

TSEM **Texas Society for Electron Microscopy**
e-NEWSLETTER



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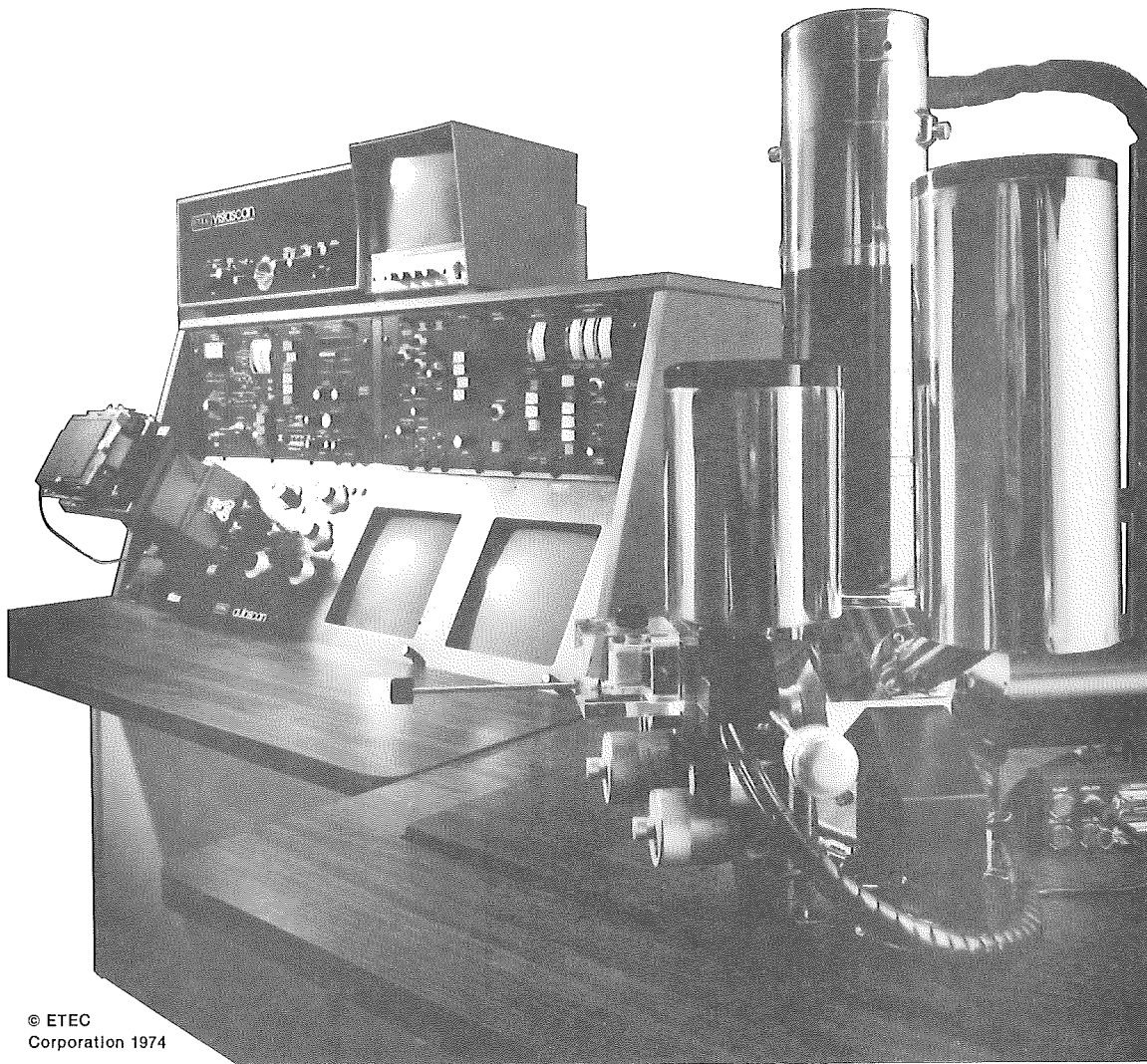
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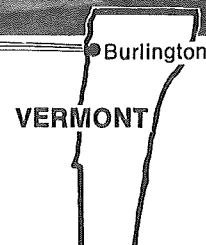
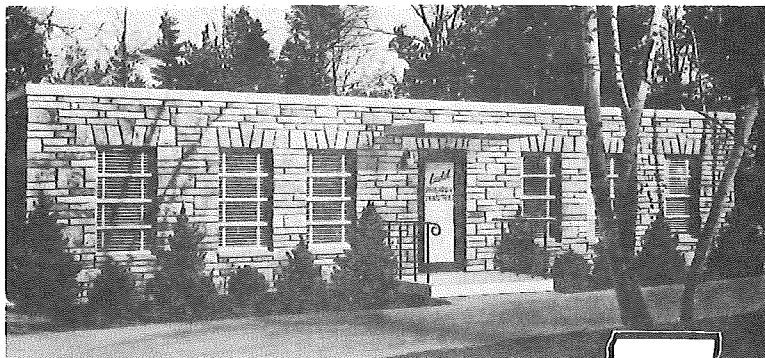
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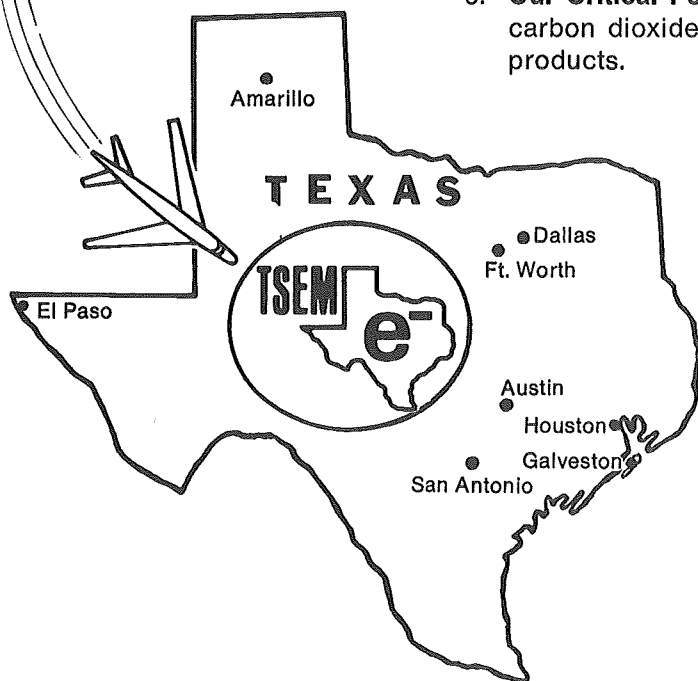
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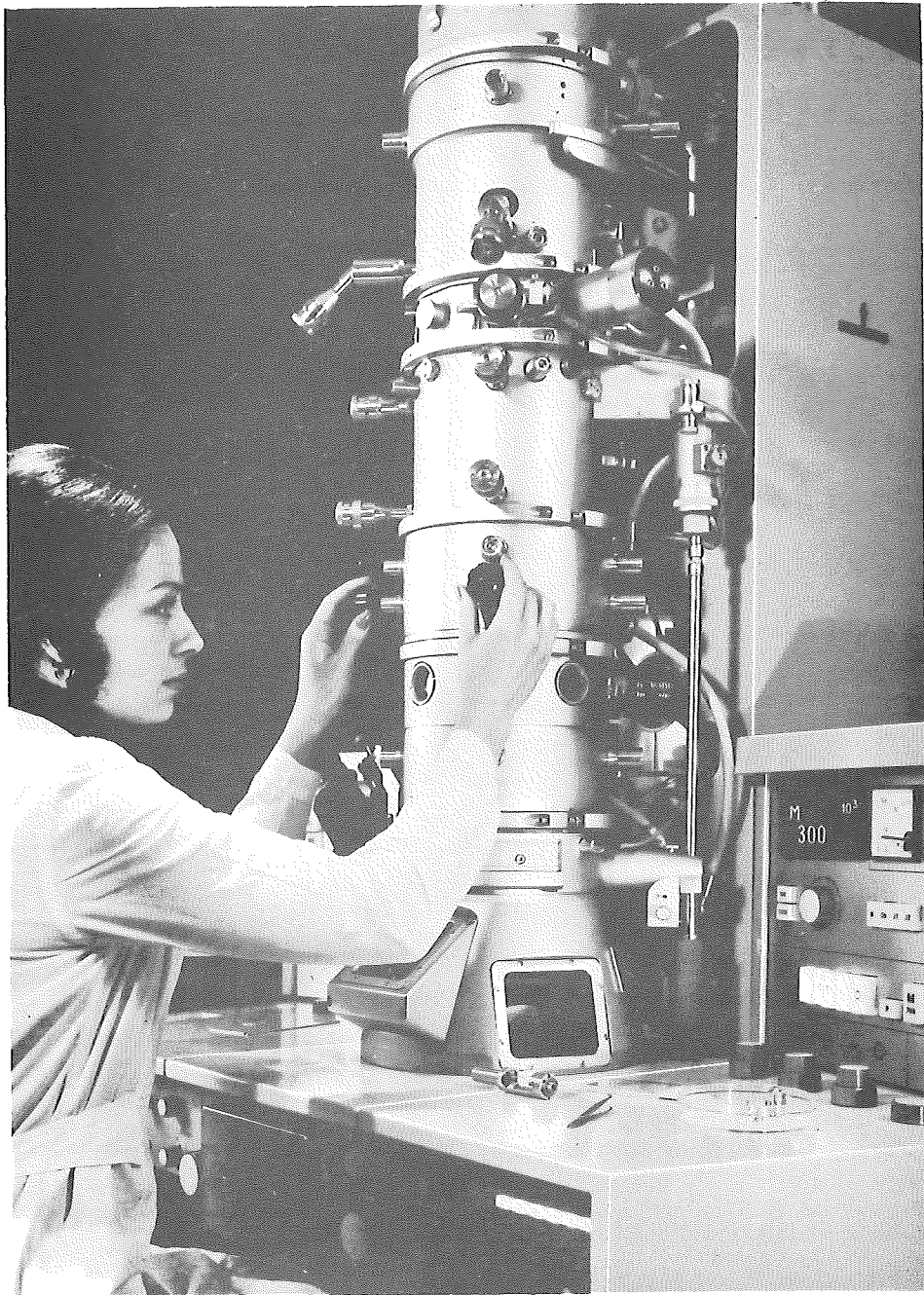


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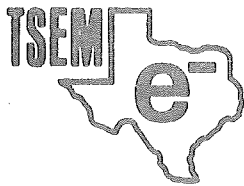
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TEXAS SOCIETY FOR ELECTRON MICROSCOPY -

" FOR THE PURPOSE OF DISSEMINATION OF RESEARCH
WITH THE ELECTRON MICROSCOPE "

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Officers 1974-1975:

Terry Hoage
President
Biology Department
Sam Houston State University
Huntsville, Texas 77340
713-295-6211 ext. 1666

Jerry Berlin
Secretary
Department of Biology
Texas Tech University
Lubbock, Texas 79409
806-742-3169

Ward Kischer
Vice-President
Department of Anatomy
Univ. of Texas Medical Branch
Galveston, Texas 77550
713-765-1809

Ivan Cameron
Newsletter Editor
The University of Texas
Health Science Center at
San Antonio
San Antonio, Texas 78284
512-696-6537

Ernest Couch
Treasurer
Department of Biology
Texas Christian University
Fort Worth, Texas 76129
817-926-2461 ext. 461

Vinita Allison
Program Chairman
Department of Biology
Southern Methodist University
Dallas, Texas 75275
214-692-2730

Robert Turner
Immediate Past President
Department of Pathology
Scott & White Clinic
Temple, Texas
817-778-4451 ext. 438

Send Letters and Inquiries to: William A. Pavlat
Managing Editor of Newsletter
Department of Anatomy
The University of Texas
Health Science Center at San Antonio
San Antonio, Texas 78284
512-696-6537

Comment

There is confusion in the land concerning the definition of science as opposed to the definition of technology.

Science is interested in but two questions: What is nature like? How is it the way it is? Science is not a compartmentalized activity, but is as much a part of the mainstream of human striving for fulfillment and betterment as are art, music, poetry, and literature. Science replaces ugly rumors, beliefs and speculations with the best facts then available. It is odd that a so-called educated person is embarrassed if he does not know who painted a particular canvas, who composed a particular symphony or who wrote a particular book, yet he cares little as to which variety of uranium atom-U-235 or U-238 undergoes fission with slow neutrons. That's a fact that could affect his whole future.

Technology, on the other hand, is interested in how to use the resources of nature more effectively to satisfy the real, or imagined, needs and desires of men. It seeks knowledge as a matter of achieving a goal or a result. Technology is the world of the engineer and the physician. It is the world of computers, nuclear weapons, nuclear reactors, missiles, antibiotics, pesticides, jet airplanes, communication satellites, automobiles, contraceptives and television sets.

Science and technology have become greatly confused in this country. We have the ironic situation in which NASA (National Aeronautics and Space Administration), which represents a technological undertaking, keeps on trying to justify its existence on the contributions it makes to science. And when scientists testify for science before Congress, they usually justify money for their research on the basis of whether their research is useful--a matter of technology.

Here is the dilemma of modern science: The scientist wants to do science but he essentially only can get funds to do technology.

America now spends billions of dollars per year on activities conveniently lumped together as research and development--R and D. But if you look very closely, most of the money is for D and only very little is for R. It is technology and not science that is getting the lion's share of governmental, industrial and university financial support. Technology would not be possible were it not for the basic research that provided the basis for development.

A great deal of commentary is made these days from scientists, because their funding has been cut back, that the country is being inundated by a wave of anti-intellectualism. This is utter nonsense because, the country never was swept previously by a wave of intellectualism. It is suggested that the commentary on funding be directed to Congressman and Senators to better facilitate communication with the granting agencies and scientists.

The above is excerpted from a talk by Irving Bengelsdorf, Director of Science Communications at Cal Tech.

President's Message

As I assume the seat of leadership of TSEM, it becomes even more obvious that the success of this, or any, society lies within the membership and their willingness to contribute meaningfully to the defined intent of the society. Our intent is to promote and disperse E. M. research information, which means that members relay to the other members their current findings. This is effectively done within the framework of our three meetings held annually. It has greatly disappointed me in the past when, as program chairman, we had to "shake the bushes" to get members to present their work. In the final assembly we see a core of contributors who respond. Where are the others? Nationally and internationally known members of TSEM have much to offer in terms of the societies' intent.

When I came to Texas in 1968 and joined TSEM, my training was limited and the conversations and contacts that I made as a member increased my working knowledge greatly as I listened to the "Old Pro's". I encouraged graduate students to go, to present our work, to mingle and listen, and all were enthused with the learning atmosphere. This is what we, as a society, must maintain through increased membership and participation. Graduate students could learn from the contacts with the older, established members. We want the old members to participate and the new members to learn and contribute.

Additionally, I would like to take this opportunity to comment on TSEM's activities during this past year. A unique fall meeting has been implemented to mix science and relaxation in equal doses. This concept is continued with the fall meeting at the Waterwood resort community. This past winter meeting was a success with excellent papers, good attendance and tremendous exhibitor participation. In May we met on the TAMU campus for our much heralded graduate student award meeting. The meeting went smoothly, the graduate student papers were superior and the workshops most informative. All we lacked was senior member attendance. As can be readily ascertained, the society has served all levels of E. M. participants in a continuing education format. In keeping with this premise, this fall meeting is centered on grantsmanship as it pertains to our research fields and endeavors. All laboratories have experienced the money crunch and I hope some of the questions you have concerning grants and funding can be answered either by our panel or national speaker.

Terry Hoage
President

T.S.E.M. MINUTES - BUSINESS MEETING

The T.S.E.M. Business Meeting was held May 25, 1974 at 9:00 A.M. on the Texas A & M Campus at College Station. President Robert Turner presided. The minutes of the last meeting were read and approved. The Treasurer's report was read and approved.

President Turner announced:

1. \$100.00 had been received from E.M.S.A. for the San Antonio meeting.
2. The February meeting with L.S.E.M. had been moved out of the French Quarters because of room costs.

The Secretary reported the election results for 1974-1975 officers as below:

Ward Kischer, UTMB, Galveston, was elected Vice President.

Venita Allison, Southern Methodist University, was elected Program Chairman.

President Turner discussed the By-Laws changes; pertinent points included:

1. The last revision in the By-Laws was in 1968.
2. Ratification of the changes would be accomplished by a ballot to the membership.

Joe Wood commends Robert Turner, and specifically noted Bob's guidance the past year as President and the fact that he is the only person who has attended every T.S.E.M. meeting.

Terry Hoage reported that a resort on Lake Livingston was being considered for the Fall, 1974 meeting.

The meeting adjourned at 10:45 A.M.

Jerry D. Berlin
Secretary

FINANCIAL REPORT

Texas Society for Electron Microscopy

through Spring Meeting 1974

Receipts:

1. Registration fees and dues at meeting	\$507.25
2. Dues received since A & M meeting	212.75
3. Corporate support of Newsletter	150.00
4. Exhibit security at San Antonio meeting	400.00
5. EMSA support of San Antonio meeting	100.00
Sub-total	<u>\$1320.00</u>

Disbursements:

1. Spring Newsletter	\$238.60
2. Mailout expenses	454.73
3. A & M. Food Services	525.00
4. Program chairman's expenses	119.03
5. Corporate seal, certificates and transfer lettering	121.26
6. Graduate student travel	132.40
7. Graduate student award	200.00
8. Other executive committee expenses	93.56
	<u>\$1884.58</u>

Summary:

Total disbursements	\$1884.58
Total receipts	<u>1320.00</u>
Amount exceeding income	<u>\$564.58</u>

Bank Balance (checking) prior to meeting	\$2430.10
Bank Balance as of August 30, 1974	1865.50
Certificate of deposit and interest	<u>1046.01</u>
GRAND TOTAL	\$2911.51

ANNOUNCEMENTS

The LSEM Local Arrangements Committee is busily making all preparations for the Fourth Joint TSEM/LSEM Symposium in New Orleans, February 20-22, 1975. A call for abstracts will soon be received by the joint membership. As usual, the abstracts from the Symposium will be published in the Texas Reports. All the Louisiana colleagues hope that our joint meeting will be as successful as always and encourage the joint membership to lend its support and attendance.



Dr. Jeffrey P. Chang

Dr. Jeffrey P. Chang, a long time TSEM member, has just received the highest scientific honor given by the Republic of China (Taiwan) - election to membership in Academia Sinica. The Academy is located in the Republic of China (Taiwan), Nankang, Taipei district and was founded in 1928. There are only 77 elected members in this International organization. One half of the Academy members are residents of the United States and other countries and the other half are residents of China. Individuals chosen for this award are the top researchers and scientists in their field, they are known for their fine academic and research contributions, as well as for their highest level of moral standing in Chinese tradition.

BYLAWS

TEXAS SOCIETY FOR ELECTRON MICROSCOPY

I. PURPOSE

The purpose of this society shall be: (a) to increase and disseminate the knowledge concerning electron microscopes and related instruments, associated technology and methodology and results obtained there from as applicable; (b) to promote free exchange of ideas and information among electron microscopists and interested participants.

II. MEETINGS

Meetings of the Society shall be held at such times and places as may be designated by the Executive Committee. There shall be a minimum of three meetings per year, one of which shall be a general symposium.

III. OFFICERS

- A. The elected officers of the Society shall be a president, president-elect, secretary, treasurer, program chairman, program chairman-elect and student representative.
- B. The current officers and the immediate past president shall constitute the executive committee and shall appoint a newsletter editor who shall also serve as a member of the executive committee.

IV. DUTIES:

- A. President shall preside at all business meetings of the Society and at the meetings of the executive committee.
- B. President-elect shall assist the president and substitute for him in his absence and perform such duties as assigned by the president.
- C. Secretary shall maintain the records of the Society other than financial, and promulgate announcements to the membership. The secretary's term of office shall be two years.
- D. Treasurer shall be custodian of the Society funds and shall account for them in accordance with accepted business practice. The treasurer's term shall be two years.
- E. Program Chairman shall be responsible for organizing the various learned undertakings of the Society and shall preside at the meetings of the program committee.
- F. Program Chairman-Elect shall assist the program chairman and substitute for him in his absence.
- G. Student Representative shall represent the student membership of the Society on the executive committee.

V. MEMBERSHIP

- A. Regular Membership - Any individual other than a student or corporate organization who has an interest in electron microscopy.
- B. Student Membership - An individual enrolled in an approved academic program and pursuing either an undergraduate or graduate degree.
- C. CORPORATE MEMBERSHIP - Reserved for those commercial organizations maintaining an interest or participation in electron microscopy. This membership entitles a corporation to designate one representative at the current corporate dues rate who shall be entitled to all mailouts and privileges. All other representatives of the same corporation may become a regular member.
- D. Honorary Membership - Restricted to the following categories:
 - (1) distinguished scientists who are not members of TSEM but who have made significant contributions to this Society.
 - (2) TSEM members who are to be recognized for extended and outstanding service to this Society and or electron microscopy in Texas. Honorary members are exempt from annual dues in the Society.

VI. DUES

- A. The amount of annual dues and date(s) of payment shall be set by the executive committee.
- B. Dues shall become payable on May 1 of each year.
- C. Any member that is delinquent in payment of dues for a period of six months shall be removed from membership.

VII. ELECTIONS

- A. In February of each year the executive committee shall appoint five members, including one student member to form the nominating committee. This committee shall nominate two candidates for each office for the next year. The secretary shall serve as Chairman of the Nominating Committee.
- B. The nominating committee shall prepare their slate of nominees with due consideration to the geographical area and fields of interest represented by membership of the Society.
- C. Additional nominations may be initiated by the membership by a petition to the executive committee signed by at least five per cent of the members in good standing.
- D. Ballots shall be mailed to the membership in March and shall be accepted by the executive committee until the fifteenth of April. The results shall be announced by the secretary at the Spring business meeting.
- E. The candidate receiving over one half of the votes on the ballots actually received shall be the winner. If more than two candidates

are running for an office, the ballots shall be arranged so that the membership ranks all candidates in order of preference. If on the first count, no candidate receives a majority vote, the ballots of the candidates receiving the lowest number of votes shall be recounted with the second choice now counting as first choice votes, and so forth, until only two candidates remain. In the event of a tie vote, the combined executive and nominating committees shall decide the winner. The ballots shall be counted by the executive committee.

VIII. AMENDMENTS

- A. Amendments to these bylaws may be initiated in two methods:
 - 1. By the executive committee, or by
 - 2. Petition of ten percent of the members in good standing
 - 3. Suggestions of amendments shall be received at any TSEM business meeting.
- B. The proposed amendment shall then be promptly submitted by mail to the membership by the secretary with signed statements of support and/or opposition. The ballots shall be accepted by the executive committee for two weeks after the date of mailing. The executive committee shall count the ballots and the amendment(s) shall be ratified if it receives a favorable majority of the votes cast.
- C. Any member in good standing can, if he so desires, be present at the counting of any ballots.
- D. These amendments to the bylaws shall be ratified by a two-thirds majority of the returned membership ballots.

AN IMPROVED FIXATION PROCEDURE FOR THE
PRESERVATION OF MICROFILAMENTS AND
MICROTUBULES IN THE ANTERIOR PITUITARY GLAND

Jerzy B. Warchol¹, Damon C. Herbert and
Edward G. Rennels

Department of Anatomy
The University of Texas Health Science Center at San Antonio

¹ Fogarty International Fellow from
The University of Poznan, Poland

In the past few years, several laboratories have focused their attention on the elucidation of the physiologic role of microtubules and microfilaments. A handful of reports have been published describing microtubules in some of the anterior pituitary cells, such as in the growth hormone and prolactin cells. We wish to describe a fixation procedure which gives excellent preservation of both microfilaments and microtubules in rat and rhesus monkey pituitary glands. We emphasize the need to use caution in avoiding extreme changes in temperature and to be sure that the osmolarity of both fixing solutions is approximately 400 milliosmoles.

The fixation procedure is as follows:

1. Dice the tissue into very small pieces with a razor blade.
2. 1-2 hrs. on a rotator at room temperature in small bottles containing 2.5% glutaraldehyde (Eastman Kodak) in 0.1 M cacodylate buffer pH 7.0 (osmolarity 400 mosm.).
3. Transfer rotator to cold room at 4°C for 1 hr.
4. Place bottles with fixative in a container of crushed ice for 10 min.
5. To two parts of the above fixative containing the tissue add one part ice cold 1% osmium tetroxide in a solution of 0.1 M cacodylate buffer pH 7.0 and 6% glucose (osmolarity 400 mosm.). Place solution in a container of crushed ice for 15 min. and decant the fixative.
6. Add new fixing solution prepared as in step 4, fix for 15 min. in ice bath and decant.
7. Add a 1:2 solution of ice cold 2.5% glutaraldehyde in 0.1 M cacodylate buffer pH 7.0: ice cold 1% osmium tetroxide in a solution of 0.1 M cacodylate buffer pH 7.0 and 6% glucose. Keep on crushed ice for 15 min. and decant.

8. Repeat step 6 using freshly prepared solution and decant after 15 min.
9. Add ice cold 1% osmium tetroxide in a solution of 0.1 M cacodylate buffer pH 7.0 and glucose. Keep on crushed ice for 1-2 hrs.
10. Decant and wash with two changes of ice cold distilled water.
11. Stain at 4°C for 16-18 hrs. with 0.5% uranyl acetate in 0.1 M acetate buffer pH 5.8.
12. Dehydrate with absolute alcohol and acetone.
13. Embed in Spurr's low viscosity embedding medium.
14. Stain for 5 min. with uranyl acetate and 20 min. with lead citrate.

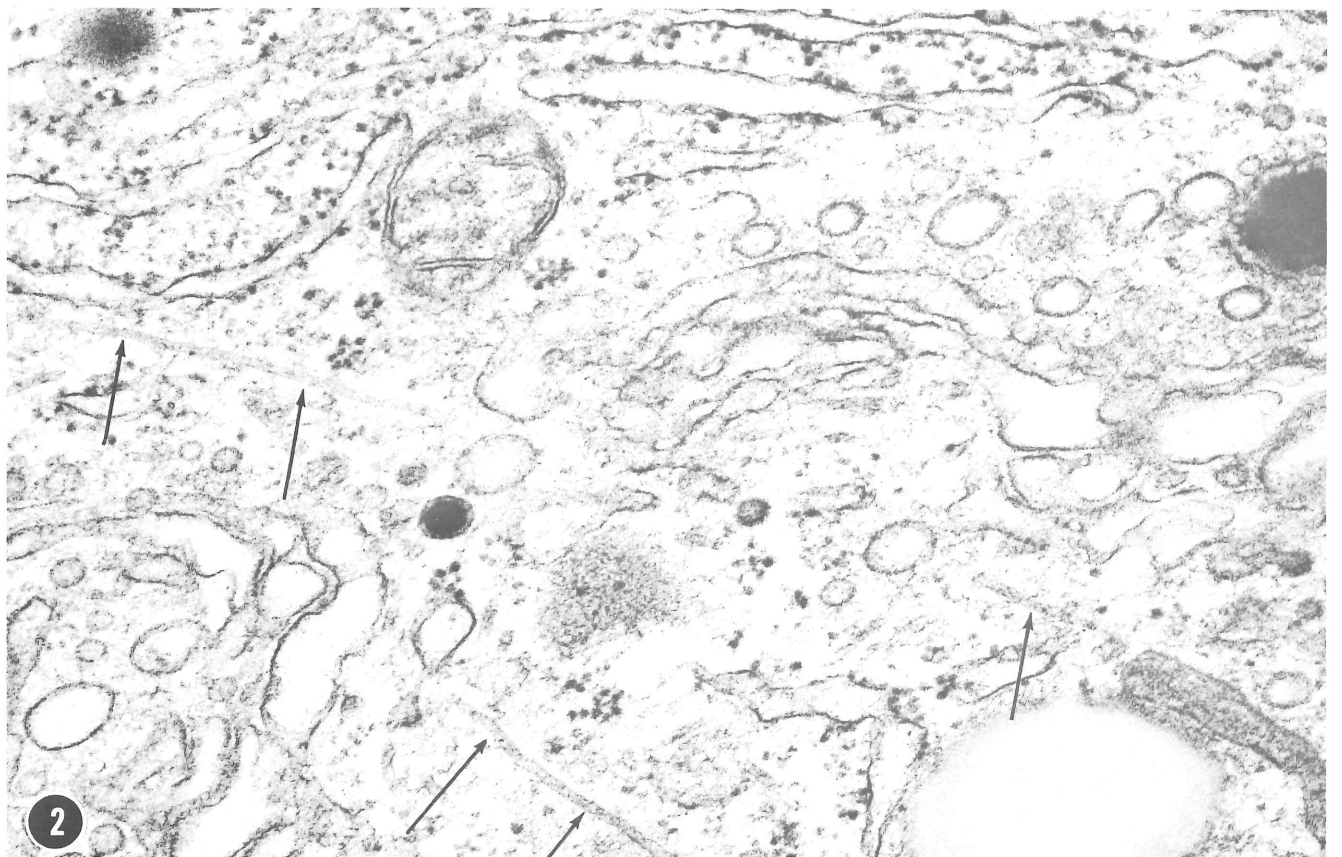
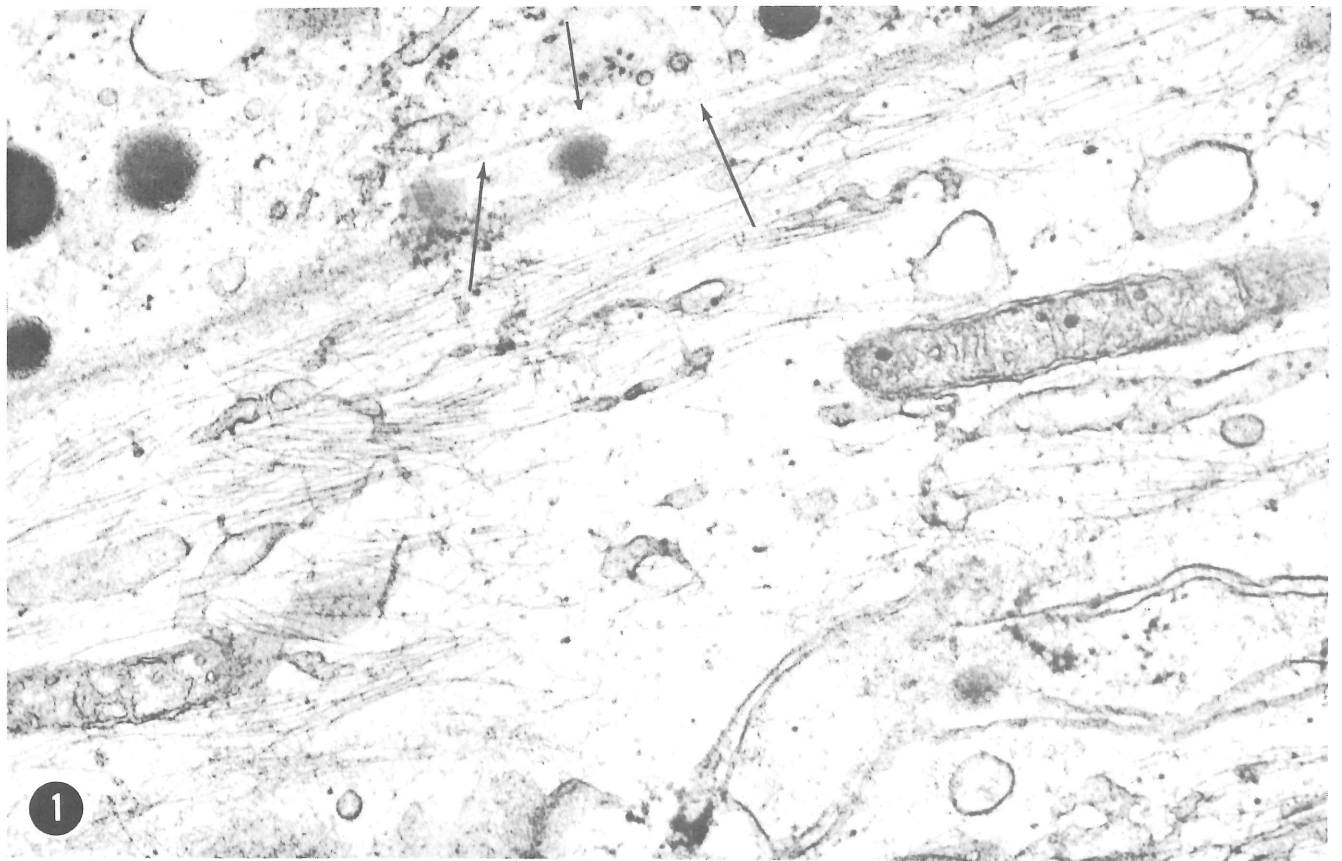
Two alternative fixative procedures were also used. In one, steps 6 and 8 were not performed. In the second alternative procedure, the tissue was fixed as described in steps 2-4. The glutaraldehyde was then decanted and replaced by ice cold 1% osmium tetroxide in 0.1 M cacodylate buffer and 6% glucose. This was kept for 1-2 hrs. in crushed ice and decanted. The tissue was then processed as described in steps 10-14.

Both microfilaments and microtubules were well preserved in the anterior pituitary cells in the rat and the rhesus monkey (Figures 1 and 2). The microfilaments were especially prominent in agranular cells of the rhesus monkey pituitary (Figure 1). There was little difference in tissue preservation when either the original or the first alternative procedure was employed. This was especially true when very small pieces of tissue were fixed. With somewhat larger pieces of tissue, better morphology was obtained when the longer procedure was followed. The least advantageous method was the second alternative method. Microfilaments and microtubules were rarely seen when this procedure was followed. This may have been due to the addition of ice cold osmium tetroxide to the glutaraldehyde solution which was at room temperature. The abrupt temperature change may have caused the disruption of a majority of the microfilaments and microtubules. An additional contributing factor to this disruption may have been the rapid exposure of the tissue to the osmium tetroxide. A gradual temperature reduction and a slow acclimatization of the tissue to osmium tetroxide may be the two most important facets of this procedure.

We believe that our fixation procedure can serve as a useful method for the preservation of microfilaments and microtubules in tissues other than the anterior pituitary. We would like to again caution anyone attempting to employ this method to be aware of the possible pitfalls associated with it and to avoid the possible disruption of these organelles due to uncontrolled osmolarity, abrupt temperature changes or shock from rapid exposure to osmium.

Figure 1. Rhesus monkey pituitary gland. Large aggregations of micro-filaments are running parallel to the cell membrane in an agranular cell. A microtubule (arrows) is noted in close association with a secretory granule in the granulated cell at the top. X 46,600.

Figure 2. Rat pituitary gland. Portion of the Golgi apparatus and adjacent areas of a somatotroph. Microtubules are indicated by the arrows. X 69,000.



THE USE OF THE ANALYTICAL ELECTRON MICROSCOPE IN THE IDENTIFICATION OF
CYTOCHEMICAL PRODUCTS IN THIN SECTIONS

Joe G. Wood

Elemental analysis by the use of energy dispersive X-ray techniques is beginning to gain in popularity. Much of the earlier work in this area, however, has been conducted toward the analysis of metals (1, 2, 3) with practical application in industry for some period of time (4). The development of increased sophistication in instrumentation, including increased sensitivity (5, 6) is now realizable. It was necessary in the past to conduct certain studies on *in vitro* products using electron probe analyses (7). However, since that time, the use of the AEM in thin sections has been the subject of a number of studies (8, 9, 10, 11) and the ability to analyze, as seen by transmission electron microscopy (TEM) (5, 6), presents a source for information which is relatively untapped. Thus, the analytical electron microscope (AEM) has practical applications to biological studies. This type instrumentation is relatively new but not difficult to assemble. Individuals are beginning to see the proper avenues in which this instrument can be used to gain new access to biological information.

Since electron optical systems can be used analytically, as well as photographically, specific reaction products can be incorporated into tissues and these products studied and identified within cell organelles. The object of this paper is to illustrate such an application showing the feasibility as to how these analytical procedures can be useful to other scientific investigators.

Tissues used in this study were those obtained from cat adrenal medulla

and monkey brain. The procedures used were those developed by Wood (12) and Wood and Matthews (13) for the detailed localization of biogenic amine reaction products. This method involves the incorporation of the heavy metal chromium (Cr). The mechanism of this reaction has been studied in detail (13). Studies in the past have shown an *in vitro* reaction product to contain Cr (7) and it seems likely that the specifically dense areas in biological tissues would also contain Cr. It also was clearly demonstrated that the JEOL Corporation and the KEVEX Corporation had produced a combination of instrumentation which can detect small amounts of Cr in tissue either in half micron or in 500Å sections. These sections make ordinary TEM photography an easy procedure.

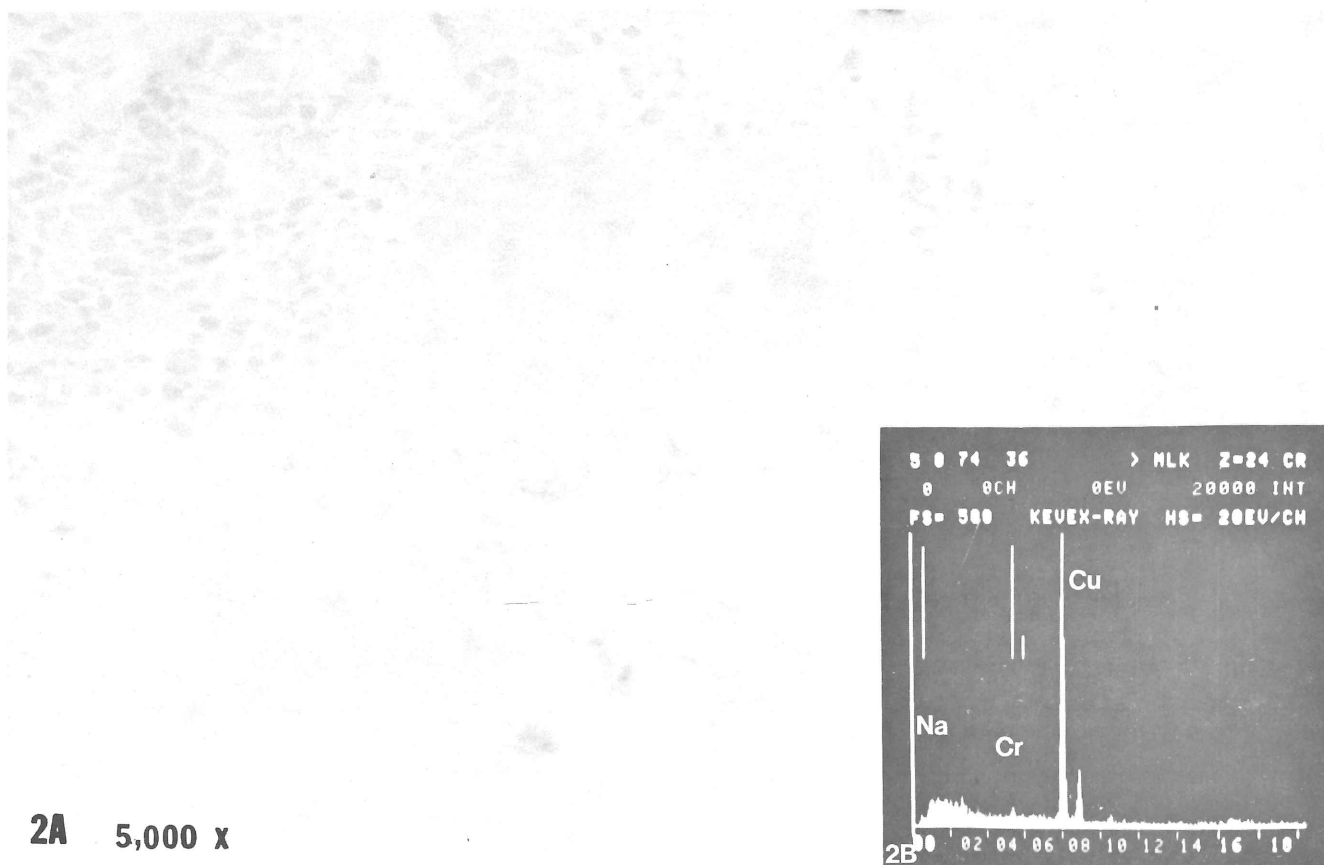
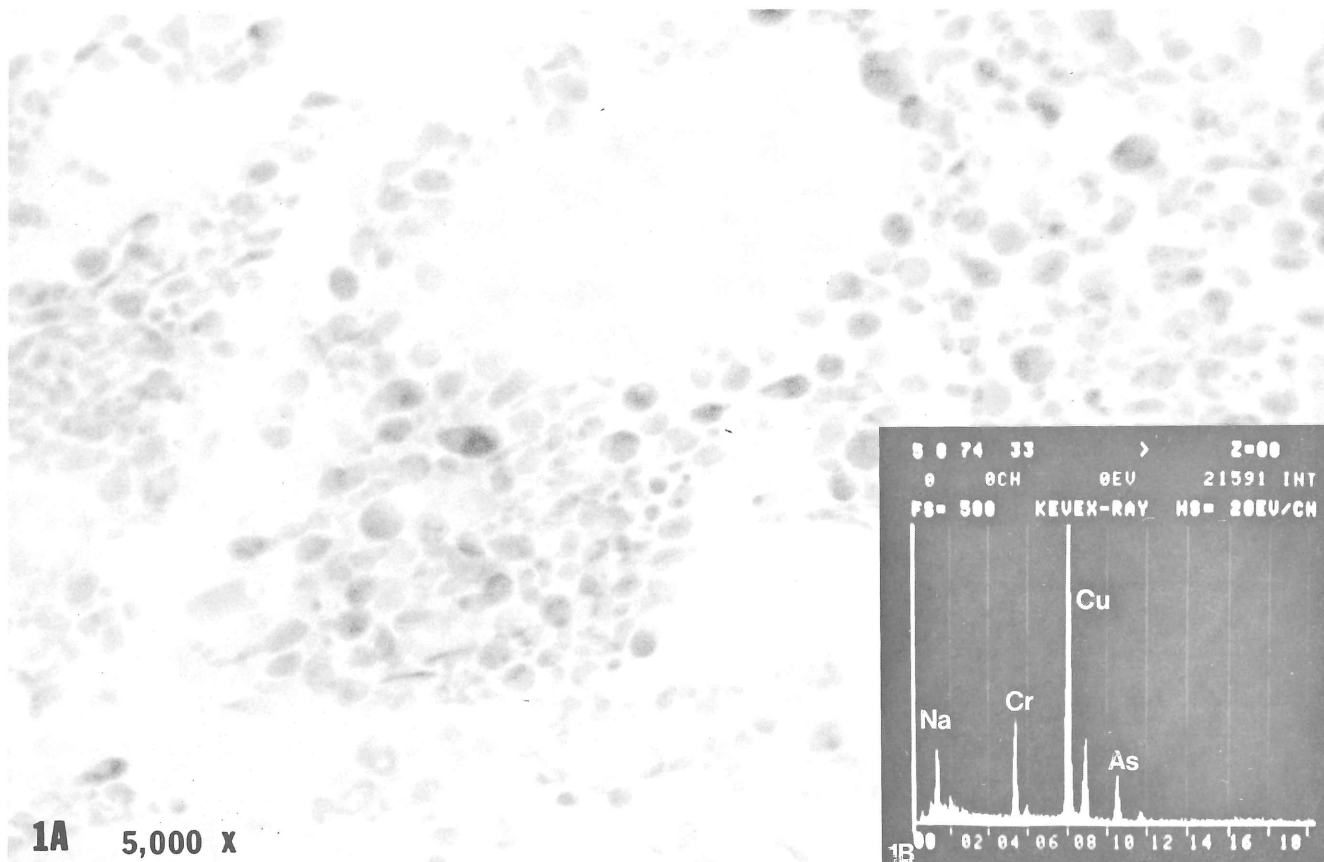
Tissues were mounted either on aluminum or copper grids placed in a JEOL 100B electron microscope with an attached KEVEX energy dispersive detector and analyser. The transmission beam used was 80KV at 40 micro-amps and had a diameter of approximately 2000Å. All counts were taken over a fixed period of time (200 seconds) and records were made either on photographic film by TEM for tissue, or taken by Polaroid photography directly from the cathode ray tube for energy dispersive analysis. Figure 1a shows cat adrenal medullary tissue with dense NE granules; this reaction has been established stoichiometrically (12) and the *in vitro* reaction product has been characterized (13). The energy dispersive analysis (Figure 1b) shows the Cr peak from these NE granules and indicates the amount of Cr present within that tissue. The other elements present in the tissue are arsenic (As) from the sodium cacodylate buffer and copper (Cu) from the grids. Other trace elements such as sodium and potassium are also present. The As and Cr are the induced materials with the Cr being the specific heavy metal incorporated which produces the electron density. Figure 2a is an epinephrine cell which is not

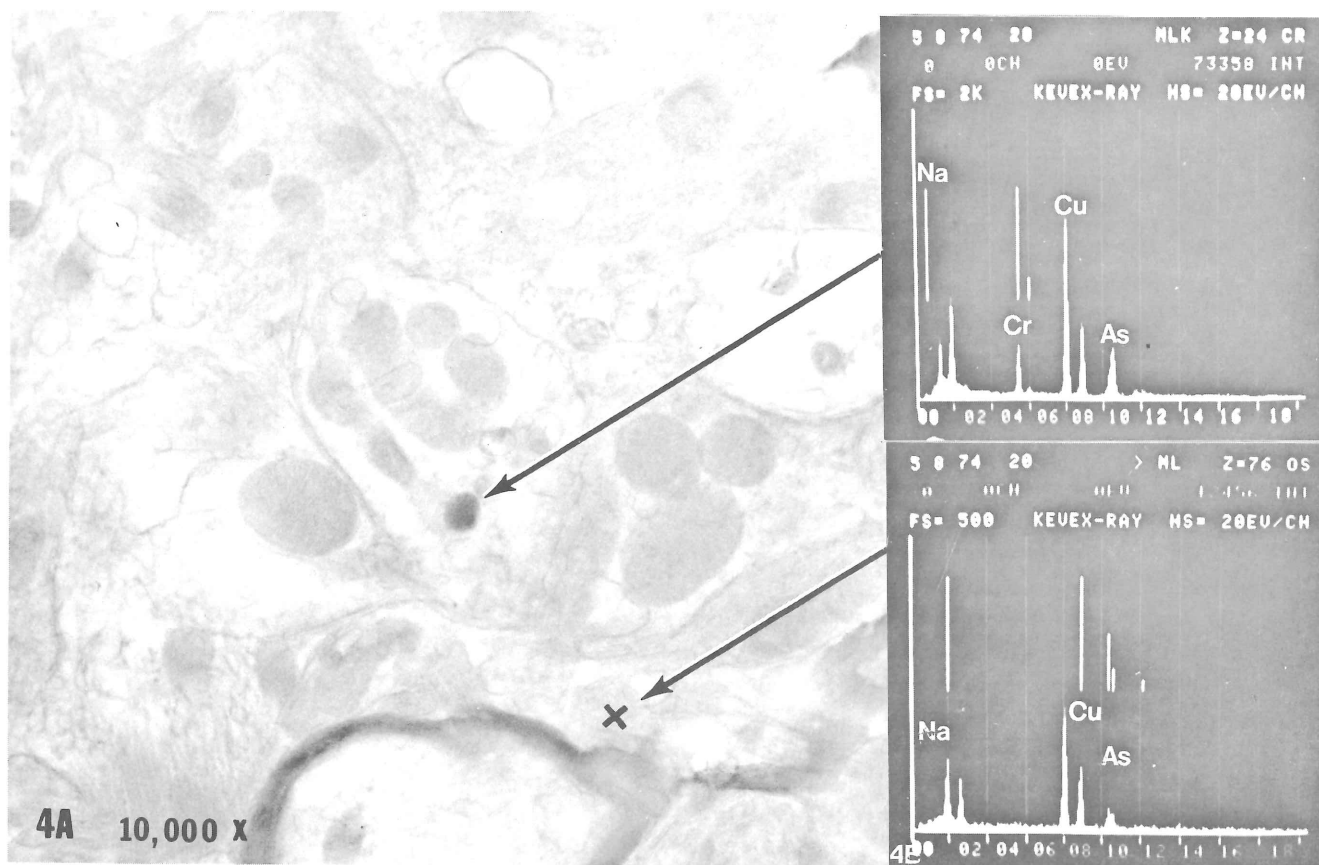
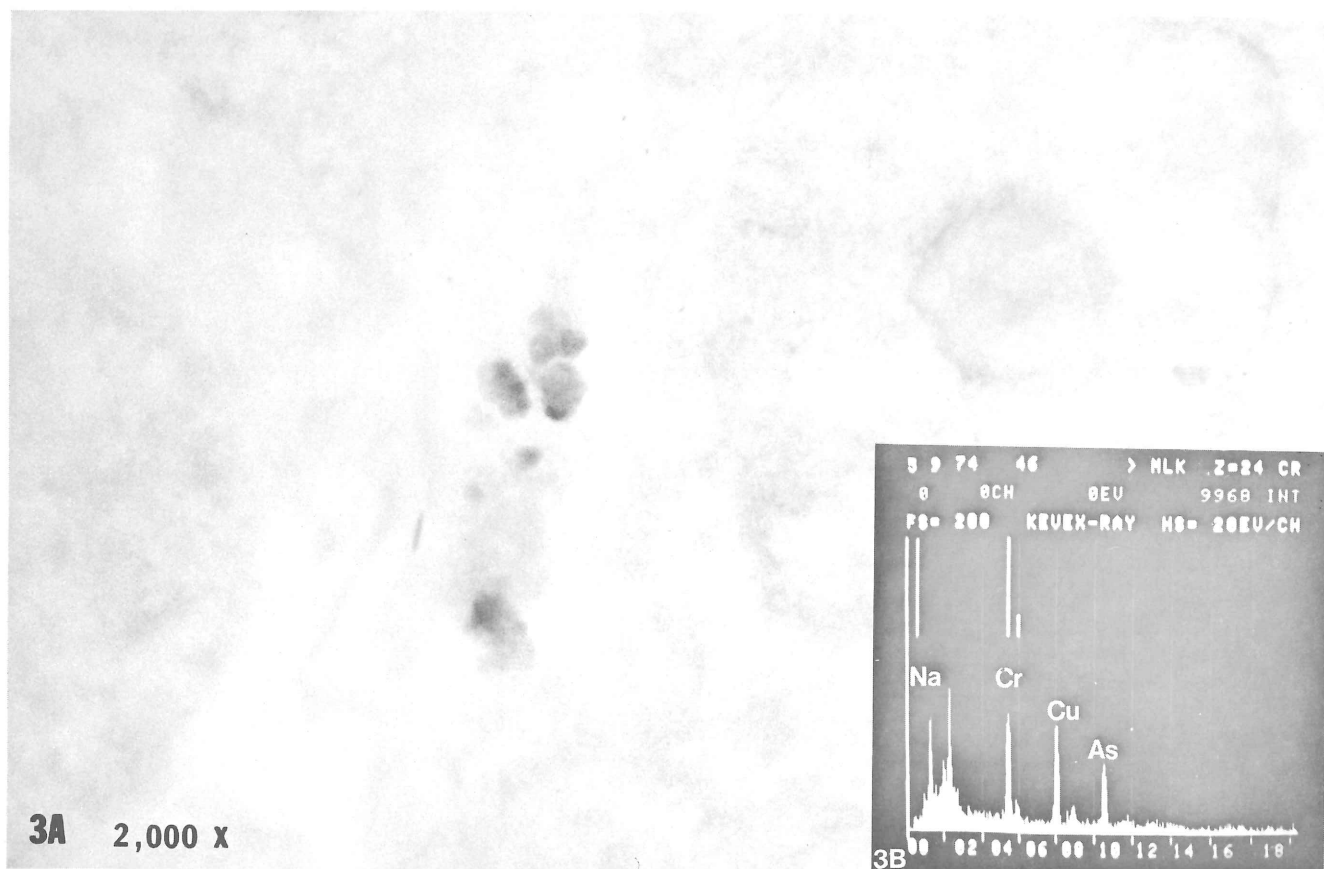
reactive for the cytochemical procedure. This is substantiated by Figure 2b (insert) which shows no significant Cr peak. Figure 3a is a TEM photograph of a biogenic amine positive neuron in the median eminence of the Rhesus monkey showing Cr positive reaction product in the cell body; Figure 3b shows the X-ray spectrum from this area. The identification of this type reaction product occurs in "lakes" in neurons of the arcuate nucleus. Fluorescence microscopy (14) has shown that these same neurons are biogenic amine positive. Figure 4a shows an osmicated axon which contains a positive Cr reaction product and the insert in Figure 4b shows the reading for Cr in this particular area. This insert (Figure 4b) shows the sensitivity of the AEM in analyzing these small structures. Figure 4c is a background count from a nonreactive area. Also, it should be pointed out that vesicles of this size could be readily studied in more detail by using the 20\AA scanning transmission (STEM) beam which was utilized in a previous study (6). The high degree of discrimination of this STEM type beam indicates exciting possibilities.

It is obvious from these comments and from recent studies that the AEM has considerable use, not only as a supportive agent to electron microscopy, but for its own qualitative, and in all probability, eventual quantitative procedures. This one technique which utilizes Cr in a reaction product has proven to be very useful in the specific identification of dense areas which are amine containing as opposed to those which contain either ferritin or osmium (15). It also seems possible that other cytochemical techniques utilizing metal reaction products such as the lead in the various phosphatase reactions (16) and the copper in the copper thiocholine technique (17) for cholinesterase can be studied by this procedure. Also, it seems highly possible that new cytochemical procedures can be developed utilizing metals which then provide a basis for tracing certain compounds, not only in the central nervous system, but in other organ systems of the body as well.

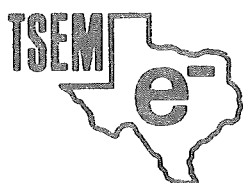
BIBLIOGRAPHY

1. Edington, J. W., Hibbert, G.: High resolution microanalysis of aluminum solid solution alloys using combined electron microscopy and energy analysis. *J. Microscopy* 99 (pt. 2): 125, 1973.
2. Fursey, Anita, Kent, B., Saunders, S. R. J.: Metal oxide systems studied by analytical electron microscopy. *J. Microscopy*, 99 (pt. 2): 147, 1973.
3. Lorimer, G. W., Razik, N. A., Cliff, G.: The use of the analytical electron microscope EMMA-4 to study the solute distribution in thin foils: Some applications to metals and minerals. *J. Microscopy*, 99 (pt. 2): 153, 1973.
4. Jacobs, M. H.: Industrial applications of analytical electron microscopy. *J. Microscopy*, 99 (pt. 2): 165, 1973.
5. Koike, H., Namae, T., Watabe, T., Mikajiri, A.: An approach to microanalysis with the electron microscope. *JEOL News*, 10E, No. 4, 1973.
6. Wood, Joe G., Harling D., *JEOL News*, in press.
7. Wood, Joe G., Seelig, L. L., Jr., Benjamin, C. P.: Cytochemistry of epinephrine and norepinephrine adrenomedullary cells. *Histochemie* 28: 183, 1971.
8. Oschman, J. L., Hall, T. A., Peters, P. D., Wall, B. J.: Association of calcium with membranes of squid giant axon. *J. Cell Biol.*, 61: 156, 1974.
9. Oschman, J. L., Hall, T. A., Peters, P. D., Wall, B. J.: Microprobe analysis of calcium deposits in squid axon. *J. Cell Biol.* 59 (2, pt. 2): 255a, 1974.
10. Oschman, J. L., Wall, B. L.: Calcium binding to intestinal membranes. *J. Cell Biol.*, 55: 58, 1973.
11. Hall, T. A., Anderson, H. C., Appleton, T.: The use of thin specimens for X-ray microanalysis in biology. *J. Microscopy* 99 (pt. 2): 177, 1973.
12. Wood, Joe G.: Electron microscopic localization of amines in central nervous tissue. *Nature (London)* 209: 1131, 1966.
13. Wood, Joe G., Matthews, H. R.: Selective metal reactions for biogenic amines, *J. Cell Biol.*, 59: 368, 1973.
14. Jonsson, G., Fuxe, K., Hökfelt, T.: On the catecholamine innervation of the hypothalamus, with special reference to the median eminence. *Brain Res.* 40: 271, 1972.
15. Wood, Joe G.: Histochemistry (in press). Use of the analytical electron microscope (AEM) in cytochemical studies of the central nervous system.
16. Wachstein, M. and Meisel, E.: Histochemistry of hepatic phosphatases at a physiologic pH. *Am. J. Clin. Path.* 27: 13, 1957.
17. Karnovsky, M. J. and Roots, L.: A "direct-coloring" thiocholine method for choline esterases. *J. Histochem. Cytochem.* 12: 219, 1964.





The author wishes to acknowledge the very able technical assistance of Jane Crick and the assistance and support of David Harling and JEOL, and of Carl Freeman and KEVEX. Supported by grant NS-10326.



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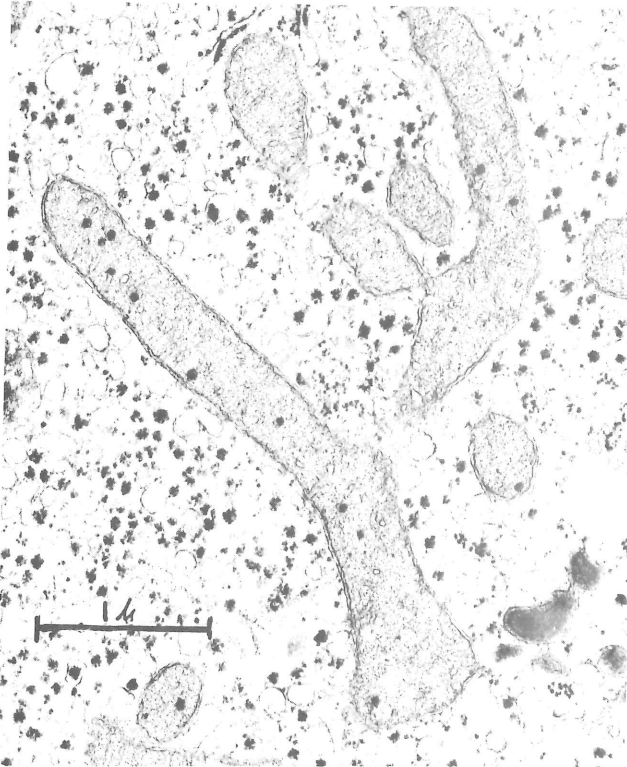
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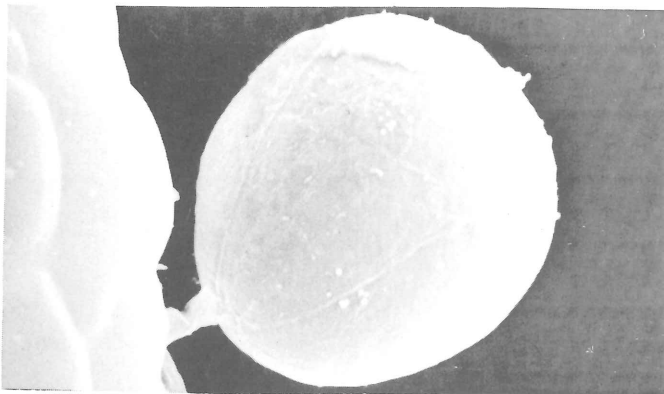
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CONTRIBUTED MICROGRAPHS

Cover micrograph- "Shades on a Cool Sperm!" An unusual section through a human sperm head. Dr. J. D. Berlin, Dept. of Biological Sciences, Texas Tech University, Lubbock, Texas.

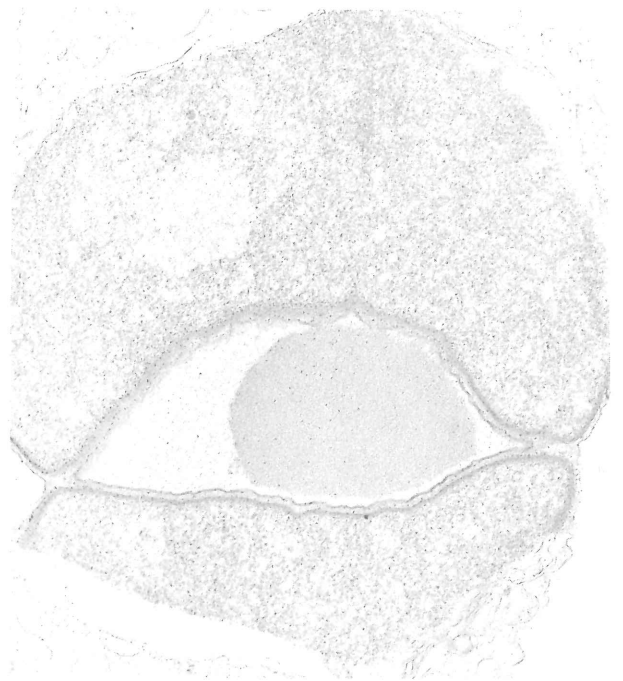


"Is it the eye of Texas? or the morning after a TSEM social hour?" Abnormal acrosomal formation in human spermiogenesis. 37,000X. Fannie E. Smith, Dept. of Biological Sciences, Texas Tech University, Lubbock, Texas.



A salt bladder on the leaf surface of Atroplex halimus. 500X. Franklin Bailey, Dept. of Biological Sciences, Texas Tech University, Lubbock, Tex.

Branching liver mitochondria associated with a hepatoma in a comatose 15 year old boy. 23,200X. Alan B. Weckerling, Dept. of Pathology, Brooke Army Medical Center, Ft. Sam Houston, Tex.



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AREA NEWS

Austin

THE UNIVERSITY OF TEXAS AT AUSTIN:

Department of Biology

New Members:

Mr. W. A. Calley (Graduate Student), Ms. Debbie Bulmer (Technician).

New Equipment:

Balzers EVM052 Electron Beam Evaporation Device for Freeze-etch unit; Sony VideoRecord System for SEM.

New Grants:

Pan Am Health Organization Grant; NSF for Fungal Ultrastructure.

Seminars Given:

Fungal Ultrastructure presented at Geneva, Switzerland and Nancy, France by G. T. Cole.

Recent Publications:

Cole, G. T. : Ultrastructure of vegetative hyphae in hyphomycetous fungi protoplasma, In press.

Cole, G. T. : Conidiophore and conidium ontogeny in Spegazzinia tessarthra. Can. J. Bot. 52: 1259-1264.

Cole, Hardcastle and Szanislo: Subbaromyces splendens: Ultrastructure and development. Can. J. Bot., In press.

Fort Sam Houston

BROOKE ARMY MEDICAL CENTER:

U. S. Army Institute of Surgical Research

Recent Publications:

Nash, D. A., Rogers, P. W., Langlinais, P. C., and Bunn, Jr., S. M.: Diabetic glomerulosclerosis without glucose intolerance. Am. J. Med., In press.

Houston

BAYLOR COLLEGE OF MEDICINE:

Department of Cell Biophysics

Grants:

"Analysis of Z Bands in Heart by Optical Diffraction" three year grant from NHLI to Margaret A. Goldstein.

NHLI Research and Demonstration Center five year grant to Michael E. DeBakey. Project entitled "Ultrastructure and Cytochemistry of Growth and Development of Normal and Diseased Heart: Role of Z Substance" by Margaret A. Goldstein.

"A Cytological Probe for $\text{Na}^+ - \text{K}^+$ ATPase" one year grant to Barry VanWinkle from American Heart Association-Texas Affiliate.

Department of Microbiology

Dr. Heather D. Mayor is on a lecture tour in Australia. The tour includes Sidney, Cairnes, Canberra and Melbourne. Dr. Mayor's lecture topics are: Virus and Cancer, Parnoviruses and Adeno and Satellitevirus.

L. E. Jordan from Dr. Mayor's group attended the recent EMSA meeting in St. Louis.

Department of Pathology

New Equipment:

Huxley ultramicrotome; Varian vacuum evaporator.

Visitors:

Dr. Mattie Bossart from Dominican College in Houston has joined Marilyn N. Smith's lab for the summer. Dr. Bossart in conducting ultrastructural studies on testicular infertility.

The Department of Pathology was represented at the recent EMSA meeting in St. Louis by Dr. Ronald F. Dodson, Marilyn N. Smith, Margie Harness and Lena Wai-Fong Chu. Mrs. Smith chaired the first section of Pathology and presented a paper entitled "Ultrastructure of Bronchiolar Origin." This paper was co-authored by Marilyn N. Smith, S. Donald Greenberg, and Harlan J. Spjut.

Recent Publications:

Klima, M., Smith, M., Spjut, H. J., Root, E. N.:
Malignant mesenchymoma: Case report with electron microscopic study.
Cancer. . In press.

Levy, M. L., Greenberg, S. D., and Harness, M. K.:
Ultrastrucutre of undifferented small ("oat") cell carcinoma of lung. Abstract
of presentation at Texas Medical Association, Houston, Texas, May 9-12,
1974.

Harness, M. K. and Greenberg, S. D.: Ultrastructure of
bronchial carcinoids. Abstract of presentation at Texas Medical Association,
Houston, Texas, May 9-12, 1974.

Greenberg, S. D. and Harness, M. K.: The "shock lung":
An ultrastructural study. Abstract of presentation at Texas Medical Association,
Houston, Texas, May 9-12, 1974.

Ross, J. N., Brown, C., Harness, M. K., Greenberg,
S. D., Tacker, M., and Kennedy, J. H.: The role of platelet aggregation
in prolonged extracorporeal respiratory support, In press.

Departments of Neurology and Pathology and the Baylor-Methodist Center for Cerebrovascular Research

Dr. Ronald F. Dodson was Chairman of the section entitled
Pathology III and Biological Techniques II at the recent EMSA meeting in
St. Louis. Dr. Dodson presented a paper entitled "Ependymal response in
acute cerebral infarction" which was co-authored by Dr. Yukio Tagashira.
Mrs. Chu also presented a paper which was entitled "Myelinated fiber response
in grey matter following acute cerebral infarction." Her co-authors were
Drs. Dodson and Tagashira.

Recent Publications:

Tulleken, C. A. F., Meyer, J. S., Ott, E. O., Abraham,
J., and Dodson, R. F.: Brain tissue pressure gradients in experimental
infarction recorded by multiple wick-type transducers. Abstract of presentation
at the Second International Symposium on Intracranial Pressure, Lund, Sweden,
June 17-19, 1974.

Dodson, R. F., Kawamura, Y.: Perivascular hemorrhagic lesions in temporal cortex following cerebral infarction (A morphological study). Exptl. Mol. Pathol. 20: 24-32, 1974.

Dodson, R. F., Kawamura, Y., Aoyagi, M., Hartmann, A.: A comparative evaluation of the ultrastructural changes following induced cerebral infarction in the squirrel monkey and baboon. Cytobios 8: 175-182, 1973.

Dodson, R. F.: Ultrastructural alterations of myelinated fibers following acute cerebral infarction in the squirrel monkey. Medikon, 19-21, June, 1974.

Dodson, R. F., Meyer, J. S., Aoyagi, M., Hartmann, A.: Acute cerebral infarction and hypotension: An ultrastructural study. Abstract read before the 50th Annual Meeting of the American Association of Neuropathologists, Boston, Mass. June 7-9, 1974.

Dodson, R. F., Aoyagi, M., Hartmann, A., Tagashira, Y.: Acute cerebral infarction and hypotension: An ultrastructural study. J. of Neuropath. and Exptl. Neurol. 33: (3) 400-407, 1974.

RICE UNIVERSITY:

M. Lea Rudee, who with Bill Philpott was one of the co-founders of TSEM, will be leaving Rice to become Provost of the new Fourth College and Professor of Applied Physics at the University of California, San Diego.

THE UNIVERSITY OF TEXAS MEDICAL SCHOOL AT HOUSTON:

The Program in Neurostructure and Function

The Program in Neurostructure and Function at UTMSH has a new staff member, Dr. Dianna Redburn, who will be an Assistant Professor, from the Department of Psychobiology at the University of California at Irvine. She completed her Ph. D. work at the University of Kansas.

Grants:

Dr. R. G. Peterson recently received a research support grant from the University of Texas Medical School for continuing work on the ultrastructure of myelin. Chuck Sea, a medical student doing his preceptorship with Dr. Peterson, was one of four students selected to present his research project "Ultrastructure and Biochemistry of Myelin after Isonazid-Induced Nerve Degeneration in Rats", to the medical students and faculty of UTMSH.

Dr. J. G. Wood is continuing research on the Cytochemical Investigations of Adrenergic Neurons funded by a grant of \$49,000 from H. E. W. He was speaker this spring to the Biology Department at Sam Houston University.

F. David Prentice is a medical student at UTMSH, who in conjunction with Dr. J. G. Wood had a paper on "Cytochemical Localization of 5-Hydroxydopamine in Adrenergic Elements of Cat Adrenal Medulla" published in *Experientia*.

Papers published recently by Dr. Wood include, "Positive Identification of Intracellular Biogenic Amine Reaction Product with Electron Microscopic X-ray Analysis", for the *Journal of Histochemistry and Cytochemistry*, and "Analytical Electron Microscopy (AEM) of Specific Cytochemical Reaction Products" for *JEOL News*.

A graduate student in Neurobiology, Bob McClung, completed his Master's Degree on "Evoked Responses and Single Units Recording from the Rat Pineal."

Margaret E. Bell, also a student, is doing her dissertation work on "Turnover Rates of Peripheral Nervous System Myelin Proteins" in the laboratory of Dr. R. G. Peterson.

Recent guest lecturers to the department include Dr. Richard Hammerschlag, who spoke on "Functional Neurochemistry"; Dr. Daniel Louie on "Biochemical Composition of Brain"; Dr. George Buletza on "Dynamics of the Cerebrospinal Fluid"; and Dr. Dianna Redburn presented a seminar on "Neurotransmitter Release in vitro: A New Approach."

New equipment purchased by the department includes a Fiske osmotic-automatic osmometer, a Zeiss fluorescent microscope, an infrared spectrophotometer, and a Metrohm/Brinkmann pH meter.

The Program in Pathology

The program in Pathology at UTMSH has a new Chairman, Donald C. Cannon, M. D., Ph. D., from Bio-Sciences Laboratory, Van Nuys, California.

Dr. Alan Broughton, who joined the staff in November, was speaker at the Texas Society of Clinical Chemistry meeting in Waco, Texas, in April.

The program in Pathology also welcomes Dr. Enrique VanSanten from Chihuahua, Mexico.

Huntsville

SAM HOUSTON STATE UNIVERSITY:

Department of Biology

Grants:

Philosophical Society Grant for Spermatheca Studies

Recent Publications:

Intranuclear and Cytoplasmic Annulate Lamellae in Grasshopper Spermatocytes (genus Melanoplus) in Cell and Tissue Research (In Press).

Louisiana

LOUISIANA SOCIETY FOR ELECTRON MICROSCOPY:

News from the Louisiana Neighbors

Dr. Robert D. Yates, Past-TSEM President and present Tulane Anatomy Chairman, is recovering nicely from a lumbar laminectomy, but is now minus the L₄, L₅, and L₅S₁ discs. Also, Bob has been selected as the Asst. Program Chairman for EMSA in 1975 (Las Vegas) and will become Program Chairman in 1976 (Miami).

Tulane Anatomy announces the following Distinguished Scientist Lecturers for the 1974-75 academic year: Drs. F. Sjostrand, Carmine Clemente, Daniel Pease, George Palade, Marilyn Farquhar, J. D. Robertson, Sanford Palay, and Don Fawcett. All of the scientific community is invited to hear these outstanding individuals lecture in their field.

Elizabeth Mary Donnell (formerly from Fort Worth) recently changed from EM to dermal pathology.

LSEM will hold its Fall Symposium on Friday, November 8, at Tulane University's Delta Regional Primate Center in Covington, La. (across lake from New Orleans).

The following publications are noted:

"Innervation of abdominal paraganglia." J. Morph., 142: 153-163, 1974, by Mascorro and Yates.

"The histology and ultrastructure of cat abdominal paraganglia after fixation and localization with glutaraldehyde/potassium dichromate." EMSA Proceedings, 1974, 290-291, by Mascorro and Yates.

"Glutaraldehyde perfusion followed by glutaraldehyde/potassium dichromate immersion: A technique for localizing paraganglia." Tex. Rep. Biol. Med., In Press, 1974, by Mascorro, Yates, and Chen.

"Morphological comparisons between aortic and carotid glomus cells in the rabbit." Tex. Rep. Biol. Med., In Press, 1974, by John T. Hansen, I-Li Chen, and Robert D. Yates.

Dr. G. W. (Bill) Bailey from Esso Research Laboratories in Baton Rouge has been appointed as Editor of the EMSA Proceedings.

Congratulations to former EMSA Proceedings Editor, Claude Arceneaux, for long and meritorious service rendered to EMSA. Claude is so dedicated that he will continue to serve as Associate Editor.

Lubbock

TEXAS TECH UNIVERSITY:

Department of Biological Sciences

New Faces:

Bill McCombs, from Scott and White in Temple, is a new doctoral student; Dr. Tom Pizzolato, formerly of the University of Miami (Ohio) is a new postdoctorate.

Grants:

The Initiation and Development of Cotton Fibers, renewal funded by Cotton Incorporated.

Seminars:

Seminars were given by Dr. Timothy Fitzharris and Dr. Tom Brady.

TEXAS TECH SCHOOL OF MEDICINE:

Department of Anatomy

New Members of the Anatomy Faculty:

Dr. John Yee, from University of Utah; Dr. Burnell Dalley, from University of Nebraska; Dr. Donald Wilbur, from Medical University of South Carolina.

Randy Brackeen is the new Director of Anatomy Laboratories of TTUSM (from Galveston, UTMB).

New Instruments:

Zeiss EM 10 Transmission Electron Microscope

Grants:

Dr. Patrick Sterrett, Contrast Materials and the Blood-Brain-Barrier. National Institute of Stroke and Neurological Diseases.

Papers Accepted for Publication:

Sterrett, P. R., A. M. Thompson, A. L. Chapman and H. A. Matzke: The effects of hyperosmolarity on the blood-brain-barrier. A morphological and physiological correlation. Brain Research 77: 1-15, 1974.

San Antonio

THE UNIVERSITY OF TEXAS HEALTH SCIENCE CENTER
AT SAN ANTONIO:

Department of Anatomy

Drs. E. K. Adrian and I. L. Cameron have recently been promoted to full Professors.

Dr. Cameron has been selected as a member of the Advisory Committee on Cell Growth Regulators in Cancer Chemotherapy for the National Cancer Institute.

Publications:

Bowie, E. Phylis, Glenn Williams, Masataka Shiino, and Edward G. Rennels: The corticotrop of the rat adenohypophyses: a comparative study. Am. J. Anat. 138: 499-520, 1973.

Bowie, E. Phylis, M. Glenn Williams and Edward G. Rennels: Evidence for PAS positive reaction of the corticotrop granules of the rat adenohypophysis. Histochemistry 38: 281-284, 1974.

Two books have been recently coedited by Dr. Cameron, Cell Cycle Controls and Drugs and the Cell Cycle for Academic Press.

Herbert, Damon C.: Histological identification and immunochemical studies of prolactin and growth hormone in the primate pituitary gland. Gen. Comp. Endocrinol. (in press).

Richardson, Lyn: Richardson's modified southgate's muricarmine stain compared to mayer's muricarmine. Am. J. of Med. Tech. 40: 207-210, 1974.

Richardson, Lyn: Richardson's reconditioning and sharpening technique for microtome knives. Am. J. of Med. Tech. 40: 371-372, 1974.

Shiino, Masataka and Edward G. Rennels: Paracrystalline aggregates of microtubules in the anterior pituitary cells of the Chinchilla (Chinchilla laniger). Am. J. of Anatomy 139: 135-140, 1973.

Warchol, J. B., D. C. Herbert and E. G. Rennels: An improved fixation procedure for microtubules and microfilaments in cells of the anterior pituitary gland. Am. J. of Anatomy (in press).

Winborn, W. B., L. L. Seelig, Jr., and C. M. Girard: Variation in the pattern of carbonic anhydrase activity in the cells of the gastric glands. Histochemistry 39: 289-300, 1974.

Winborn, W. B., and D. L. Guerrero: The use of a single tissue specimen for both transmission and scanning electron microscopy. Cytobios. (in press).

Departments of Physiology and Pathology

The departments of Physiology and Pathology have obtained jointly a new Philips 301 electron microscope.

Temple

SCOTT AND WHITE MEMORIAL HOSPITAL:

Robert A. Turner, Immediate Past President of TSEM, was recently appointed as interim affiliates representative on the Executive Council of EMSA at the August meeting in St. Louis. This appointment holds the rank of Director. At the February EMSA Council meeting the position is expected to be formalized by the Council, and Turner will begin serving a two year term.

Turner's duties will be to coordinate the activities and correspondence of the state and local affiliate EM societies. He will serve for two years after which the appointment, made by a vote of the affiliate officers, must pass to another affiliate.

New members to TSEM:

Dr. Don Jutzy; Dr. A. K. Brown; Dr. Louis Dieterman;
Dr. Allen Jay; Mr. Kenneth Mazur; Mrs. Elaine McCoy.

Visitors:

Dr. Gerald Beathard, M. D. , Ph. D. , Department of Pathology, Galveston: lecture entitled "Latest in Nephrotic Syndrome Studies. "

Abstracts of Papers
presented at the
Fall 1974 Meeting of the
TEXAS SOCIETY FOR ELECTRON MICROSCOPY
at Waterwood
Huntsville, Texas
October 4-6, 1974

THE CLINICAL ELECTRON MICROSCOPE, B. E. P. Beeston, AEI Scientific Apparatus Inc., 500 Executive Boulevard, Elmsford, New York 10523.

It is frequently felt among clinical pathologists that reliance entirely upon the light microscope for the microscopical examination of biopsy material for the purpose of diagnosis, prognostication and evaluation of therapeutic treatments is rather less than satisfactory, and the additional use of the electron microscope for more accurate diagnoses and confirmations is strongly advised.

In the light of this situation it is expedient to analyze the requirements necessary to optimize the design of the electron microscope for clinical investigations in order to further the application of the instrument in this vital field. These requirements are:

1. Large field of view for low magnification survey work.
2. A magnification range overlapping that of the light microscope.
3. A high contrast, high definition image.
4. Multiple specimen facilities for rapid throughput and comparison work.
5. Large screen viewing and ample means for indicating unambiguously to colleagues important features of the images.
6. Ease of operation and instrument reliability.

All these features are now available in a new microscope, called the Clinical Corinth.

ACID PHOSPHATASE ACTIVITY IN DEVELOPING SPERMATID OF CHINESE HAMSTER
Jeffrey P. Chang, Hiroshi Mayohara and B. R. Brinkley. Division of Cell Biology, The University of Texas Medical Branch, Galveston, Texas.

Seminiferous tubules of the normal mature Chinese hamsters were dissected into 2% glutaraldehyde in 0.1M cacodylate buffer at pH 7.4 at 4° and were halved longitudinally for fixation. The Gomori's lead technique was used for demonstration of acid phosphatase (ACPase).

For the first time, we have recorded stepwise localization of reaction product of ACPase activity in various early stages of acrosome formation during Golgi phase of spermiogenesis. The enzyme apparently appeared first in the Golgi complex of the spermatocytes. It was subsequently incorporated and concentrated into the acrosomal granule but later became non-detectable cytochemically when the acrosomal cap was formed. Concurrently reduction and diminution of ACPase was also observed in the Golgi apparatus following its caudal migration to the posterior end of the spermatid. The results were consistent in our repeated observations.

It may be assumed that ACPase actually diminished in acrosome after Golgi phase or that the amount of the enzyme present was below the sensitivity of the cytochemical determination. It is also possible that ACPase has been masked by some substance and became nonreactive to histochemical reaction. Finally, the acrosomal membrane had changed its properties and caused an inaccessibility between the enzyme and the substrate. (Supported by a Research Contract, NIH-NICHD-69-2139, from the USPHS.)

ADRENAL MEDULLARY CHANGES DURING CHRONIC ETHANOL INTOXICATION AND FOLLOWING WITHDRAWAL. D. K. Bynum*, J. G. Wood, H. R. Matthews*. University of Texas Health Science Center, Houston, Texas.

Activation of the peripheral sympathetic nervous system and the adrenal medulla have been demonstrated during chronic ethanol intoxication and following acute withdrawal. A study was carried out to determine the effects of chronic ethanol intoxication and acute withdrawal on the storage of catecholamines (CAM) in the adrenal medulla using cytochemical and biochemical techniques. Mice were maintained in a continuously intoxicated state for a period of 72 hr using a vapor chamber. Norepinephrine (NE) and epinephrine (E) levels were determined fluorometrically, while changes in the storage of CAMs as well as associated ultrastructural changes were studied by transmission EM using specific heavy metal cytochemical reagents. Animals were sacrificed and tissues obtained every 24 hours during intoxication and every 6 hours after withdrawal. Tissues were fixed in 4% glutaraldehyde plus 1% sodium molybdate (pH 7.2) followed by incubation in 2.5% potassium dichromate (pH 4.1) both solutions in 0.2M sodium cacodylate buffer. Results indicate that during ethanol intoxication there is an apparent increase in NE content in the adrenal medulla which is manifest by an increase in the number, size and density of NE containing granules. Following withdrawal a marked decrease in NE content occurs. This decrease is most noticeable 13 hours after withdrawal and correlates temporally with the occurrence of the most marked symptoms of the withdrawal syndrome. Twenty-four hours after withdrawal, NE content appears to have returned to normal. This transient increase in NE content during intoxication followed by a decrease during withdrawal correlates with known neurochemical events. Thus the adrenal medulla may serve as an adequate model for the study of the effects of ethanol on noradrenergic neurons. Support from NS-10326 and AA-00975.

ER PROLIFERATION AND DICYTOSOMAL ACTIVITY IN HYPOXEROUS FUNGI. Garry T. Cole, Department of Botany, University of Texas at Austin.

Stacked arrangements of smooth endoplasmic reticulum and short, swollen cisternae in vegetative hyphae of the imperfect fungus, *Trichia sorokhiana* are examined. Similar ER complexes in plant and animal cells have been reported and ascribed several different functions, one of which is a dicytosomal-like activity. However, evidence is weighted against the likelihood of the laminated SER in this fungus having such a function. Suggested functions of the ER in *T. sorokhiana* are discussed and compared to the probable secretory activities of apical vesicles which accumulate at the tip of vegetative hyphae.

INTRAMITOCHONDRIAL GRANULES IN ISOLATED MUSCLE BEFORE AND AFTER ACUTE ANOXIA. Margaret A. Goldstein, David L. Murphy, Per T. Thyrum and James H. Martin. Dept. of Cell Biophysics, Baylor College of Medicine, Houston, Tx. and *Dept. of Pathology, Baylor Medical Center, Dallas, Tx.

Left posterior papillary muscles (diam. < 1mm) from guinea pigs were bathed in Tyrode's (95% O_2 -5% CO_2), held at 0.15 gms. tension and stimulated at a rate of 1.0 Hz. After 1 hour, 5 controls were bathed 1/2 hr. in Tyrode's (95% O_2 -5% CO_2), and 7 experimentals were bathed 1/2 hr. in Tyrode's (95% N_2 -5% CO_2). A rapid decline in developed tension was observed during early hypoxia but at 30 min., the activity stabilized at 28% of pre-hypoxia value. At this time muscles still held isometrically were bathed in 2% glutaraldehyde in Tyrode's ($+O_2$ or $+N_2$) for 1 hr. and processed for EM. Controls had normal morphology, resembling that of intact muscles fixed in situ, except for the more frequent presence of intramitochondrial granules (IG). IG were more numerous in control muscles isolated for longer time periods (1 1/2-2 1/2 hrs.). Cross sections of the entire muscle showed that cells at the periphery contained more IG than those in the interior. Mitochondria in anoxic muscles were swollen and seldom contained IG. Unstained sections of post-oximicated control and anoxic muscle containing IG were examined. Electron-dense IG were not removed by flotation on 2% HCOOH, 2 mM EDTA or 2mM EGTA alone. IG were removed by 5% HIO_4 or 2% H_2O_2 followed by 2mM EDTA and by HIO_4 or H_2O_2 alone. IG contained some residual material after treatment with HIO_4 which stained with uranyl acetate and lead citrate. Inorganic material in IG was not seen after microincineration at 500°C. The increased number of IG with prolonged exposure to O_2 and decreased number after anoxia may be related to respiratory and energy dependent calcium movements into heart mitochondria. IG may serve as organic precursors for calcium-containing granules.

ABORTIVE GROWTH OF WILD STRAINS OF INFLUENZA B IN HUMAN LUNG CELLS by Cameron E. McCoy, William B. McCombs and Albert Leibovitz. Scott and White Clinic, Temple, Texas.

Attempts were made during the influenza B outbreak in February-March, 1974, to isolate this agent in human embryonic lung fibroblasts. Although no evidence of viral activity could be detected by cytopathogenicity or hemadsorption tests, electron microscopy revealed that the wild virus proliferated in the lung cells on primary inoculation. Infected cells exhibited significant changes in both the nucleus and cytoplasm. Aggregated chromatin and dispersed nucleoli were characteristic of the nucleus. Amorphous inclusion bodies resembling distended endoplasmic reticulum proliferated in the cytoplasm. These inclusions gave way to filamentous particles that ultimately filled the entire cytoplasmic area.

The budding virions appeared as numerous pleomorphic, villous projections along the thickened plasma membrane. The lack of internal structure in many of these indicated that essentially non-infective virus was being produced which explains the failure of subcultures.

The abortive growth cycle of influenza B is similar to studies attempting to grow Newcastle disease virus in mouse "L" cells.

OPTICAL DIFFRACTION ANALYSIS OF Z BANDS IN HEART MUSCLE. Margaret A. Goldstein, John P. Schroeder* and Ronald L. Sass* Depts. of Cell Biophysics and Medicine, Baylor College of Medicine, *Depts. of Chemistry and Physics, Rice University, Houston, Tx. 77025.

Anomalous Z bands in mammalian muscle, which we call Z crystals, have been observed in a variety of physiological and pathological states. The Z crystals resemble Z bands in that they have the same electron density, they are often continuous with adjacent Z bands of normal width and they bind anti- α -actinin. Using an optical diffractometer on electron micrographs, we have tested the assumption that Z crystals are chemically and structurally related to Z bands. Diffraction patterns from electron micrographs of both longitudinal and cross sections of normal and anomalous cardiac Z bands were compared. Diffraction patterns generated from cross sections of Z bands and Z crystals were identical. A face-centered rectangular lattice, containing a rhombic lattice ($\sigma = 75^\circ$) as well, was predicted from the diffraction data of cross sections. The model unit cell has the dimensions of $172\text{Å} \times 240\text{Å} \times 395\text{Å}$. Optical diffraction analysis has provided structural and chemical information regarding the substructure of the cardiac Z lattice that is not obtained by visual examination of electron micrographs. The shape and dimensions of the lattice suggest a structural unit common to both Z crystal and Z band.

CONFIGURATIONAL CHANGES IN THE MITOTIC APPARATUS OF MECHANICALLY COLLECTED CELLS TREATED WITH COLCEMID. Monley McGill and D. P. Highfield, Division of Cell Biology, Department of Human Biological Chemistry and Genetics, The University of Texas Medical Branch, Galveston, Texas 77550.

Populations of mitotic cells were collected by shaking confluent monolayers of Chinese hamster ovary (CHO) cells at 10-min intervals and pooling the "mitotic shakes" at 4°C. Eighty-five to 93% of the cells collected by this method were in mitosis. The distribution of mitotic configurations in these cells included 80% metaphase and anaphase cells and 20% telophase cells. Following resuspension of these cells in fresh medium and reincubation at 37° for 30 min, 90% of all mitotic cells were in telophase. Duplicate collections of mitotic cells pooled in the cold were reincubated at 37° for various time periods in fresh medium containing 0.06 $\mu\text{g}/\text{ml}$ Colcemid. After 30 min reincubation in Colcemid, a significant drop in the mitotic index indicated anaphase and telophase cells continued through mitosis into G₁. However, all cells in metaphase remained blocked. By light microscopy it appeared that Colcemid, applied to metaphase cells recovering from cold shock, induced C-metaphase-like configurations around both poles of the mitotic apparatus in some cells while other cells appeared to be blocked in normal metaphase. These results suggest that the arrangement of the mitotic apparatus in the presence of Colcemid, at metaphase, may be dependent on the degree of pole separation at the time of cold storage. An ultrastructural examination of these mitotic configurations is in progress and will also be reported. (This study was supported by Research Grant DHEW 5R01 CA 14675 from the National Cancer Institute.)

PARASITISM OF MALLOPHAGA BY TREMONYCES HISTOPHORUS. Shirlee M. Neale and Joyce A. DeVaney, Veterinary Toxicology and Entomology Research Laboratory, ARS, USDA, College Station, Texas 77840.

Little interest has been shown in the fungi of the order Laboulbeniales since the majority of these ectoparasites of arthropods do not penetrate beyond the integument of their host and therefore do little or no damage to their host. Four species of Mallophaga cultured in our laboratory were found to be parasitized by this fungus. SEM studies showed that the fungus grew on all three body segments of the lice, as well as on the appendages, but the reproductive structures only emerged from the cuticle of the host at the intersegmental membranes. Both the glycerine technique developed by Allison, et al, 1972 (Jour. Parasitol. 58(2): 414-416) and the critical point techniques for preparing tissue were used in order to determine the best method of preserving the delicate perithecium.

EFFECT OF TRYPSIN ON INTRAMEMBRANOUS PARTICLE DISTRIBUTION Randy L. Moses and John J. Bieseke, Department of Zoology, University of Texas, Austin, Texas.

Changes in both number and spatial distribution of intramembranous particles (IMP) have been documented as a result of differentiation, transformation, and changes in cell to cell contact. Distribution of particles can also be a function of concentration of specific reagents, ionic strength, or pH. In this study we observed the effect of varying concentrations of trypsin on the cell membrane via the freeze-etch technique. Unfixed confluent cultures of BHK-21-13 cells, a fibroblast-like cell line, were incubated with either balanced salt solution or varying concentrations of trypsin (.0025%, .025%, .125%, .25%) for 30 minutes at 37°C. Cells were harvested, incubated in a graded series of glycerol-Sorenson's buffer solutions, and freeze-etched in a Blazer's freeze-etch device. Etch time was 2 minutes, and replication was with platinum-carbon and carbon. In our cells we found numerous 70 to 90Å particles, the majority appearing on the inner fracture face. The concentration of trypsin did not drastically alter the number of IMP/micron². However, the distribution of IMP was dependent on trypsin concentration, more particle aggregates being present at low trypsin concentrations than at high ones. It is suggested that particle aggregates play a role in cell to cell contact and/or adhesion. Trypsin could act by removing peripheral membrane components essential to this aggregation.

STRUCTURAL RELATIONSHIP BETWEEN GOLGI APPARATUS AND ENDOPLASMIC RETICULUM: EVIDENCE FOR TRANSITION ELEMENTS. Hilton H. Kollenhafer, Veterinary Toxicology and Entomology Research Laboratory, ARS, USDA, College Station, TX 77840. Golgi apparatus are composed of dictyosomes which always appear to be in the same functional state. This synchrony implies that the dictyosomes are interassociated by direct dictyosome-dictyosome continuity, by some association with the endoplasmic reticulum, or by a constituent of the cell sap.

When dictyosomes are closely aggregated as, for example, in testicular germ cells, then synchrony is probably achieved by tubular connections between adjacent cisternae; at least tubular connections can be followed from dictyosome to dictyosome throughout the aggregate. An endoplasmic reticulum association also exists and is usually indicated by the close proximity of endoplasmic reticulum to the forming poles of the dictyosomes and by small vesicular profiles between the endoplasmic reticulum and the dictyosomes.

When dictyosomes are dispersed, as they are in most plant cells and in many animal cells, then the relationship between dictyosomes and between dictyosomes and endoplasmic reticulum is difficult to follow. What is indicated, however, is that the Golgi apparatus is composed of groups of interconnected dictyosomes with one or more transitional zones of endoplasmic reticulum near each dictyosome group. The transitional zones appear as anastomosing networks between the sheet-like and tubular portions of endoplasmic reticulum. In this configuration the endoplasmic reticulum could act to synchronize dictyosome function even if the dictyosome groups were not themselves interconnected.

AN ULTRASTRUCTURAL COMPARISON OF MATURE SPORES OF ASTOMUM AND FISSIDENS. Mueller, D.M.J. and Frank Seabury, Department of Biology, Texas A&M University, College Station, Texas 77843.

A comparison of the ultrastructure of Fissidens limbatus Sufl. and Astomum muhlenbergianum (Sw.) Grout revealed extensive differences in the condition of the cytoplasm at maturity. In the former, the protoplast contains numerous, uniformly shaped plastids with well developed grana and interconnecting lamellae. Little or no starch is seen. The cytoplasm is dense, organelles and lipid inclusions are evenly dispersed. In Astomum the protoplast of the mature spore is displaced peripherally by large lipid bodies. In addition, many small vesicles with similar electron density are included in the protoplast. Plastids are few in number, contain starch and apparently have a poorly developed internal membrane system. A study of wall structure in several genera indicates a three layered wall is of common occurrence in mosses. The sources of wall materials and methods of deposition are varied. Fissidens appears to have a wall which is of uniform thickness. Astomum has a similar three layered structure but exhibits an asymmetrical intine. The exine contributes to spore ornamentation in Astomum while in Fissidens ornamentation is due to perine deposition only.

ULTRASTRUCTURE AND BIOCHEMISTRY OF MYELIN AFTER ISONIZID-INDUCED NERVE DEGENERATION IN RATS. Richard G. Peterson and Charles P. Sea. Program in Neurostructure and Function. UTMSH, Houston, Texas 77025.

Sciatic nerves of rats showed marked changes following treatment with isoniazid (an antituberculous drug) for 5-15 days at about 250 mg per kg of body weight per day. Treated rats showed either loss of weight or a slower rate of weight gain than control animals. Electron microscopic observations reveal initial axoplasmic changes followed by destruction of the myelin interperiod band. As a result of this process the intra-period band may either split or become discontinuously aggregated. As degeneration continues myelin is often seen to be further compressed to take on the appearance of lipid bilayers. The myelin subsequently loses its structure as void globules are transformed into osmophilic droplets. The Schwann cells apparently undergo further change and appear to transform into macrophages. The principle change in protein composition when the nerve is solubilized and the proteins are run on SDS gel electrophoresis is reflected in a loss of the main protein band (P_0) of the myelin.

BRANCHING MITOCHONDRIA ASSOCIATED WITH A HUMAN HEPATOMA Alan B. Weckerling, M.S., Dept. of Pathology, Brooke Army Medical Center, Ft. Sam Houston, Texas 78234.

A 15 year old white male was admitted to the hospital in a comatose condition. A needle biopsy of the liver revealed large, multi-branched mitochondria while the other cellular organelles were within normal limits. Surgery revealed an extensive hepatoma which grossly had three types of nodes: a tan node, a white "fatty" node, and a red "fleshy" node. The latter also contained branching mitochondria as did a second liver biopsy which was taken during surgery. It is possible that these branching mitochondria had their division inhibited by some substance produced by the hepatoma. It is also possible that this same unknown substance contributed to the patient's comatose condition.

A LOW COST TECHNIQUE FOR RECORDING DIRECT POSITIVE IMAGES FROM A SEM USING STANDARD PRINT PAPERS. E. L. Thurston, A E Sowers, & J R Scott. Dept. Biology, Electron Microscopy Center, Texas A&M University, College Station, Texas 77843.

A major expense incurred recording images on a SEM is the Polaroid film. Some investigators use Polaroid type 55 film to produce a positive & negative image while others are mainly interested in the positive & therefore use Polaroid type 52 film. The above mentioned technique is designed to produce a positive image similar to type 52 film at a fraction of the cost. The system requires that the polarity on the photographic CRT be reversed & that ordinary print paper (Kodabromide, Polycor Contrast Rapid or Ekamatic) be loaded in 4x5 Graflex film holders & exposed. Since a variety of papers exist for such use, the paper, speed, contrast & developers can be altered to produce various effects. This type of versatility is a distinct advantage over Polaroid type 52 film. If a laboratory has access to an Ekamatic processing unit, the images can be processed in the SEM room providing results as rapidly as the Polaroid films. Various microscope operational parameters need to be altered to accommodate the slow paper speeds, etc. & such operational alterations will be presented.

A COMPARISON OF THE FINE STRUCTURAL CHARACTERISTICS AND PHAGOCYTIC PROPERTIES OF EARLY AND LATE PASSAGE WI-38 CELLS. Sheila Long, Marilyn Smith, and Janet Aune. Department of Biology, Texas Woman's University, Denton, Texas 76204.

The events of senescence have been studied quantitatively and qualitatively at a molecular level using the WI-38 cell. This is a normal diploid fibroblast which was derived from human fetal lung and has a finite in vitro life span of 40-50 doublings followed by a decline in cell viability and eventual death of the culture. This inevitable decrease in proliferative capacity is considered by many to be an example of aging at the cellular level. To examine functional changes associated with aging, the phagocytic response of early and late passage level WI-38 cells to latex beads was determined morphologically by transmission and scanning electron microscopy. In addition, the number of beads ingested by large populations of young and old cells was quantified by dioxane digestion and ultraviolet spectrophotometry.

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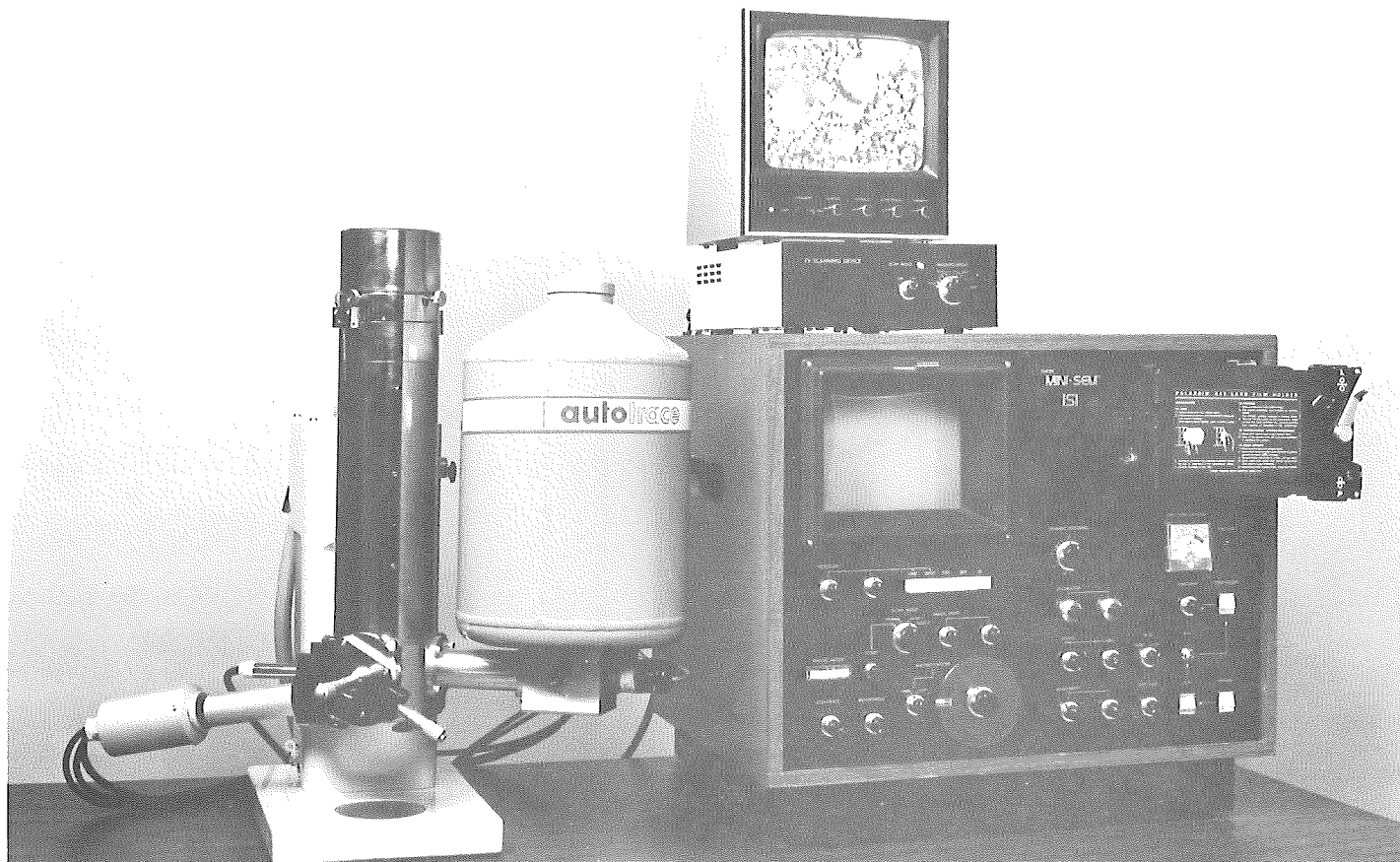
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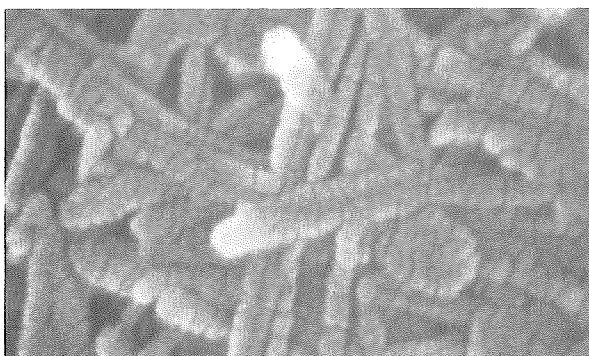
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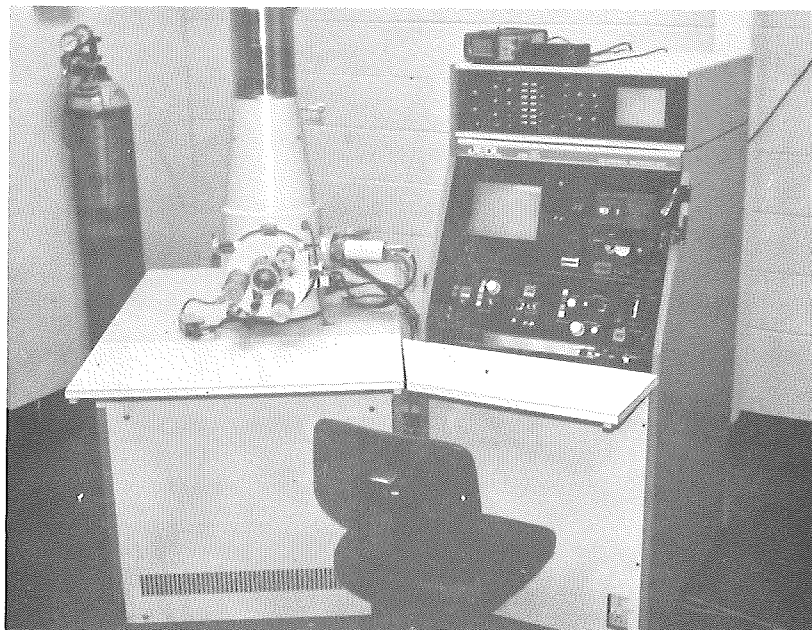


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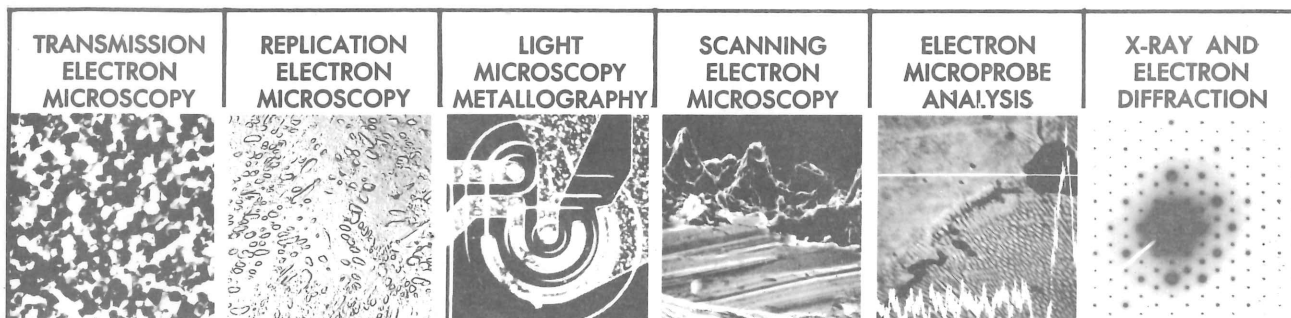
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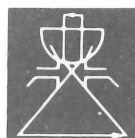


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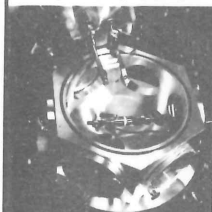


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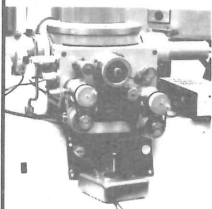
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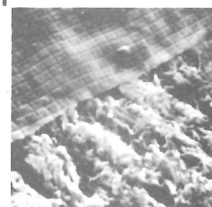
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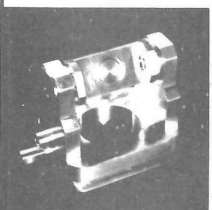


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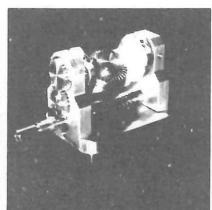


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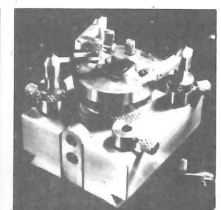
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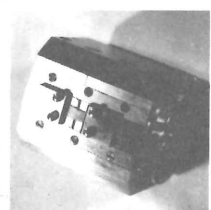
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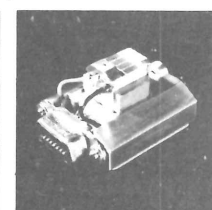
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