plastic strips. The community structure shifted from predominantly rod shaped bacteria to phytoplankton between the 9th and 11th day. Detritus platelets were the site of attachment of stalked diatoms and filamentous algae. The diversity of the microorganisms increased during the second stage of development and the bacteria became associated with plant surfaces rather than the plastic surface. Results of this study indicate that bacteria are the initial invaders of the described habitat. In the second stage of succession, the diversity of microorganisms increased and it is hypothesized that they function as decomposers.

STUDIES ON THE EYES OF BIGEYES (TELEOSTEI: PRIANCANTHIDAE) WITH SPECIAL REFERENCE TO THE TAPETUM LUCIDUM. R. T. Wang, J. A. C. Nicol, E. L. Thurston, and M. McCants, Department of Zoology, The University of Texas at Austin 78712, and Department of Biology, Electron Microscopy Center, Texas A&M University, College Station, Texas 77843.

Eyes of priacanthids (glasseyes, Priacanthidae) have conspicuous eyeshine and possess a brilliant tapetum in the chorioid. The tapetum occupies the entire fundus; it is composed of several rows of reflecting cells which contain stacks of flat crystals lying parallel to the retinal surface in the central fundus, and obliquely towards the periphery. Reflection is orange and specular; the reflection spectrum is a broad band centered at about 630 nm. Processes of the pigment epithelium contain black pigment in some parts of the eye, especially dense in a horizontal central band, and in the lower field; pigment is absent from the cell bases. The ultrastructure of the pigment epithelium and of the tapetum is described. The crystals and intervening cytoplasmic lamellae are organized as quarter wavelength films to give maximal reflection of long wavelengths (orange and red light) by constructive interference. Rods and cones are present; there is not retinomotor activity. The mechanism of reflection, efficiency of the tapetum and role of the retinal pigment are discussed.

SURFACE MORPHOLOGY OF THE GILL FILAMENTS OF THE MULLET, MUGIL CEPHALUS. Fred E. Hossler and John R. Ruby, Department of Anatomy, LSU, New Orleans, Louisiana, and Thomas McIlwain, Gulf Coast Research Lab., Ocean Springs, Mississippi.

In preparation for studies of the effects of salt water-fresh water adaptations on mullet gill, we have conducted a scanning electron microscopic investigation of normal gill filament surface morphology. Micrographs reveal that each gill arch supports two parallel rows of gill filaments-the inner row being about 1-1/4 times as long as the outer row. Secondary lamellae (respiratory leaflets) project from the lateral surfaces of each filament along its complete length. Pavement cells (~4x7 μ) with obvious, concentrically arranged surface ridges (~0.1 μ in. width) cover the entire surface of the filaments in a mozaic pattern. Pavement cells on the secondary lamellae lack the surface ridges. On the inner and lateral surfaces of both rows of filaments numerous pores can be observed opening along the pavement cell borders. Two types of pores can be distinguished: the first is a rounded hold (0.6-0.8 μ in diameter) with small microvillous projections occasionally visable in its interior; the seond is irregular in shape (2-5 μ in diameter) and contains numerous rounded projections protruding from its opening. These may represent pores from chloride cells and mucous cells.

BRAIN STEM LESIONS IN THE SPONTANEOUSLY HYPERTENSIVE RAT. P. M. Klara, H. Kawamura, C. Knox and E. Frohlich, Tulane University, Department of Anatomy and the Ochsner Foundation.

The spontaneously hypertensive rat (SHR) is the best model of essential hypertension currently available for investigation. Preliminary studies of brain stem centers known to be involved in blood pressure regulation revealed the presence of neuronal lesions in the SHR strain. Similar lesions could not be demonstrated in the normotensive Wistar-Kyoto (WKY) rat of the same age. Accordingly, a light and electron microscopic study was undertaken in order to determine: (1) At what age these lesions first appear; (2) What areas of the brainstem are involved; (3) What is the ultrastructural morphology of the lesions; and (4) Could the lesions be involved in the eliology of the hypertensive state. Results indicate that the lesions first appear at approximately age 4-5 months; (2) Lesions are limited to the caudal portion of the medulla in the area of the nucleus gracilis and cuneatus; (3) The ultrastructural morphology of these lesions resemble fibrinoid degeneration; (4) Since the lesions develop after the onset of hypertension at age 6 weeks it appears that the lesions may be the result of the hypertensive state. However, ultrastructural studies revealed changes in the morphology of the area postrema; a structure thought to be involved with blood pressure control. Altered morphology was evident as early as two days after birth. These findgings may indicate early disfunction of neural blood pressure control centers. The possibility of such alterations and their role in the etrology of hypertension will be discussed with regard to the present findings.

Acknowledgements: This work was funded by a grant from the Ochsner Foundation. Special thanks also to Dr. R. D. Yates who provided facilities and encouragement.

PERMEABILITY CHANGES IN ARTERIOLES, VENULES AND CAPILLARIES FOLLOWING EXPERIMENTAL CEREBRAL ANGIOGRAPHY. R. L. Casady, G. T. Kitten and P. R. Sterrett, Department of Anatomy, Texas Tech University School of Medicine, Lubbock, Texas 79409.

Various neurological sequelae such as paresthesias, weakness, transient blindness and nausea may accompany cerebral angiography. The injected contrast media causes a breakdown in the blood-brain barrier, and the resulting edema and/or chemical toxicity of the contrast media produces a neurological problem. The object of this study was to determine both the type of vessel affected and the nature of the blood-brain barrier breakdown. Seventeen rabbits underwent experimental cerebral angiographies, via the left internal carotid artery, with methyglucamine iothalamate (Conray 60, Mallinckrodt) or methylglucamine diatrizoate (Reno-M-60, Squibb). Animals were killed at 1.5, 5, 30, or 60 minutes after injection of contrast media. After perfusion fixation with aldehydes (modified Karnovsky), the brain were removed, sliced and processed for light and electron microscopy. Six brain areas were selected for study: cerebral cortex, thalamus, cerebellum, dorsal hippocampus, caudate nucleus and hypothalamus. Arterioles (and very small arteries) comprised 59% of the total leaking vessels (horseradish peroxidase was the tracer); venules comprised 28%; and capillaries comprised 13%. Large amounts of leakage occurred in some of the 1.5 minute group. This suggested opening of the normally tight, impermeable intercellular junctions since intracellular passage (vesicles or otherwise) would be too slow to account for such quantities so quickly.

This research was supported by NIH Grant NS 11883 and General Research Support Grant 22-A027-400000.

ADENOSINE TRIPHOSPHATASE AND ALKALINE PHOSPHATASE LOCALIZATION IN THE CEREBRAL VASCULATURE OF SPONTANEOUSLY HYPERTENSIVE RATS. C. A. Knox[1], P. M. Klara[1], R. D. Yates[1], and E. Frohlich[2], Department of Anatomy, Tulane Medical School[1] and the Ochsner Foundation Hospital[2], New Orleans, Louisiana.

Increased vascular permeability to exogenous dyes and proteins during acute and chronic hypertension is well documented. However, a few studies have demonstrated the ultrastructural localization of enzymatic activity in the blood vessels of hypertensive animals. We have investigated the distribution of two enzymes found in the endothelial lining of the cerebral cortical vasculature in adult normotensive, Wistar-Kyoto (WKY) and spontaneously hypertensive (SHR) rats with the electron microscope. These enzymes, adenosine triphosphate (ATPase) and alkaline phosphatase (AkPase), have been hypothesized to have roles in transport phenomena. ATPase (Wachstein-Meisel technique) was localized in the adluminal endothelial plasmalemma and in the basement membrane; the density of reaction product in the basement membrane made it difficult to assess activity in the adluminal plasmalemma. Pinocytotic vesicles rarely contained reaction product. A light increase in density of reaction product at the adluminal surface was observed in some SHR as compared to WKY. No other change in distribution or density of ATPase activity was observed in hypertensive animals. AkPase activity was localized in both endothelial plasmalemmae and in numerous pinocytotic vesicles. Slight amounts of reaction product were observed with the basement membrane. No change was observed in distribution or density of AkPase activity in the hypertensive animals. ACKNOWLEDGEMENTS: This research was funded by a grant from the American Heart Association, Louisiana, Inc. Special thanks to Drs. I. Chen, L. Wade and H. Kawamura and Mr. J. Mascorro for facilities and helpful discussions.

ULTRASTRUCTURAL CHANGES IN MYOCARDIUM OF DOGS FOLLOWING SUPRATHRESHOLD TRANSCHEST DEFIBRILLATING ELECTRIC SHOCK. Joyce S. Davis, MD, Hilton H. Mollenhauer, PhD, Texas A&M University College of Medicine; J. T. Lie, MD, Mayo Medical School; W. A. Tacker, MD, PhD, Purdue University.

Transchest electrical cardiac defibrillation, often life-saving in patients with cardiac arrhythmias, may, under certain circumstances, cause damage to myocardial fibers. In dogs, the administration of transthoracic current and energy greater than twice the threshold defibrillating dose of one ampere per kilogram of body weight is associated with a characeristic pattern of myocardial necrosis, the extent of which is directly related to the "dose" of the shock. Lesions range from small subepicardial foci of myocardial necrosis to transmural necrosis in a dog that received 10 times the threshold defibrillating dose. Ultrastructural cellular changes such as disruption of myofilaments, z-bands aligned out of register or compressed into tranverse "contraction bands," dissolution of z-bands, disruption of mitochondrial christae and mitochondria containing numerous large electron dense particles stop abruptly at the intercalated disc. Separation of cell membranes from other cellular constituents is seen. Early lesions are devoid of inflammatory cells. By the fourth day abundant pale amorphous material and moderate numbers of mononuclear phagocytes and spindle-shaped cells are seen in the interstitial spaces. By light microscopy, myocardial fibers show coagulation necrosis, followed by evidence of healing. In one animal there was extensive distrophic calcification of the damaged fibers. These lesions are non-specific and have been seen in a variety of human and experimental conditions. Reichenback and Benditt suggest that a common pathogenetic mechanism may be catacholamine release from sympathetic nerve endings in the myocardium or from the adrenal gland causing increased work or uncoupling oxidative phosphorylation.

ULTRASTRUCTURAL STUDIES OF CARDIAC MESENCHYME CULTURED ON COLLAGEN SUBSTRATES. D. H. Bernanke, R. R. Markwald, J. M. Krook, Department of Anatomy, Texas Tech University School of Medicine, Lubbock, Texas 79409.

Explanting microdissected valvular primordia (endocardial cushions) from 96-hour embryonic chick hearts onto glass (2-dimensional) or collagenous (3-dimensional) substrates resulted in outgrowth of fibroblastic cells. Tissues were placed onto gels of repolymerized collagen or onto glass coverslips in 35mm Falcon dishes containing Medium 199 with HEPES buffer and 10% fetal calf serum. Collagen was extracted from isolated rat tail tendons with acetic acid and dialyzed against 1:10 diluted Medium 199 at pH 4.0. The gels were repolymerized by mixing the collagen extract with NaOH and 10x Medium 199 to obtain a neutral isotonic solution. Gels of varying density were formulated by diluting the collagen extract with water prior to adjusting pH and salt concentration. The cultures were washed with buffered salt solution, fixed with glutaraldehyde, postfixed in osmium, dehydrated in alcohols and critical-point dried for SEM or embedded in Epon for TEM. This explanting method provided a homogenous population of cells, which, when grown on collagen, more closely resembled the **in situ** cushion tissue cells than those grown 2-dimensionally on glass. The cells migrated into the 3-dimensional substrate in proportion to the density of the gel. It was believed that the collagen gels provided a microenvironment for the cells more akin to that found **in situ**. The cells were tentatively identified as cushion tissue cells by morphological criteria. The culture methods were compared for influences on biochemical function of the cells. The collagen substrates are being tested for compatability with other cell types and suitability for studies of development, motility, or biochemical function. Supported by Texas Heart Association grant.

CELL: MATRIX INTERACTION DURING CUSHION TISSUE CELL (CTC) IN VIVO MIGRATION. R. R. Markwald, J. M. Krook, Department of Anatomy, Texas Tech University School of Medicine, Lubbock, Texas 79409.

Formation of chick and rat cardiac valvular and septal tissue involves an epithelial/mesenchymal transformation with subsequent migration of "seeded" CTC through an expansive extracellular matrix (ECM) rich in glycosaminoglycans (GAG), glycoprotein (GP) and collagen microfibrils. The study objective was to determine, through scanning, transmission and high-voltage EM and **in vivo** techniques, if one ECM component more than any other physically interacted with motility processes. Premigratory matrix (stage 19, chick; day 11.0 rat) consisted of randomly dispersed collagenous microfibrils upon which was superimposed GAG-GP matrix preserved by adding cetylpyridinium chloride to the fixative. Newly-seeded CTC (stage 20, day 11.5) initially probed the ECM by cork-screw shaped filopodia which correlated temporally with coalescence of microfibrils into matrical "tracks." Pioneering CTC of sequentially-aged embryos migrated centrifugally along the microfibrillar tracks. Association of CTC filopodia with the tracks was characterized by subplasmalemmal modifications. The GAG-GP matrical component (CPC1-dependent) enshrouded motility processes without forming physical attachments. Pioneering CTC examined **in vivo** by Nomarski optics were attenuated in profile and had no evidence of "ruffled" membranes characteristic of **in vitro** cell locomotion. Treatment in organ cultures (chick-stage 21-23) with 100mg/ml collagenase or intraperitoneal injection (rat, day 10.5) of 75mg/kg BAPN resulted in the aggregation of pioneering CTC. Results suggests CTC movement engenders new matrical ordering involving collagenous "tracks" that subsequently served as a migratory template. Supported by NIH grants HL 19136, HL 02028.

THE CYTOLOGY OF CANINE PANCREATIC ACINAR CELLS FOLLOWING HYPOPHYSECTOMY. A. H. Crenshaw, Jr. and W. A. Shannon, Jr., Veterans Administration Hospital and Department of Cell Biology, Southwestern Medical School, Dallas, Texas.

Specimens of exocrine pancreas were studied from hypophysectomized dogs after postoperative periods of 4 to 300 days. Normal dogs or sham hypophysectomized dogs served as controls. Zymogen and prezymogen granules, seen especially in the apical region of acinar cells in the controls, were few or missing in the experimental animals. In addition, the zymogen material present appeared atypical in density and shape. A considerable number of the acinar cell population appeared electron dense very often with pyknotic nuclei and dense bodies. Some cells, especially in the longer hypophysectomized animals, showed extreme cisternal dilations of the rough endoplasmic reticulum as well as a loss of its normal organization. Nuclei in these cells were very pyknotic. Some cells exhibited connective tissue infiltration and the basal laminae appeared thickened in many cells. Most of both light and dark acinar cells contained greater number of lysosomes, lipofuscin granules, and large dense bodies than present in the controls. The granular endoplasmic reticulum frequently appeared in extensive arrays often as parallel lamellae exhibiting loose and swollen cisternae.

PANCREATIC DUCT LIGATION AND RESULTING FINE STRUCTURAL ALTERATIONS IN THE CANINE PANCREAS. A. H. Crenshaw, Jr., W. J. Brown, G. Patton, R. E. Dobbs, and W. A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Physiology, Southwestern Medical School, Dallas, Texas.

Studies on canine pancreas following seven months ligation of both pancreatic ducts and any accessory ducts present indicated gross changes in the fine structural morphology of the exocrine pancreas. The pancreas remnant appeared devoid of all acinar cells, in effect representating an endocrine organ. Rare dense cell remnants with pyknotic nuclei were thought to represent degraded acinar cells. The entire remnant was profusedly infiltrated by vasculature and thick layers of connective tissue. A relatively normal population of endocrine cells remained. Some of the A and B cells appeared partially degranulated relative to controls. Other cells thought to be of an endocrine nature but indistinguishable, due to a paucity of cytoplasmic granules, were observed. Some apparent lipid inclusions as well as multivesicular bodies, especially in A and B cells, were also present. The physical and numerical relationship of the cells to each other and to the underlying vasculature appeared normal.

ABNORMAL STRUCTURES IN LYMPHOCYTES FROM PATIENTS WITH OCULOCRANIOSOMATIC NEUROMUSCULAR DISEASE. W. J. Brown, J. D. Cook, and W. A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Neurology, Southwestern Medical School, Dallas, Texas.

The presence of mitochondrial paracrystalline inclusions in skeletal muscle and other tissues from a number of myopathic conditions suggests a generalized systemic disorder. A preliminary ultrastructural study of the circulating leukocytes from three patients with oculocraniosomatic neuromuscular disease with ragged-red fibers indicates the presence of virus-like and tubuloreticular structures in the lymphocytes. The virus-like structures, found within large cytoplasmic vacuoles, consist of spherical bodies 30 nm in diameter with up to twenty having been observed within a single vacuole. The tubuloreticular structures, also seen within vacuoles, consist of parallel arrays of electron dense tubules 25 nm in diameter. The electron dense portions are 7.5 nm wide and are separated by 10 nm spaces. Also, honeycomb or tubulopolygonal structures have been observed within vacuoles containing tubuloreticular structures. The dimensions of the honeycomb structures correspond with those of the tubuloreticular arrays, therefore, the honeycomb structures probably represent cross sections through a stack of parallel tubules. Based upon morphology alone, the 30 nm spherical bodies appear to be virus-like particles, although no direct evidence is available to show that virus-like particles are indeed viruses. Also, the tubuloreticular structures do not resemble the paracrystalline inclusions seen in skeletal muscle; they are probably related to the virus-like structures.

MITOCHONDRIOPATHY IN OCULOCRANIOSOMATIC NEUROMUSCULAR DISEASE. W. J. Brown, J. D. Cook and W. A. Shannon, Jr., Veterans Administration Hospital and Departments of Cell Biology and Neurology, Southwestern Medical School, Dallas, Texas.

The fine structure of skeletal muscle mitochondria in patients with oculocraniosomatic neuromuscular disease with ragged red fibers was investigated. A definite mitochondriopathy exists with apparently two distinct morphological diversities. Mitochondria within the myofibril bundles appear quite normal while clusters of mitochondria at the periphery appear bizzare. One type of mitochondria (Type I) exhibits stages in the apparent formation of paracrystalline structures while the other (Type II) exhibits a concentric lamellation of its matrix components within its outer compartment. The apparent chronology in the formation of these two types appears to be distinct. Type I begins with mitochondrial elongation and matrix

reorientation with cristae extending end to end. Paracrystalline arrays appear, eventually filling the mitochondria and finally displacing them so as to make them unrecognizable. The paracrystalline inclusions consists of four closely associated groups of four parallel linear densities. Each group is 130 nm wide and the periodicity of the parallel densities is 6 nm. Each group of four linear densities is separated by a 10 nm space. Type II forms as the matrix and cristae begin to assume a whorled pattern resulting in a lamellated structure indistinguishable from normal mitochondria.

MORPHOLOGICAL AND ULTRASTRUCTURAL CHANGES IN GLANDLESS COTTONSEED (GOSSYPIUM HIRSUTUM L.) INDUCED IN A NON-EXTRUDED TEXTURIZED VEGETABLE PROTEIN. M. V. Taranto and K. C. Rhee, Food Protein R&D Center, Texas A&M University, College Station, Texas.

An ultrastructural study of the cotyledonous tissue of the cottonseed kernel reveals that the major portion of the protein is located in the protein bodies or aleurone grains and the lipid is localized in the spherosomes. Globoids, the storage site for phosphorus and metallic ions, are embedded in the matrix of the protein bodies. While the cellular structure of the kernel is disrupted, the protein bodies remain intact during dehulling, defatting and grinding of the kernel into a flour.

Non-extrusion texturization of defatted glandless cottonseed flour was found to be time dependent. Texture was developed in a series of well defined stages. The process begins with the fusion of flour components after a two second retention time. Between two and eight seconds, the protein bodies are deformed, ruptured and fused forming the product's protein-carbohydrate matrix. The texturization process is essentially complete after a ten second retention time with the fibrillation of the protein-carbohydrate matrix.

The globoids were the only structures of the kernel which were not disrupted and fused during processing. These data may offer an explanation for the significantly higher rheological properties of textured soy protein over textured cottonseed protein.

The results of this study indicate that a specific ultrastructural reorganization of the cottonseed kernel is induced by the non-extrusion texturization process. Funded by NSF Grant ENG 76-17269.

AN ANATOMICAL ANALYSIS OF THE EPIDERMAL SURFACE ELABORATIONS OF CARYA ILLIONENSIS. Larry Grauke, Department of Horticulture, Texas A&M University.

The absorption of foliar applied nutrients is affected by leaf surface features such as wax distribution, venation, trichomes and stomata. As a preliminary step in the investigation of the mechanism of the foliar absorption of zinc in pecan **(Carya illinoensis)**, it was necessary to perform an anatomical analysis of leaf surface features as they are affected by leaf and tree age.

Three types of trichome were found: awn-like hairs, peltate scales and capitate glands. All types were found predominately on the abaxial surfaces and on the immature leaves. Awns and capitate glands were more frequent on leaves from juvenile (non-fruiting) tress. Peltate scales occurred with equal frequency on leaves from both juvenile and adult (fruiting) trees.

Stomata are confined to the abaxial surface, where they occur in two sizes. Stomata reach their maximum density and development on mature leaves. Differences in stomatal densities on leaves taken from adult and juvenile trees were insignificant.

An unusual stomate-like structure was found on the abaxial midrib of immature leaves from both juvenile and adult trees. Vestiges of the structures were found on mature leaves.

FERROMANGANESE MICROACCRETIONS FROM THE MID ATLANTIC RIDGE FAMOUS AREA. L. A. Barnard, Department of Oceanography, Texas A&M University, College Station, Texas 77843.

Ferromanganese microaccretions were hand picked from four sieved size fractions (100, 200, 325 and 400 mesh) of sediment samples from four depths in a one meter gravity core taken in the mid-Atlantic ridge Famous area. This area is one of intense hydrothermal activity with high rates of metal input. Scanning electron microscopy and energy dispersive x-ray analysis were used to relate surface morphology and qualitative metal content (Mn, Fe, Cu, Ti) to depth of burial, size fraction, and apparent sediment redox potential. Upper sediment (0-2 cm) microaggregates were typified by a well-formed pillowy appearance with large amounts of Fe, Mn and Cu. Microaggregates taken from the region of the redox boundary (42-46 cm) showed some signs of dissolution and tended to have depressed Mn-Fe ratios. Cu was present in trace amounts. Fe-Mn particles from the base of the core (86-98 cm) were strongly leached in appearance and contained either Fe or Mn in small amounts but rarely both together. Basal particles clearly showed the biogenic kernel material while all particles were rich in biogenic detritus. Well crystallized pyritihedra aggregates were found in samples taken from the base of the core (96-98 cm) as well as in samples from the redox boundary. Framboids were found to be contained within foraminifera from the redox boundary area (44-46 cm).

SEM USED TO EVALUATE FORMATION DAMAGE. D. F. Feuerbacher and M. H. Cordova, Dresser Industries, Inc.

One of the serious problems encountered in oil-well drilling or completion is damage to oil-or-gas producing formations. The scanning electron microscope has been successfully used to study three types of formation damage: drilling mud invasion, perforation damage, and damaged caused by particle migration.

All types of formation damage were studied using either actual oil-field cores or berea sandstone cores. Mud invasion damage, determined by examining a fractured mud-drilled core from the outer edge radially inward, showed minimal penetration of mud solids. Perforation damage was limited to the area near the end of the perforation, where the core was "pulverized." Damage due to particle migration was determined by examining both pressure-stressed (by fluid flow) and unstressed sections from the same core. On one case, a "filter-effect" due to particle build-up in pore spaces along the direction of flow was noticed; a simultaneous decrease in permeability to fluid flow was also found.

A prediction of possible formation damage may be made by studying the minerals adjacent to pore spaces in the core. Easily hydratable or swellable clays, such as montmorillonite or illite. or migratable particles such as kaolin or chlorite, for example, could help determine the type of drilling or completion fluids best suited to minimize permeability damage.

EVALUZATION OF THE THIN FILM ASSUMPTION IN QUANTITATIVE X-RAY ANALYSIS. By D.P. Skinner and N. C. Barbi, Princeton Gamma-Tech, Inc.

Most of the commonly used algorithms to convert X-ray intensity data to concentrations in the analysis of thin sections or small particles are based on the thin film assumption. For example, the relation $C_1/C_2 = k_{12} I_1/I_2$ is often employed to calculate the ratio of concentrations between two elements in a thin section, where k_{12} is either measured or calculated by a variety of

schemes. For this and other similar expressions to give accurate results, interelement effects on X-ray intensity must be negligible. To evaluate the thin film assumption, the absorption expected from Beer's Law can be computed. If, for example, up to 10% relative error is to be tolerated from invoking this assumption, then $I/I_0 = e^{-}X\ (\varrho z) \geq 0.9$

where $X=\mu_m \csc\gamma$; μ_m=mass absorption coefficient; γ=take-off angle; ϱz=mass thickness; ϱ=density and $z=t/2$; t=specimen thickness.

A computer program written in BASIC computes the mass thickness beyond which the thin film assumption is said to be invalid. This thickness will be different for each element in the section. Alternatively, the program can be used to input mass thickness, and output fractional X-ray intensity absorbed for each element.

ULTRASTRUCTURAL OBSERVATIONS ON THE MOTILE CELLS OF THE GREEN EPIPHYTIC ALGA PHYCOPELTIS EPIPHYTON (CHROOLEPIDACEAE.) B. H. Good and R. L. Chapman, Department of Botany, Louisiana State University, Baton Rouge, Louisiana 70803.

Phycopelitis epiphyton reproduces zsexually by the formation of zoospores in abscised, wind-borne zoosporangia and sexually by gametes formed in sessile gametangia (modified vegetative cells). Ultrastructural observations reveal that both gametes and zoospores contain multilayered structures, spline microtubules, and bilaterally keeled flagella. This is comparable to what has been observed in the related algal genera, **Cephaleuros** and **Trentepohlia.**

Early light microscopic reports indicated that both gametes and zoospores were biflagellate. However, recent ultrastructutal observations have shown that zoospores are quadriflagellate and are not biflagellate as reported by earlier workers. Also, it is suggested that the biflagellate gametes of **P. epiphyton** contain on multilayered structure while the quadriflagellate zoospores contain two multilayered structures. As has been reported, the quadriflagellate zoospores of **Cephaleuros** also contain two multilayered structures and suggests that the presence of two multilayered structures in each biflagellate motile cell of **Trentepohlia** is a particularly intriguing observations. These observations may be an important phylogenetic indicator among green algae and certain land plants.

ULTRASTRUCTURE OF SENSORY SENSILLA ON THE ANTENNAE OF SOME PARASITES IN THE CHALCIDOIDEA (INSECTA: HYMENOPTERA). Margaret R. Barlin, Department of Entomology, Texas A&M University.

Many species of Hymenoptera parasitize insect pests and are therefore of interest as biological control agents. These parasites have a complex behavioral pattern preceding mating or oviposition. Chemical and mechanical cues in the environment initiate these behavioral patterns. These cues are perceived by sensory sensilla in the antennae. Female parasites have three types of hair sensilla, one with fluted, thick cuticular walls, the second with thinner, multiporous walls, the former functions as a mechanorecptor and the latter as a chemoreceptor. In addition two or three terminal, non-porous, thick-walled hairs are present. There are two types of plate organs, one with a thin wall and numerous pores and one with a thick wall and few pores, both probably functioning as chemoreceptors. In addition, bulb-shaped sensilla with pores in a thick cuticular wall occur. Males differ from the females in having one type of plate organ, intermediate between those found on the female antenna, and a row of long hairs along the proximal edge of the basal segments. These hairs have numerous pores indicating a chemoreceptive function. As they are lacking in the female they may serve to detect the female sex pheromone. These sensory sensilla play a vital role in the complex behavior of the highly evolved parasitic Hymenoptera.

ELECTRON MICROSCOPY, AN ART FORM. Ruth E. Lewis, Electron Microscopy Center, Texas A&M University, College Station, Texas 77843.

A pictorial essay demonstrating the use of various graphic techniques on electron micrographs to produce special effects is displayed.

DIFFERENCES IN CONCENTRATION OF ELEMENTS IN BRAIN CELLS DUE TO: OPIATES, CELL TYPE AND SUBCELLULAR LOCATION. A X-RAY MICROANALYSIS STUDY. Ivan Cameron, Peter Sheridan and Nancy Smith, Department of Anatomy, The University of Texas Health Center at San Antonio, San Antonio, Texas.

Adult female Sprague Dawler rats were divided into three groups and given two daily doses of (1) morphine (25 mg/kg at each injection), or (2) methadone (5 mg/kg), or (3) saline for seven days. Two hours before the rats were killed they were given a double dose of the drugs. Fifteen minutes before being killed some of the morphine treated rats were given the opiate antagonist, naloxene (2.5 mg/kg), which caused a sudden arousal in these rats. At the time of killing the region of the hypothalmus containing the medial preoptic nucleus was rapidly removed and frozen in liquified propane to prevent translocation of elements in cells. Frozen-dried 4μm sections were electron probed in a SEM and energy dispersive x-ray spectra were collected. The characteristic peak to continuum ratio for all detectable elements was determined in both the nucleus and the cytoplasm of ependymal cells and neurons. The data from 9 cells of each type in each rat brain were then subjected to one- and three-way analysis of variance. The results show significant differences in the distribution of elements (Na, Mg, P, S, Cl, K and Ca) which are dependent upon: (1) subcellular localization, (2) cell type, and (3) particularly opiate treatment. The behavioral state produced by the opiates is directly correlated with the effects they have on intracellular concentration of several elements, most notably, sodium. (Supported by NIH grants CA16831, S01 RR05654 and DA01611)

ULTRASTRUCTURE AND CYTOCHEMISTRY OF OOGONIAL DEVELOPMENT IN ACHLYARECURVA. Ellzey, Joanne T., Elaine Huizar and Michelle Baca, Ultrastructure Laboratory, Biological Sciences, The University of Texas at El Paso.

The cleavage of the oogonium into oospheres within **Achlya recurva** has been studied utilizing Nomarski interference microscopy, transmission electron microscopy and ultrastructural cytochemistry. Meiotic nuclei containing synaptonemal and ultrastructural cyctochemistry. Meiotic nuclei containing synaptonemal complexes were localized in the peripheral cytoplasm of cleaving oogonia. The distribution of other organelles including dictyosomes and associated vesicles, endoplasmic reticulum, "fingerprint" vacuoles, lipids and mitochondria were compared to other developmental stages such as antheridia, hyphae, resting oospores and germinating oospores. Catalase, an indicator for microbodies was deteched with 3.3'-diaminobenzidine. The Barka and Anderson modification of Gomori's medium was used to detect acid phosphatase activity.

INTRACELLULAR DISTRIBUTION OF ELEMENTS IN MOUSE PANCREATIC ACINAR CELLS BY X-RAY MICROANALYSIS. T. B. Pool, I. L. Cameron and Nancy Smith, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas.

In order to determine the intracellular distribution of elements in a protein secreting tissue, 1 mm cubes of pancreas were rapidly excised from Ajax mice and frozen in liquid propane chilled to liquid nitrogen temperature. Sections (4 μm) were made at -30° prior to cryosorption at that temperature. To reduce background x-rays, the sections were suspended over a single hole in a copper EM grid using no adhesives and were analyzed at 25 kV in a JEOL JSM-35 SEM equipped with a Nuclear Semiconductor Si(Li) detector. Data were stored and processed with a Tracor Northern NS-880 pulse height analyzer system using appropriate programs and reference standards. Peak to continuum ratios were calculated from characteristic x-rays of Cl, K, P, S and Ca from nuclear, cytoplasmic and zymogen-enriched areas of acinar cells. Although Ca is uniformly distributed throughout the cells, both Cl and S are significantly higher in zymogenic regions as compared to surrounding cytoplasm or nuclei; K and P are significantly lower in zymogenic regions. All elements were seen to be uniformly distributed between nuclei and non-zymogenic areas of the cytoplasm in pancreatic acinar cells. (Supported by USPHS Grant CA16831)

A COMPARISON OF INTRACELLULAR ELEMENTS IN NORMAL AND TUMOR CELLS AS DETERMINED BY X-RAY MICROANALYSIS. Thomas B. Pool, Department of Anatomy, The University of Texas Health Science Center at San Antonio, San Antonio, Texas.

Studies have shown that, in general, cancer cells possess a lower transmembrane potential than do their normal counterparts. Further, the hypothesis has been proposed that intracellular levels of sodium and other elements are involved in the regulation of mitosis in both normal and transformed cells (Cone, 1971). Results from several cellular systems tested **in vitro** have supported this contention, however data from **in vivo** systems are lacking. We have used a scanning electron microscope equipped with an energy dispersive x-ray detector and pulse height analysis module to examine the intracellular levels of Na, K, Cl, S, Mg, and P in three transplantable tumors and the normal tissue counterparts of these tumors. The tumors studied were Morris hepatoma #7777 carried in Buffalo rats, H6 hepatoma carried in A/Jax mice, and a transitional cell bladder tumor carried in C3H-HeJ mice. Peak to continuum ratios for nuclear Na were elevated 1.76-fold in Morris hepatoma 7777 cells as compared to host rat liver; 1.78-fold in C3H bladder tumor cells as compared to normal mouse transitional cells; and 3.72-fold in H6 hepatoma cells as compared to host mouse liver cells. Additionally, K was elevated in both rat hepatoma cells and mouse bladder tumor cells whereas Cl was elevated in both rat hepatoma and mouse hepatoma cells. These data support the conclusion that elevated levels of intracellular Na are correlated with increased mitotic activity. (Supported by Grant No. CA16831 from the National Cancer Institute)

EFFECT OF CHLOROQUINE ON FETAL HEART LYSOSOMES. Robert M. Ridout, Robert S. Decker, and Kern Wildenthal, University of Texas Health Science Center at Dallas, Dallas, Texas 75235.

In order to study the effect of chloroquine on the cardiac lysosomal system **in vitro**, the drug (0.1mM) was added to the

(Continued on page 22)

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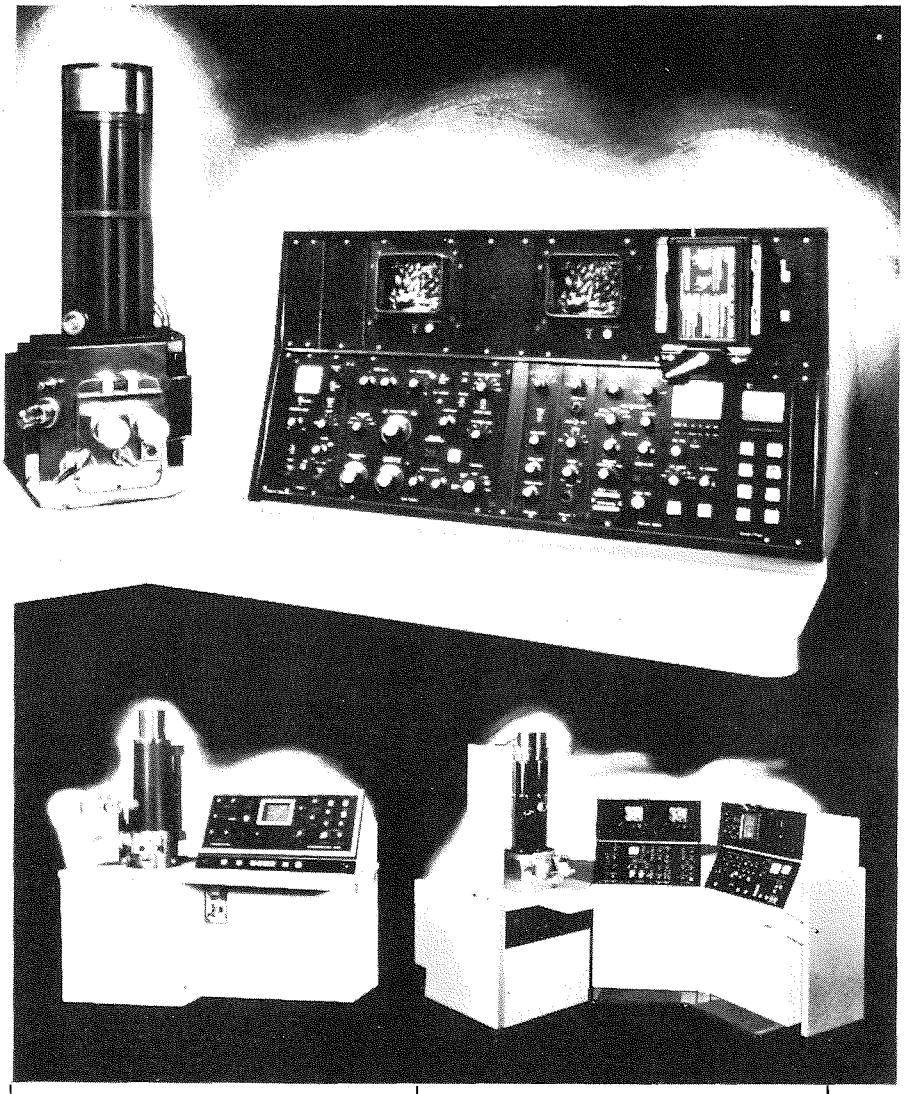

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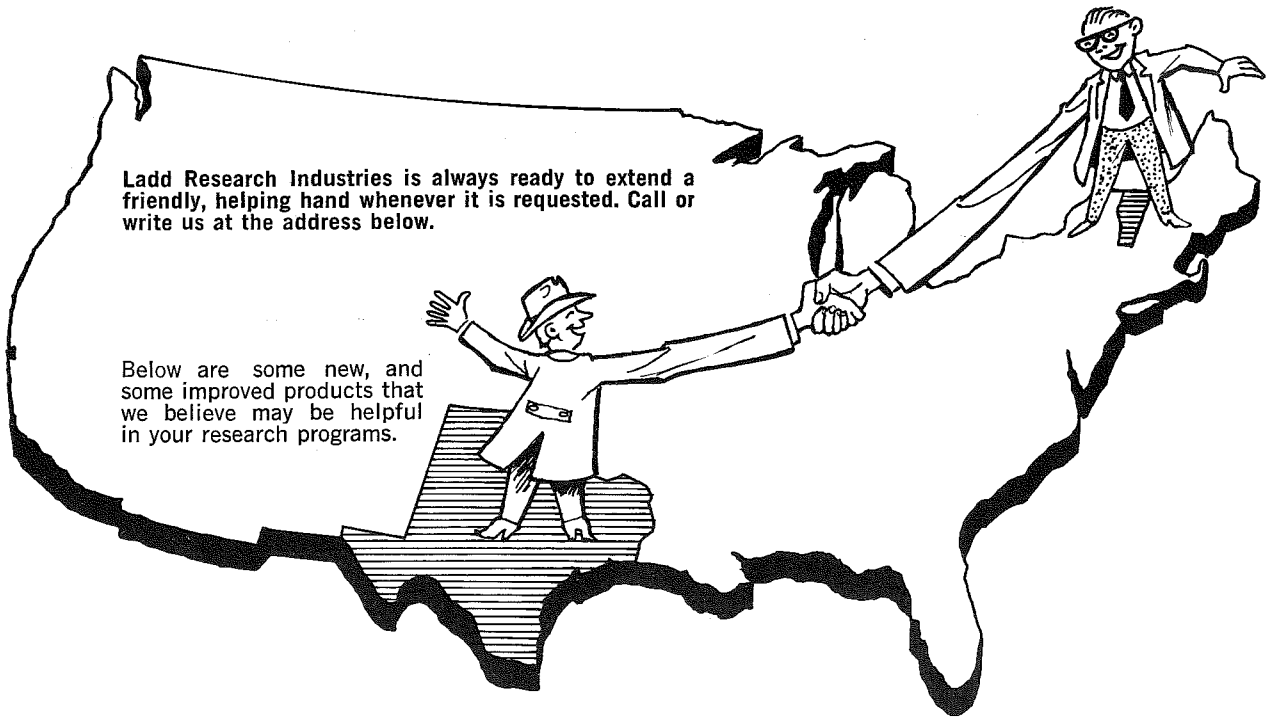

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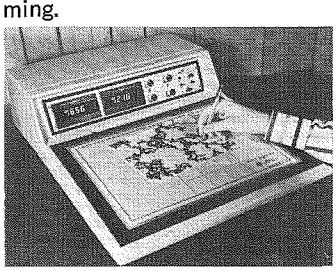

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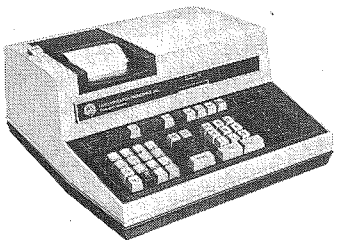

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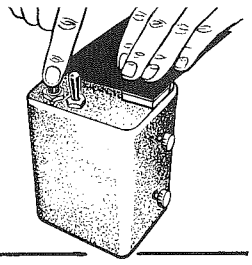

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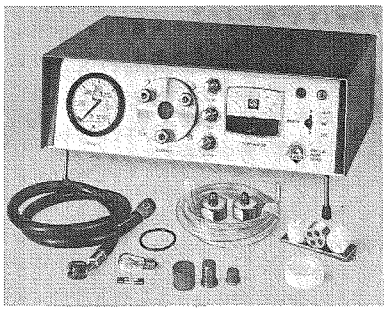

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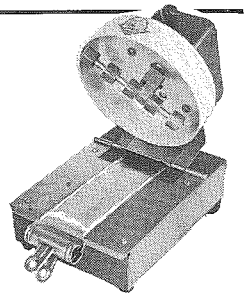

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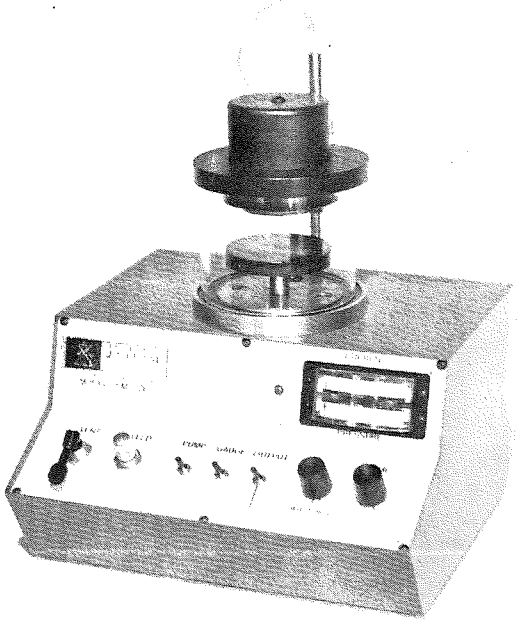

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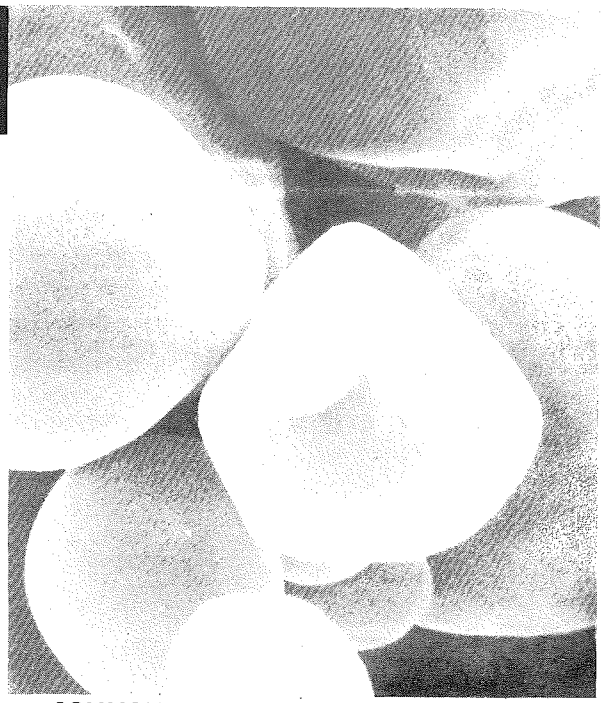

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Micrograph courtesy of PERKIN ELMER–HITACHI.

Regional News

HOUSTON: University of Texas System Cancer Center, M.D. Anderson Hospital and Tumor Institute.

Department of Laboratory Medicine

A Siemans 102 has been added to the EM Laboratory and is now operational. Mr. Ken Hannah and Mr. Mark Grogan have recently joined the Department's Ultrastructure Section. Mr. Hannah was formerly in Dr. Mims' laboratory at Stephen F. Austin State University. Dr. Ahearn presented a paper, Diagnostic Implications of Electron Microscopy in Acute Undifferentiated Leukemia, at the December Meeting of the American Society of Hematology in San Diego.

Recent Publications: Ahearn, M. Trujillo, J., Dicke, K.: Electron Microsopic Identification of Leukemic Cells in Human Bone Marrow Colonies. Proceedings of the Third International Symposium on Detection and Prevention of Cancer, H.E. Nieburgs, editor, Marcel Dekker, Inc., New York, In Press, 1977.

Ahearn, M., Trujillo, J.: Acridine Orange Staining of Euchromatin in Human Acute Leukemia Blast Cells Following PHA Stimulation. Proceedings of the American Association for Cancer Research, 8:150, 1977.

Youness, E., Ahearn, M., Drewinko, B.: Simultaneous Occurence of Non-Hodgkin's Lymphoma and Spontaneous Acute Granulocytic Leukemia. American Journal of Clinical Pathology, In Press, 1977.

Department of Molecular Carcinogenesis and Virology.

Recent Publications: "Laboratory diagnosis of immune and autoimmune reactions (Basic principles and practical application), introductory review" by J.M. Bowen and L. Dmochowski. **American Journal of Clinical Pathology.** 68(5):635-637, 1977.

"Monolayer cultures for immunofluorescent staining from tumor primary explants" by G. Seman and R.C. Nairn. **Stain Technology,** November, 1977.

"Viruses and human cancer: History and current status of ESP-1" by L. Dmochowski and J.M. Bowen, a chapter in **Prog. Exp. Tumor Research,** Vol. 22, pp. 162-197, 1978.

Accepted for publication: "Electron microscopic studies of intracisternal virus particles in Soehner-Dmochowski murine sarcoma virus-induced bone tumors of New Zealand black rats" by Y. Ohtsuki, L. Dmochowski, G. Seman, and J.M. Bowen. **Cancer Research,** March, 1978.

Presentations; Participation at Meetings: Dr. James C. Chan presented a seminar to the Carcinogenesis Study Section on September 8, 1977, entitled, "Viral carcinogenesis: A model system of retrovirus-induced bone tumors".

Dr. James M. Bowen made a presentation entitled, "An overview of viral oncogenesis" to Residents and Fellows of the Department of Medicine on September 8, 1977.

The Southwest Section of the American Association for Cancer Research meeting in New Orleans, November 3-5, 1977, was attended by Drs. J.M. Bowen, J.L. East, J.E. Knesek, G. Seman, L. Dmochowski, N.M. Kouttab, J.W. Davis, J.C. Chan, and other personnel from our department. Eight papers were presented. Also Dr. Bowen gave a lecture at this meeting entitled "Antigenic markers in human breast cancer: Are they viral?"

Dr. Gabriel Seman presented a paper entitled "Paraffin rafts for organ cultures" at the fall meeting of the Texas Branch of Tissue Culture Association, Houston, Texas, October 28, 1977.

The Texas Branch of the Tissue Culture Association held its semi-annual meeting on October 28 at the University of Texas Medical School. Several members of the department participated in a workshop entitled, "Mycoplasma Detection".

Several members of the department attended the 22nd Annual Clinical Conference on "Immunotherapy of Human Cancer" in Houston at the Shamrock Hilton, November 9-11, 1977.

Dr. Leon Dmochowski attended the annual meeting of the American Association for Laboratory Animal Science in Anaheim, California, October 3-7, 1977.

Dr. Leon Dmochowski attended the Board of Trustees meeting of the Leukemia Society of America, Gulfcoast Chapter, on September 8, 1977, and Executive Committee meetings of the chapter on October 20 and November 17, 1977.

Dr. Leon Dmochowski attended the annual meeting of the National Leukemia Society of America, Inc., in Birmingham, Alabama, October 27-29, 1977. He presented talks to the members of the Executive Committee, Patient Aid Committee, National Board of Trustees, and to the Corporate Delegates.

Special Items: Dr. James M. Bowen has been appointed a member of the Search Committee for President of the University of Texas System Cancer Center.

Dr. James M. Bowen participated in the Comprehensive Cancer Center Site Visit November 29-30, 1977. Dr. Bowen and Dr. J.C. Chan participated in the site visits for both NCI Clinical Core Grant and Environmentals Science Park Core Grant during the week of October 24-28, 1977.

Dr. L. Dmochowski attended on November 29-30 the Comprehensive Cancer Center Site Visit and also the Clinical Core Grant Site Visit on October 24-26.

Grant Award: Dr. James East was notified that his competitive renewal grant, "Relatedness of RNA Tumor Viruses and Human Neoplasia" from NCI has been funded for the 04-06 years.

Visitor: Dr. Yohei Ito, Professor and Chairman, Department of Microbiology, Faculty of Medicine, Kyoto University, Kyoto, Japan, spoke to members of the Leukemia Lymphoma Study Section and other interested personnel on November 8, 1977, on "RNA Virus Search for Virus-Related Etiological Factor(s) in Human Leukemia; RNA-Reverse Transcriptase Complex from Cultured Myeloma-Leukemia Cells". On November 9, 1977, Dr. Ito presented an informal discussion on viruses and cancer to members of the Department of Molecular Carcinogenesis and Virology.

HOUSTON: Baylor College of Medicine, Department of Medicine, Section of Cardiovascular Sciences.

Presentations: Dr. Ann Goldstein gave a seminar to the research group of the Jerry Lewis Neuromuscular Disease Center in October.

Dr. Barry VanWinkle gave a seminar in the Department of Medicine at Baylor in November.

News Brief: We have recently purchased a rotation tilt stage for our Philips 201 EM.

Publications: Goldstein, Margaret A., Schroeter, John P. and Sass, Ronald L. Optical Diffraction of the Z Lattice in Canine Cardiac Muscle. J. Cell Biol, 75, 1977 (Dec.).

HOUSTON: The University of Texas Health Science Center at Houston.

Department of Anatomy. The following faculty members of the Department of Neurobiology and Anatomy, University of Texas Medical School at Houton attended the 7th Annual Meet-

ing of the Society for Neuroscience in Anaheim, California, November 6-10, 1977:

Dr. S.J. Enna chaired a session entitled "Receptors: GABA, Peptides". He is author of the following papers that were also presented: "Neurotransmitter Receptor Alterations in Parkinson's Disease", "Ontogenesis of Calcium-Dependent GABA Release in Rat Brain" and "Specific ^{3}H-Muscimol Binding to Synaptic GABA Receptors".

Dr. Yvonne C. Clement-Cormier present a pper entitled "Dopamine-Sensitive Adenylate Cyclase in Retina: Subcellular Distribution" and "Cyclic Nucleotides in Neuroendocrine Function: The Role of Dophamine and Chlorpromazine".

Dr. N. Dafny presented his paper entitle "Investigation of the Cholinergic Pathway in the Nucleus Accumbens".

Dr. Dianna A. Redburn presented a paper entitled "Otogenesis of Calcium-Dependent GABA Release in Rat Brain". Dr. Redburn is author of the following papers which were also presented: "Dopamine-Sensitive Adenylate Cyclase in Retina: Subcellular Distribution" and "Uptake of Neurotransmitters by Synaptosomal Fractions from Retina".

Dr. Richard Wiggins presented a paper entitled "CNS Teratology of Halothane".

Dr. Jon DeFrance presented his paper entitled "Investigation of the Cholinergic Pathway in the Nucleus Accumbens".

Dr. Joe G. Wood, Chairman of the Department of Neurobiology and Anatomy and Dr. W.G. Dail, University of New Mexico School of Medicine, Albuquerque presented a paper entitled "Studies on the Possibility of an Extra-ganglionic Source of Adrenergic Terminals to the Superior Cervical Ganglion". Dr. Wood also presented papers entitled "Analytical Microscopy of Amino Positive Structures in Primitive Substantia Nigra Neurons" and "Alternate Section Histofluorescence/Cytochemistry of Identified Aminergic Neurons".

Cleatus Wallis visited the department October 6-7, 1977. Dr. Wallis is from The University of California, Los Angeles, Department of Anatomy. She presented a seminar entitled "Neuroendocrine Influences on Gamma Aminobutyric Acid Metabolism in Rodent Brain Tissue".

Dr. Gerald P. Kozlowski visited the department December 1, 1977. Dr. Kozlowski is from Colorado State University, Department of Anatomy. He presented a seminar entitled "Brain pathways which secrete hormones.".

Dr. Robert B. Chronister visited the department December 2, 1977. Dr. Chronister is from the University of Southern Alabama, Mobile, Department of Anatomy. He presented a seminar entitled "Morphological Analysis of A10 Relationships".

Margaret E. Bell, M.S. from the University of Texas, Graduate School of Biomedical Sciences, Houston, presented a special neuroscience seminar October 20, 1977, which was sponsored by the Department of Neurobiology and Anatomy. Ms. Bell's seminar was entitled "Myelin Synthesis in Sciatic Never Dysfunction".

Ronna C. Kaplan, A.A., B.A., has joined the Department as the new Administrative Assistant. Ms. Kaplan was previously employed as the Administrative Assistant at Northwestern Memorial Hospital, Chicago, Illinois, in the Department of Surgery.

The Department of Neurobiology and Anatomy has recently purchased a JEOL 100C electron microscope with a Kevex energy dispersive X-ray detector plus a JEOL 100B electron microscope.

TEXAS A & M

Presented Papers: W.M. Kemp, S.C. Merritt, and J.G. Rosier, Department of Biology, "Evidence for membrane modulation in Schistososmiasis" at the American Society of Tropical Medicine and Hygiene, Nov. 11, Denver, Colo.

S.L. White and D.F. Bay, Department of Entomology, "Olfactory sensilla of the horn fly antenna" at the Entomological Society of America, Nov. 27, Washington, D.C.

D.E. Bay and S.L. White, Department of Entomology, "Sense organs on the ovipositor of the horn fly" at the Entomological Society of America, Nov. 27, Washington, D.C.

Edson, K.M., Department of Entomology, "The poison glands in adult braconids: a comparative morphology" at the Entomological Society of America, Nov. 27, Washington, D.C.

Petralia, R.S., Department of Entomolgy, "Developmental morphology and feeding behavior of larvae of the imported fire ant, **Solenopsis invicta** Buren" at the Entomological Society of America, Nov. 27, Washington, D.C.

Phillips, S.A. Jr. Department of Entomology, "Physiology of the post-pharyngeal gland of the imported fire ant, **Solenopsis invicta**" at the Entomological Society of America, Nov. 27, Washington, D.C.

Barlin, M.R. Department of Entomology, "Ultrastructure of sensory receptors of the antennae, ovipositor, and maxillary palps of a cockroach-egg parasite" at the Entomological Society of America, Nov. 27, Washington, D.C.

Droste, T.E. Department of Entomology, "The morphology of the ventral eversible (defense?) gland of walnut caterpillar **(Lepidoptera notodontidae)**: SEM, TEM, and light microscopy" at the Entomological Society of America, Nov. 27, Washington, D.C.

Graduations: Thomas M. Dreier received his Master of Science degree in Wildlife Sciences on December 9, 1977; the title of his thesis is "A technique for quantitative and qualitative viewing of aquatic bacteria using scanning electron microscopy". T.M.D. will stay on at TAMU and work on an expanded version of his M.S. problem for his dissertation.

Arthur E. Sowers received his Doctor of Philosophy degree in Biology on December 9, 1977; the title of his dissertation is "Effects of silicic acid analogs on silica body production and wall ultrastructure in stinging cells of **Urtica pillulifera**". A.E.S. received a Muscular Dystrophy Association post-doctoral fellowship to work under Charles R. Hackenbrock at UN.N.C. Medical School in Chapel Hill, North Carolina.

GALVESTON: University of Texas Medical Branch.

Division of Cell Biology: Jeffrey Chang has recently received a research grant from Rohm and Haas Co. The award is for $43,383 and the project entitled "Determination of no effect levels of ethylenethiourea". He has also received an NIH renewal grant for $63,856.

Paul Baur is also a co-prinicpal investigator on a grant awarded by the Rehabilitation Services Administration. The study is entitled "A comprehensive medical rehabilitation approach for burns" and has been funded for $449,358.

Peter Moller presented a paper at the fall meeting of the Texas Branch of the Tissue Culture Association held in Houston at the UT Medical School on October 14, 1977. The paper was entitled "Lectin receptors hepatoma ascites cells **in vivo** and **in vitro.**

Publications: Baur. P.S., T.R. Stacey, The uses of PIPES buffer in the fixation of mammalian and marine tissues for electron micscroscopy, J. Microscopy, **109,** 315-327 (1977).

Baur. P.S., D.L. Larson, D.F. Sloan, G.F. Barrat, An in situ procedure for the biopsy of pressure wrapped hypertrophic scars, J. Invest. Derm., **68**, 385-388 (1977).

Baur, P.S. G.F. Barratt, D.L. Larson. Scar contraction and myofibroblasts, Norelco Reporter, **24**, 19-23 (1977).

Chang, J.P., An ultracytochemical Study of the fate of Golgi apparatus in mitotic cells with special reference to regenerating livers (in press-book chapter).

Moller, P.C. J.P. Chang, Internalization of cationized ferritin receptors by rat hepatoma ascites cells, Exp. Cell Res., (in press).

Yokoyama, M.J.P. Chang, D. Hom, S.L. Wilson, An electron microscopic study on concanavalin A binding sites and their mobility in normal and cystic fibrosis fibroblasts **in vitro**, Pediat. Res., **11**, 765-769 (1977)

Department of Anatomy: Recent publications include: Payer, A., An ultrastructural study of the nerve plexus accompanying the ovarian artery and vein in the rat, Anat. Rec. (in press).

Callas, G. Fine structural observations on the origin and development of lamellar bodies in alveolar type 2 pneumocytes. In: pulmonary Macrophase and Epithelial Cells. ERDA Conference Symposium Series. (1977).

Department of Physiology: Recent publications include: Krauhs. J.M., L.A. Sordahl, A.M. Brown, Isolation of pigmented granules involved in extra-retinal photoreception in **Aplysia californica** neurons, B.B.A., **471**, 25-31 (1977).

Krauhs, J.M. Mahler, H.R., J.W. Moore, Protein turnover in photoreceptor cells of isolated **Limulus** lateral eyes, J. Neurochem. (in press).

LUBBOCK: Texas Tech University Health Sciences Centers, School of Medicine.

Department of Anatomy: Publications: Bartone, F.F., P.J. Gardner and J.C. Hutson. 1977. Polyglactin 910 Suture in the Urinary Tract. **Urol.**, 11:521.

Yee, J.A. and D.L. Wilbur. 1977. Utrastructure of Thyroid Follicular Cells Following Hypophyseal Portal Vascular Infusion of the Thyrotropin Releasing Hormone. **Amer. J. Anat.**, (In press).

Wilbur, D.L., J.A. Yee and S.A. Raiguel. 1977. Hypophyseal Portal Vessel Infusion of TRH in the Rat: An Ultrastructural and Radioimmunoassay Study. **Amer. J. Anat:** (In press).

Markwald, R.R., T.P. Fitzharris and D.H. Bernanke. 1977. Structural Analysis on the Matrical Organization of Glycosaminoglycans in Developing Endocardial Cushions. **Develop. Biol.**, February (In press).

Ledford, B.E., J. Rankin, V.C. Froble, M.J. Serra, R.R. Markwald and B. Baggot. 1977. The decidual cell reaction in the mouse uterus: DNA synthesis and autoradiographic analysis of responsive cells. **Biol. Repd.** (In press).

TEMPLE: Scott & White Clinic.

Publications: Brasher, G.W., Starr, J.C., McCombs, W.B. III, and Bourland, P.D.: T-lymphochtes in children with allergic respiratory disease. Annals of Allergy 39, 4:237-239, Oct. 1977.

Presentations: Quarles, J.M. Morris, N.G., and Leibovitz, A.: "Growth and Characterization of Human Colorectal Tumor Cells in Capillary Culture Devices" Presented at the Texas Branch Tissue Culture Association Association meeting in October.

Col. Leibovitz gave a seminar on "Techniques for the Establishment of Permanent Cell Lines from Human Colorectal Adenocarcinomas and Their Classification by Cytogenetic, Morphologic, and CEA Production Techniques" at the Department of Biochemistry and Molecular Biology and Surgery of the University of Texas School of Medicine in Houston.

Col. Leibovitz has been elected president of the Central Texas Research Society, Sigma Xi.

ARLINGTON: The University of Texas at Arlington, Department of Biology.

News Briefs: Prof. K. Kurosumi, Professor of Microscopical Anatomy at the Institute of Endocrinology, Gunma University, Maebashi, Japan, while a guest of Dr. Ernest Couch at T.C.U., visited the U.T.A. electron microscope laboratory and stereology facility.

Invited Papers: Dr. Howard J. Arnott, "Carbonates in Higher Plants" to the Third International Symposium on the Mechanism of Biomeneralization in Invertebrates and Plants, Kashikojima, Japan.

AUSTIN: The University of Texas.

Cell Research Institute: Dr. W. Gordon Whaley will be going to the USSR during the Spring semester of 1978. He will go on a Fulbright-Hays appointment and a contribution from the University of Leningrad. He will give a series of lectures at the University of Leningrad and other cities in Russia.

Department of Botany. Grants: Cole, G.T. has been granted an NSF US/Japan Cooperative Science Program Fellowship for the period January 1 to August 31, 1978. He will do cooperative research with Dr. Y. Hosawa and Dr. Y. Kitajima at the Department of Biochemistry of Gifu University School of Medicine. The research topic will be ultrastructural, chemical and immunological activities of the surface wall layers of pathogenic microfungi.

The University Research Institute has approved a grant to G.T. Cole, L.J. Berry (Department of Microbiology, Division of Allied Health and Life Sciences, University of Texas at San Antonio) and L.Pope (Department of Botany, University of Texas at Austin) entitled "Colonization and ultrastructural studies of gastrointestinal and systemic Candidiasis in Mice."

Cole, G.T.: A research proposal has been submitted to NSF requesting continuation of his grant on "Experimental Taxonomic Studies of Conidial Fungi."

Articles: Guentzel, M.N., L. Field, G.T. Cole and L.J. Berry. 1977. The localization of **Vibrio cholerae** in the ileum of infant mice. Proc. of Worshop of Medical Microbiol. IIT Research Institute, Chicago, pp. 275-282.

Olah, G.M., G.T. Cole and O. Reisinger. 1977. The role and chemical composition of secretory microvesicles in the hyphal apex and conidiogenous cells. Ann. Sci. Nat. Bot. et Biol. Vegetal. (Paris) (In press).

Cole, G.T. 1977. Conidiogenesis in the black yeasts. Mycoses (Pan American Health Organization Pub.), (In press).

Books: Cole, G.T. and R.A. Samson. 1978. Patterns of Development in Conidial Fungi. Pitman Publishing, Ltd. (In press).

Lectures: Gary T. Cole was invited by Dr. R.L. Shaffer to present a lecture entitled "Patterns of Conidial Development in the Fungi" in the Botany Seminar Series, University of Michigan, Ann Arbor.

Meetings: Gary T. Cole presented a paper entitled "Ultrastructure and Conidiogenesis of the Black Yeasts" during the Fourth International Conference on Mycoses, Brazilia, Brazil, June 6-8, 1977. He was also invited to present three papers during the Second International Mycological Congress, Tampa, Florida, August 27-September 3, 1977. The papers were: "Conidial Ontogeny of Some Dematiaceous Fungi Pathogenic to Man" in the symposium on "Taxonomy of Pathogenic Dematiaceous Hyphomycetes"; "Blastic and Tahllic Conidial Development in the Fungi Imperfecti" in the symposium on "Cytology and Biochemistry of Conidiogenesis"; and "Conidiogenesis in Marine Hyphomycetes" in the symposium on "Activities of Marine Fungi."

New Members: Robert D. Slocum from Ohio State University began work for a Ph.D. in Botany under Dr. Gary T. Cole.

L. Pope, a new graduate student, will be co-supervised by Gary T. Cole and will be working on systemic Candidiasis in mice.

Sharon Mattox, graduate student under Dr. G. Thompson is working on the effects of cations on membranes of **Tetrahymena.**

SAN ANTONIO: Department of Anatomy

Publications. Hanse, J.T., 1977. Spontaneous atherosclerosis: An ultrastructural study in the White Carneau pigeon. Virchows Arch. Path. Anat. and Histol. 375: 147-157.

Hansen, J.T., and W.W. Morgan, 1977. Effects of 6-OHDA on rat carotid body glomus cells. J. Cell Biol. 75: 108a.

Williams, M.G., and E.K. Adrian, 1977. The use of elemental iodine to enhance staining of thin sections to be viewed in the electron microscope. Stain Technol. 52: 269.

1977 — Seelig, Leonard L., Jr., William B. Winborn, and Elliot Weser. Effect of small bowel resection on the gastric mucosa in the rat. Gastroenterology 72: 421-428.

1977 — Winborn, W.B., D.L. Guerrero, and E.E. Hodge. Scanning and transmission electron microscopic studies on the effects of bile and aspirin on the gastric mucosa of the rat. Gastroenterology 72: 1168 (Abstract).

1977 — Seelign, L.L., Jr., W.B. Winborn, and E. Wesser. Changes in gastric glandular cell kinetics following small bowel resection in the rat. Gastroenterology (In press).

1978 — Winborn, W.B., D.L. Guerrero, and E.E. Hodge. The effects of bile on the gastric mucosa. Cytobios (In press).

Gravis, C.J., R.D. Yates, and I. Chen (1977). Ultrastructure and cytochemistry of epinephrine induced testicular degeneration. In: Male Reproductive System: Fine Structure Analysis by Scanning and Transmission Electron Microscopy. R.D. Yates and M. Gordon, eds. Masson Publishing USA Incorporated, New Yor.

Chen, I., C.J. Gravis, and R.D. Yates (1977). Fine structure of Lipid Doplets in Sertoli cells. In: Male Reproductive System: Fine Structure Analysis by Scanning and Transmission Electron Microscopy. R.D. Yates and M. Gordon, eds. Masson Publishing USA Incorporated, New York.

Gravis, C.J., and F.J. Weaker (1977) Testicular Involution following Optic Enucleation: an Ultrastructural and Cytochemical Study. (In Press). Cell Tiss. Res. 184:67

Gravis, C.J. (1977) A scanning electron microscopic study of the Sertoli cell and spermination in the Syrian hamster. (In press). Am. J. Anat.

Gravis, C.J. (1977) Testicular involution following optic enucleation: the Leydig cell. (In press).Cell Tiss. Res.

Gravis, C.J. (1977) Testicular involuation after optic enucleation: Ultrastructure and alkaline phosphatase cytochemistry of the peritubular tissue. (In press). Am. J. Anat.

Congress: 1) Effect of fetal brain extract on the differentiation of pituitary anlage cells. Ishikawa, H. and M. Shiino Anatomical Rec. 187, 611-612, 1977 (abst.).

2) Four subtypes of prolactin producing clonal strains derived from Rathke's pouch epithelium. Shiino, M.H. Ishikawauand E.G. Rennels. 58th annual meeting p. 123, 1977 (abst.).

3) Effect of estrogen, dopamine, and TRH on prolactin release from clonal pituitary cells. Herbert, D.C., H. Ishikawa, M. Shiino and E.G. Rennels. 59th annual meeting p. 275, 1977 (abst.).

4)In vitro accumulation of secretory granules in clonal cells of the rat anterior pituitary gland. Shiino, M.H. Ishikawa and E.G. Rennels. J. Cell Biol. 75(2), 88a (abst.).

Paper:

1) Functional clones of putuitary cells derived from Rathke's pouch epithelium of fetal rats. Ishikawa, H., M. Shiino, A. Arimura and E.G. Rennels. Endocrinology 100, 1227-1230, 1977.

2) Effects of fetal brain extract on the growth and differentiation of rat pituitary anlage cells. Ishikawa, H., M. Shiino and E.G. Rennels. Proc. soc. Exp. Biol. Med. 155, 511-515, 1977.

3)In vitro and in vivo studies on cytodifferentiation of pituitary clonal cells drived from the epithelium of Rathke's pouch. Shiino, M., H. Ishikawa and E.G. Rennels. Celland Tiss. Res. 181, 473-485 (1977).

4) An immunocytochemical study of a rat pituitary multipotential clone. Bowie, E.P. H. Ishikawa, M. Shiino and E.G. Rennels J. Histochem, Cytochem. (In press).

5)Secretory granule accumulation in pituitary clonal cells derived from Rathke's pouch epithelium. Shiino, M.H. Ishikawa and E.G. Rennels Cell Tiss. RES. (In press).

6) Specific subclones derived from a multipotential clone of rat anterior pituitary cells. Ishikawa, H., M. Shiino and E.G. Rennels. In Vitro, (Submitted).

News Brief: Dr. Earle Adrian has received grant funds to study "Cell Proliferation in Injured Nervous Tissue" from the National Institute of Neurological and Communicative Disorders and Stroke for the period December 1, 1977 through November 30, 1978. This is the ninth year of a ten year project.

Dr. D. Herbert has received a NIH grant, "gonadotrophic hormones in malnourished rats." (3 years).

Damon C. Herbet and Edward G. Rennels 1977.

FT. SAM HOUSTON: Brooke Army Medical Center.

The Diagnostic Virology Laboratory and Electron Microscopy Laboratory of the Department of Pathology are combining efforts to compile an Atlas of Virology. This project was funded through Clinical Research at BAMC. Steven K. Koester is joining the society. Steve has a M.S. in Microbiology from University of Wisconsin at La Crosse, and is currently working in the Virology Section.

SAN ANTONIO: Weckerling's World.

Alan B. Weckerling has announced the expansion of his business enterprise, Weckerling's World, to include useful ultramicrotomy tools. The first product, a special thin section "herding" hair, will be introduced at the San Antonio meeting. Future directions for growth are now being studied with special attention being given to the feasibility of a sectioning service. Alan has been a member of TSEM since 1971 and has just become a corporate member. The address is 6702 Cypress Lake, San Antonio, Texas 78244. 512/661-7000.

Letter to the Editor

One day a few years ago while at that great saltwater fount of medical education, the chief officer in the Office of Medical Education said to me, "This class is certainly different from any I have seen before. I've had virtually no complaints from the students. I don't understand it."

"I do," says I, unabashedly. He replied quizically, "Oh! Well, tell me why!" I felt embarassed really, swelling from the strength of my conviction, instructing a career professional in educational psychology with such a basic observation: "It's simple. They had better toilet training!"

His shock was registered by subsequent stammering and incoherent babbles as I strode away, first feeling somewhat superior, then feeling just sorry as hell that education had been inundated by "educationists" who see but do not perceive.

We have been embraced by a massive takeover in the American educational system by "educationists" where every student is classified as to percentiles, stanines, placements, accelerations and declarations, or what have you. Every conceivable device has been employed to discover how to develop all those latent and hidden talents **they know** to exist in all of our students. And this has come at the expense of proven techniques from training, such as spelldowns, word lists, vocabulary sheets, number drills, arithmetic tables, latin, classic readings and foreign languages. Still, we are perplexed as to WHY JOHNNY CAN'T READ!

Our schools have become a proving ground from John Dewey's philosophy of development of the **whole child.** This has led to the "do your own thing", and not just in the classroom; the striving for free expression, the compromise of authority and the abdication of responsibility from faculty and especially administrators. Somehow, the notion has been established as quasi-fact that this is what our country is all about: the do-your-own-thing — freedom of expression — anything goes as long as it's between two consenting adults and it's not hurting anyone else!

Educationists have made a shambles of our educational system, first in the primary grades, then in the high schools, and now in the colleges, universities and even the professional schools. Students are patronized unmercifully until they are pushed onto faculty committees, originate evaluations and design new curriculae. Two years ago the first-year class President even had the gall to propose that a student committee review and approve all test questions before they were given to the class! When I told of this to our esteemed "educationist" he said,". . . maybe that wouldn't be such a bad idea."

Just one year ago Alan R. Shucard wrote in The Chronicle of Higher Education that in his class during a discussion of Willy Loman (Death of a Salesman) and the attempt:

"to open up vistas of understanding, broad and deep . . . I had attempted through the use of those references to connote . . . the dimensions of a life grasping, pushing, of a human being's sensing that he had somehow been transmogrified into an insect . . . Willy is, after all, in a large measure, Tantalus, Sisyphus and Kafka, and in discomforting measure, us — skewered on credit and grilled over the obsolescence of the things he buys and, worst of all, of himself. The matter was illuminated for the class after a while, but, I had to reinvent fire for them; they had not heard of Prometheus, either."

Shucard went on to say that, "we find that we cannot really speak to each other . . . We do not pay attention" to some common language in which we might meet through the input of our own sensibilities; instead, "the thunder has grown louder in our classrooms and in political life."

We have fostered in our schools, colleges and even professional schools a dependency syndrome. Administrators and teachers, like new mothers, are more concerned with every little squeak and cry from the students, reacting knee-jerk responses, than with providing the **stimulus** for response, the discipline for a common learning situation. As a result, principals, deans, presidents have been smothered with petitions because students **know** they will respond and will respond favorably. In the eyes of others, however, the responses are timid and weak, perhaps because of fear of the courts or as a defensive measure attempting to push off responsibility elsewhere. As a consequence our basic problems simply won't go away.

William Raspberry writes in the Washington Post that Time magazine recently attacked the "soft-sell approach to education, the dizzying array of elective courses (and the diminishing list of required ones) the everybody do-your-own-thing and we'll give-it-a-passing-grade nonsense, the 'relevant' courses, such as interior decorating." He argues, however, that back-to-basics cannot be implemented because the classrooms now tend to keep those in school who are disruptive and perhaps unteachable. Efforts toward any basic learning process by teachers are compromised because they are preoccupied with discipline problems.

Even in the professional schools it is virtually impossible to remove the few who really do not belong because the administration is loathe to sacrifice one cent of state and federal money, which is allocated on a per-head count of students.

Very recently our tax dollars were committeed by the National Research Council to evaluate federal spending on education. Their investigating committee concluded that spending had been wasteful, and that there "is no . . . basis for understanding human intellectual development in the adult years". They called for research on (1) the nature of cognitive development, (2) education and reading, (3) the brain and neural processes, (4) innovation (there's that word again) and change in educational institutions, and **ad infinitum.** In short, their study concluded that we do not know how college students and other adults learn! This is a fine state of affairs and a rotten testimonial to American educational institutions especially since they first appeared in this country more that 300 years ago!! **IF** you listen to our "educationist" researchers! Johnny could not read in 1930; he couldn't read in 1950, nor in 1978. He is not going to be reading in the year 2000 either with this kind of support.

Now, what does toilet training have to do with all of this? The intent of my remark to the medical "educationist" was to emphasize early discipline in an individual's learning experience. We saw a dramatic change in attitude, industry and behavior of many students approximately four years ago. The reason was that those students were not war-babies. The fathers were in the home when they were infants, and it made the difference. This opinion, and that is precisely what it is, is not meant as an indictment of womanhood; rather, it points out the need each child has to identify his or her position within the family structure. Children search for authority and they usually are taught to respond to authority. Certainly, when they enter school the groundwork has already been laid, and the schools should not be unfairly burdened with teaching students how to recognize proper order. It is that environment which creates the best learning situation possible.

How important is cognition of order and authority and independent action within that scope in one's early life? Our National Research Council study could not say. Perhaps it is everything! Recently two psychiatrists finished a four-year study of the criminal personality and found there was no qualitative difference between that and so-called normal personality; but, they found significant quantitative differences and characterized the criminal mind with 52 errors of thinks, which dramatically began appreaing between the ages of 1 to 5!

Perhaps we are coming to see that the mission of our educational institutions is not to reveal the full expression of the individual, **a priori,** for we well may "inherit the wind", but to provide that foundation, the basic fabric, of creative expressions with sound design. Hopefully when we have suffered to the last from all the analyses of distributions, standard errors, percentiles, and the like, we can once more look into the eyes of Johnny and find that spark of innocent passion which in his simple and uncofusing way asks, "will you teach me to read". At that point we will not be questioning ourselves; we will begin, again, to teach what needs to be taught.

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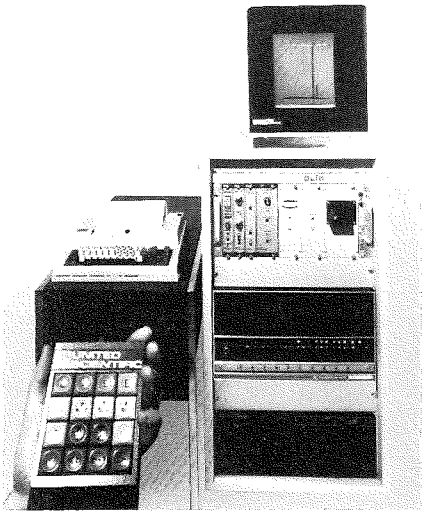

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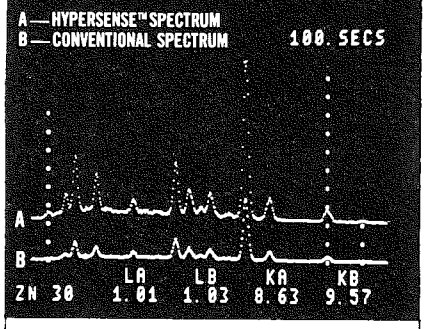

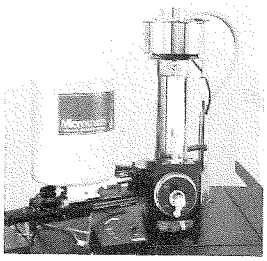

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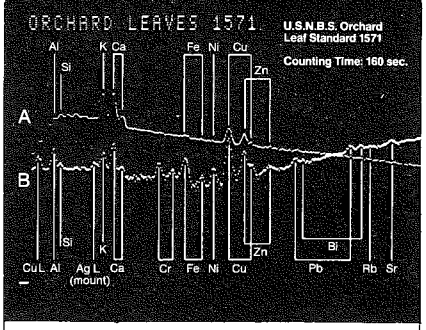

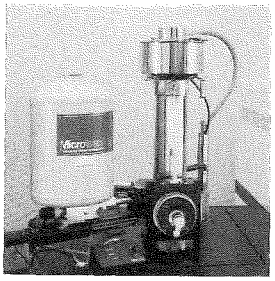

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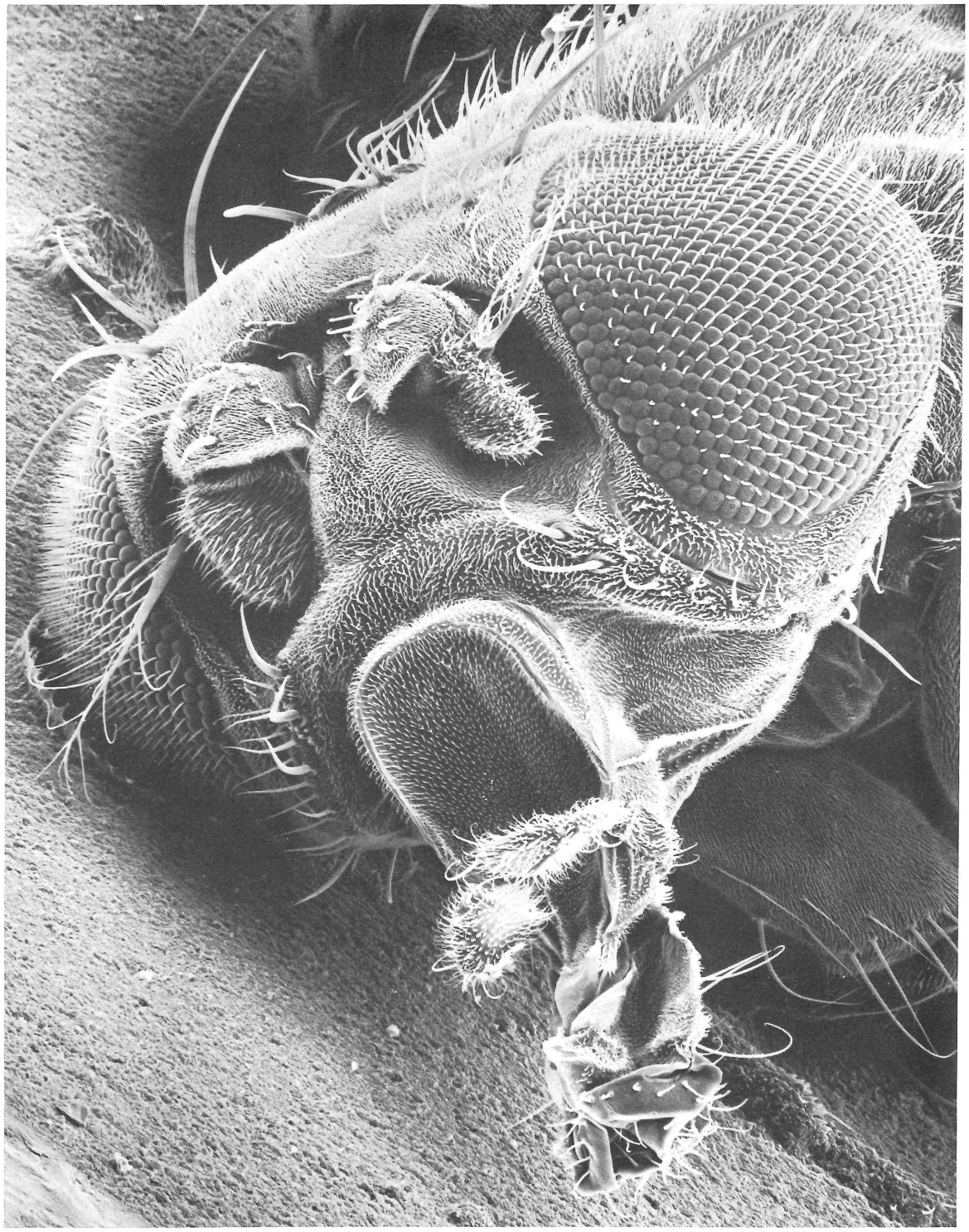

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Situation Wanted — Barbara Bruton, M.A., Eight and one-half years research experience under Dimitrij Lang doing quantitative electron microscopy of nucleic acids. Prior experience in microbiology especially psychology and food industry quality control. Seeking position requiring initiative, responsibility for a smooth running laboratory, and involvement in research projects. Current address: University of Texas at Dallas, Biology Programs, Mail Station FO 3.1, P.O. Box 688, Richardson, Texas 75080.

Situation Wanted — Raul Joseph Alvarado, 5300 Tropicana, El Paso, TX 79924. (915) 751-0691. Single, 5-10, 180 lbs, born July 24, 1951. Wants career in medical field as a Laboratory Technician. Majored in Microbiology at El Paso Community College, GPA 3.6 on a 4.0 scale. Presently employed as Bio-Lab aide, electron microscopy, Dept. of Pathology, William Beaumont Army Medical Center, El Paso, Texas. Has been recommended by Bernhard E.F. Reimann, Dr., rer. nat., Chief, at William Beaumont Army Medical Center. Dr. Reimann is in the process of training Mr. Alvarado and will be available for full-time job on Jan. 21, 1977. Other references and a complete resume are available.

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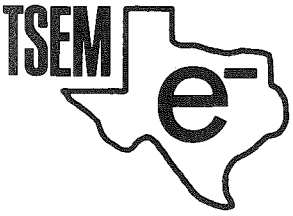

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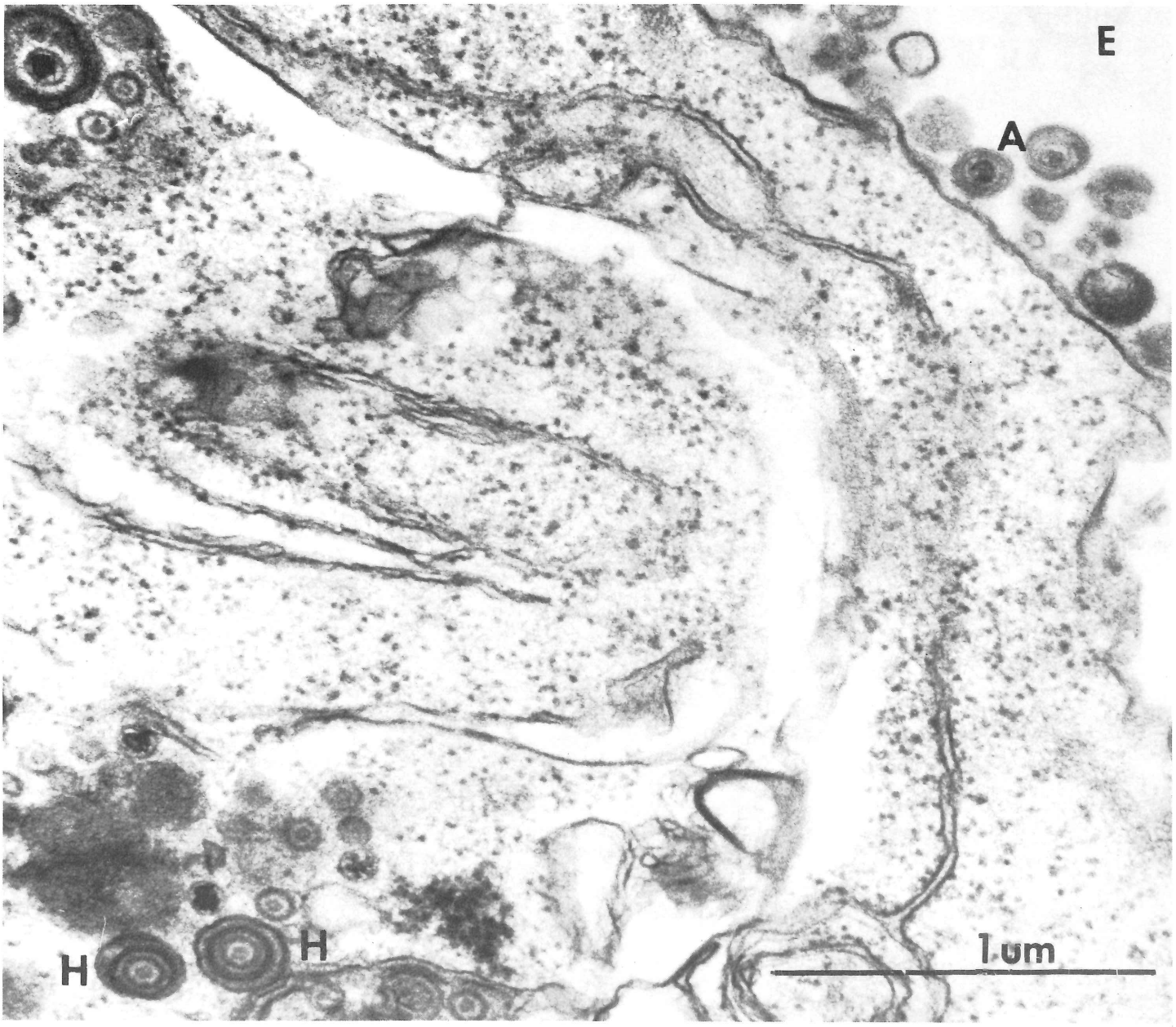

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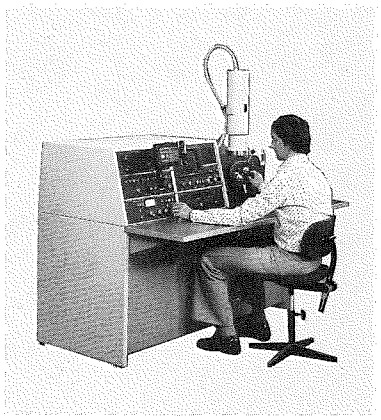

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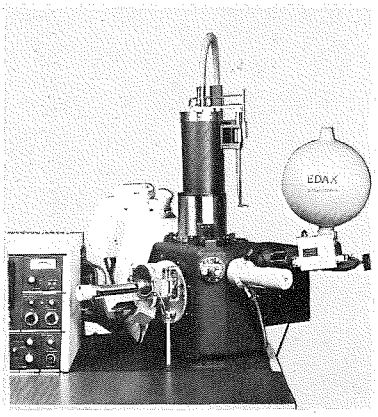

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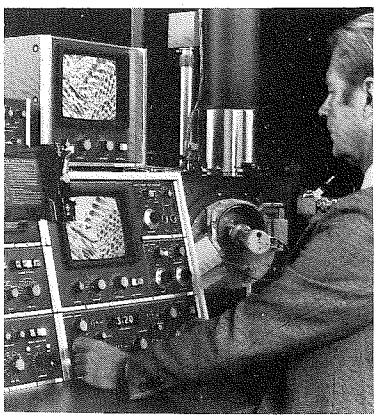

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