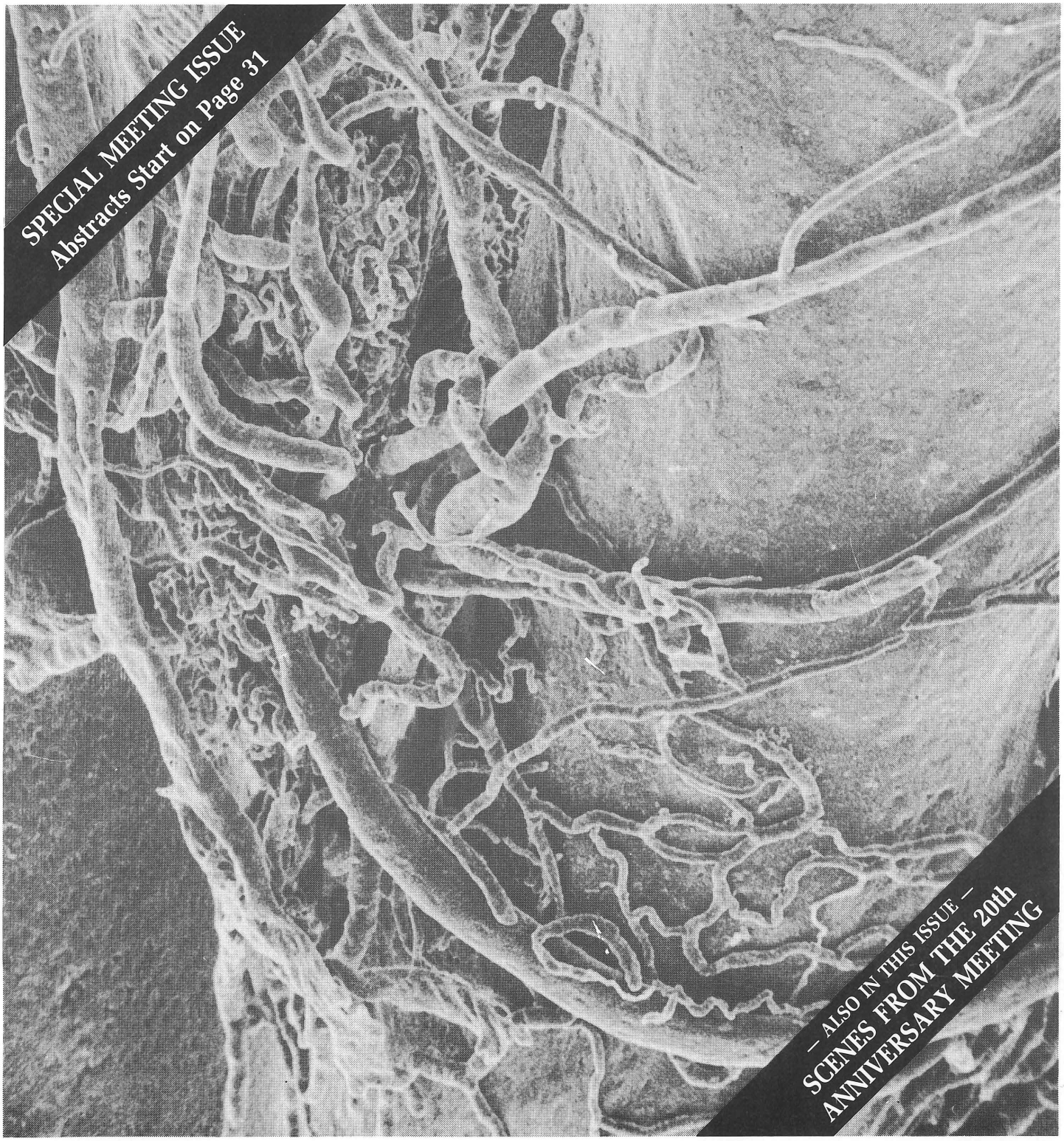
TSEM

Texas Society for Electron Microscopy

JOURNAL
VOLUME 16, NUMBER 2, 1985
ISSN 0196-5662



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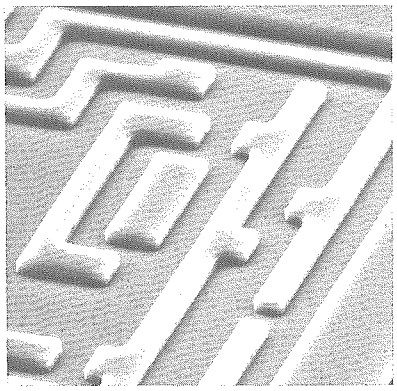

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JOURNAL
VOLUME 16, NUMBER 2, 1985
ISSN 0196-5662

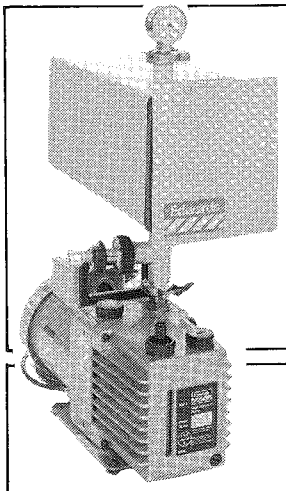
Randy Moore, Editor
Department of Biology, Baylor University, Waco, Texas 76798

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

ON THE COVER

Scanning micrograph of a Mercox vascular cast showing the vasa vasorum of the carotid sinus baroreceptor zone. The microvasculature of the carotid body chemoreceptor also is visible in the upper left of the micrograph. X425. See related story beginning on page 27.

Buyers Guide to EM specimen preparation



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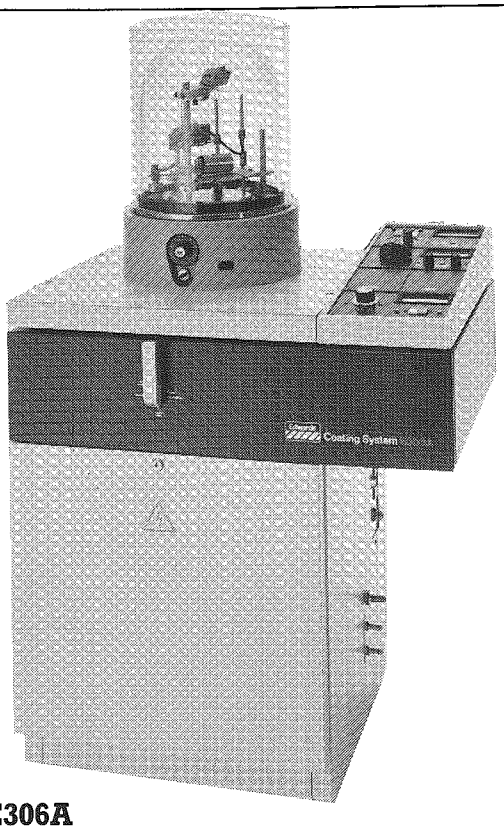

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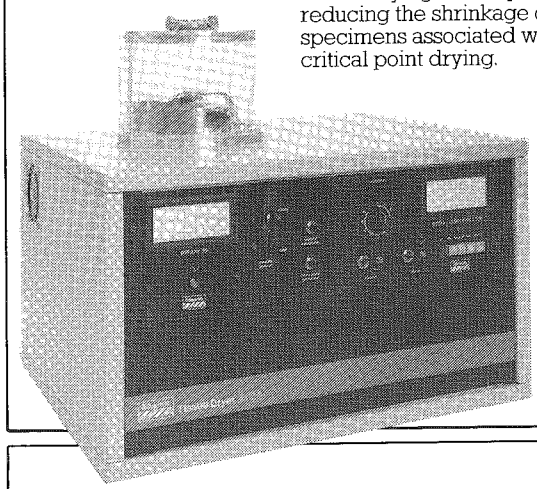

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President's Message

From all accounts the Spring Meeting of TSEM in San Antonio was an outstanding success. Much of the credit for the meeting should go both to our past president, Charles Mims, for excellent planning and leadership and to those members of TSEM who worked long and hard in making the meeting a reality. However, the success of this meeting, as well as any other endeavor, ultimately depends on support from the entire membership. We had this support in San Antonio from past presidents to industry to general membership. I encourage continued support for our society and attendance at meetings whenever possible. I especially encourage the senior investigators and previous TSEM officers whose participation and monetary support is necessary for both students and technicians.

By the time you read this letter at our Fall Meeting, TSEM will have become a corporation. The move to incorporate was taken at the advice of council to help protect the individual members from legal action that might be directed against the Society. The cost for legal council and preparation of all paperwork has been exceedingly modest (less than $300) and I believe that this has been a good move. Except for the "Inc." there should be no noticeable change in any aspect of the Society.

Many of us are again looking forward to a joint meeting with our neighbors in Louisiana as in years past. The LSEM is a society of exceptional quality whose members are a delight to be with. I harbor many fond memories of past meetings and past acquaintences. It's good to be back again.

The Spring, 1986 meeting of TSEM will be in Nacogdoches on March 20-22. A mini-symposium on "aging and electron microscopy" is being planned along with the other usual activities. Program planning is well underway and the indications are that this will be another outstanding meeting. A dinner at the lake on Thursday evening will be a pleasant way to start the meeting and to visit with friends.

I look forward to seeing all of you at Beaumont.

Sincerely,

Hilton H. Mollenhauer
President

EDITORIAL POLICY

LETTERS TO THE EDITOR

Letters to the editor are printed as they are received in the order of their arrival. These letters reflect the opinion of the individual TSEM member and do not necessarily reflect the opinions of the editor or the society. The content of the letters should be concerned with the philosophical or operational aspects of the TSEM, the Journal and its contents, academic or national policies as they apply to TSEM and/or its members and electron microscopy in general. Editorial privilege may be evoked to insure that the LETTERS SECTION will neither be used as a political forum nor violate the memberships' trust.

ELECTRON MICROGRAPHS AND COVER PHOTOS

Micrographs submitted for cover photos should be marked as such. The choice of photographs will be made by the editor. Photograph receipt and/or dispensation will not be acknowledged. Photographs will not be returned. Electron micrographs to be used for cover photos and text fillers are welcome and should be selected with some attention to aesthetic appeal as well as excellence both technique and in scientific information content.

REGIONAL NEWS

News items should be submitted through the regional editor in your area and made to conform to the standard format used by the regional news section. Regional contributions should be sent to the Regional News Editor. Editorial privilege may be executed for the sake of brevity or to preserve the philisophical nature of the TSEM Journal.

The JOB OPPORTUNITIES section will be comprised of a "Jobs Available" and a "Jobs Wanted" sub-section. Anonymity of individuals listing in the Jobs Wanted or Jobs Available sub-sections may be maintained by correspondence routed through the Regional News Editor's office.

TECHNICAL SECTION

The Technical Section will publish TECHNIQUES PAPERS, HELPFUL HINTS, and JOB OPPORTUNITIES. The TECHNIQUES PAPERS will describe new or improved methods for existing techniques and give examples of the results obtained with methods. The format of the Technique Papers will be the same as that used for regular research reports. HELPFUL HINTS will be in the form of a brief report with an accompanying illustration, if required for clarity. Helpful Hints should embody techniques which will improve or expedite processes and/or procedures used in EM.

PUBLICATION PRIVILEGES

The right to publish in the TSEMJ is restricted to TSEM members or to those whose membership is pending. A membership application form can usually be found in each issue of the TSEMJ. Membership dues are as follows: students $2.00; regular members $10.00; Corporate members $75.00. Individuals who belong to TSEM by virtue of a corporate membership are invited to participate in Journal submissions as are our regular or student members. However, papers of a commercial nature, either stated or implied, will not be accepted for publication as a Research Report or Techniques Paper. Such papers may be acceptable as advertising copy.

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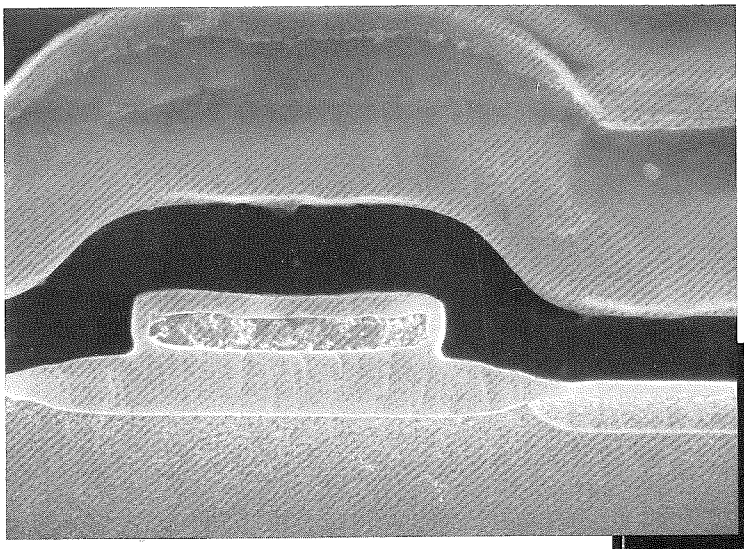

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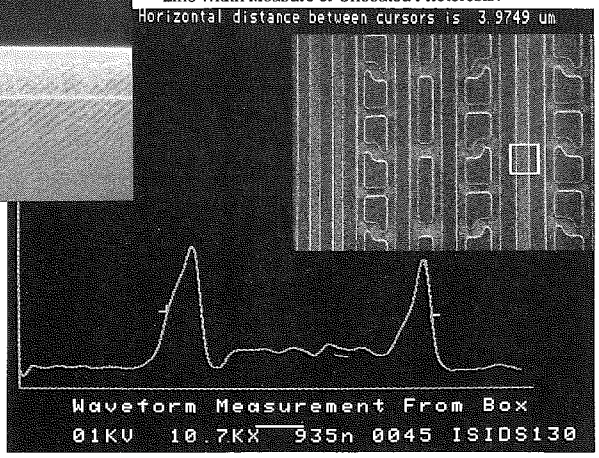

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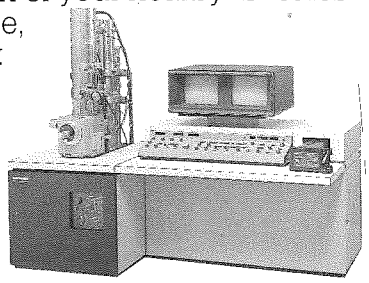

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Treasurer's Report

ASSETS ON 25 OCTOBER 1984:		
Certificate of Deposit No. 64766, Univ. Natl. Bank, Galveston	$ 2,000.00	
Certificate of Deposit No. 10-0475417, United Savings of Texas	3,245.65	
Certificate of Deposit No. 240-0064030, Republic Bank of Waco	2,243.56	
Checking Account No. 7914-448-1, Republic Bank of Waco	6,375.27	$ 13,864.48
RECEIPTS:		
Membership Dues	$656.00	
Arlington Meeting, Registration and Donations[1]	1,569.00	
Interest		
Checking Account No. 7914-448-1	149.00	
Certificate of Deposit No. 10-0475417	82.73	
Certificate of Deposit No. 7914-448-1	94.71	
Certificate of Deposit No. 64766	101.52	
TSEM Journal		
Subscriptions	60.00	
Advertising Revenue	$ 4,742.50	
San Antonio Meeting		
Exhibitors Fees	1,000.00	
EMSA Local Support	500.00	$ 8,955.46
EXPENSES:		
Arlington Meeting[1]		
Materials Sciences Group — John Lange	$ 274.80	
Honoraria	200.00	
Expenses of Beth Grant	533.33	
Xerox Costs for Registration Materials	19.34	
Equipment Rental	129.08	
Student Travel	67.29	
Rodeway Inn — Room Rental	1,036.27	
TSEM Journals for Distribution	217.00	
Secretarial Expenses	1,408.70	
Plaques for Past Officers	75.74	
TSEM Journal Printing Costs[2]	1,600.00	
Secretarial Help for John Lange	400.04	$ 5,961.59
ASSETS ON 11 APRIL 1985:		
Certificate of Deposit No. 64766	$ 2,000.00	
Certificate of Deposit No. 10-0475417	3,328.38	
Certificate of Deposit No. 240-0064030	2,338.27	
Checking Account No. 7914-448-1	9,191.70	$ 16,858.35

[1]Net loss on Arlington Meeting = $908.11

[2]Net profit on Vol. 15, No. 3 of TSEM Journal = $1,200.00

Respectfully submitted,

Randy Moore, Treasurer
Texas Society For Electron Microscopy

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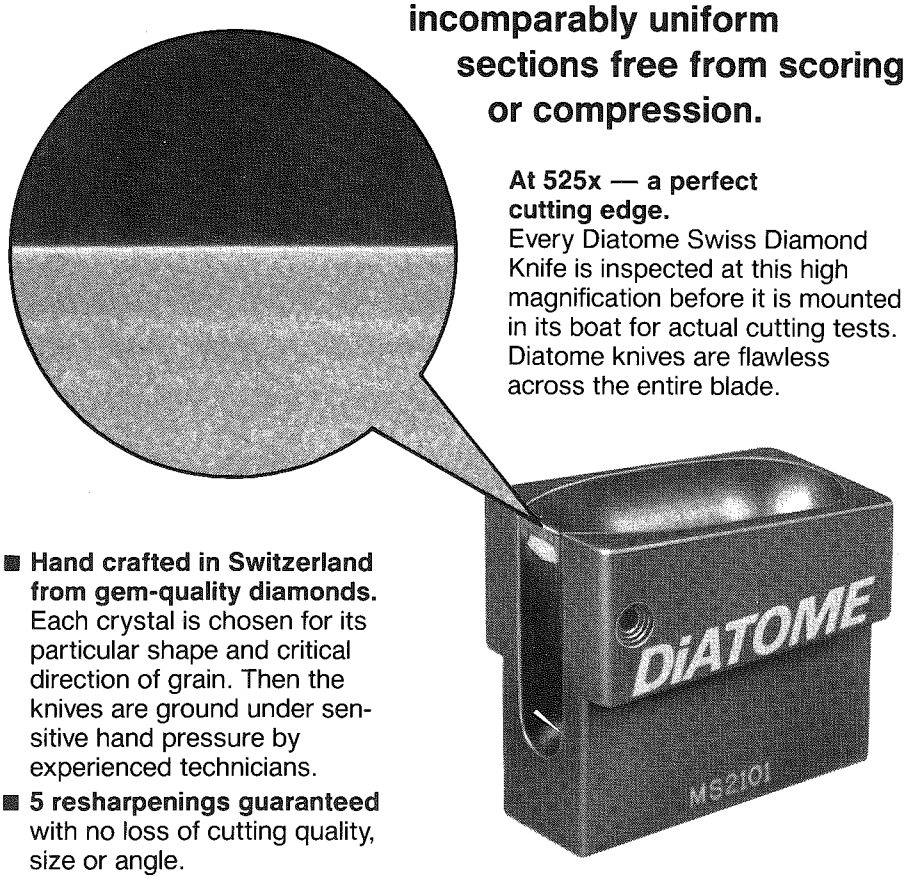

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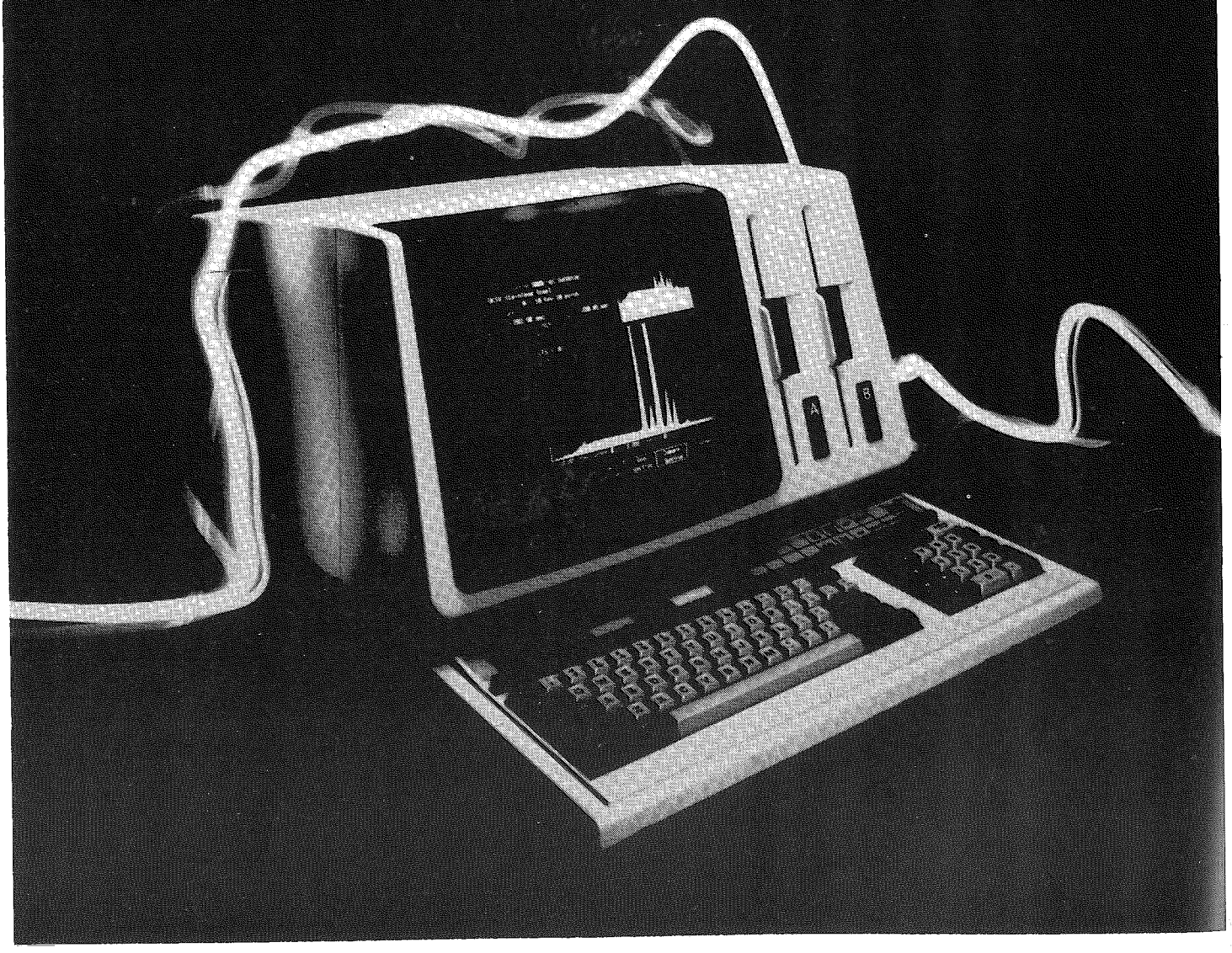

PAST PRESIDENTS OF THE TSEM

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At the 20th Anniversary Meeting
of the TSEM, 1985, San Antonio

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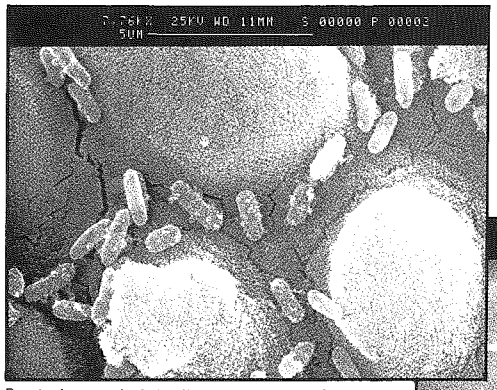
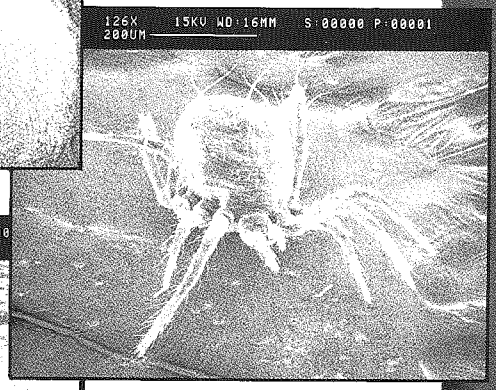
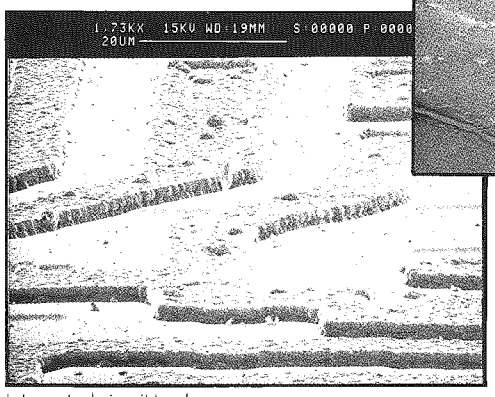


Bacteria on pig intestine.



Squirrel flea.



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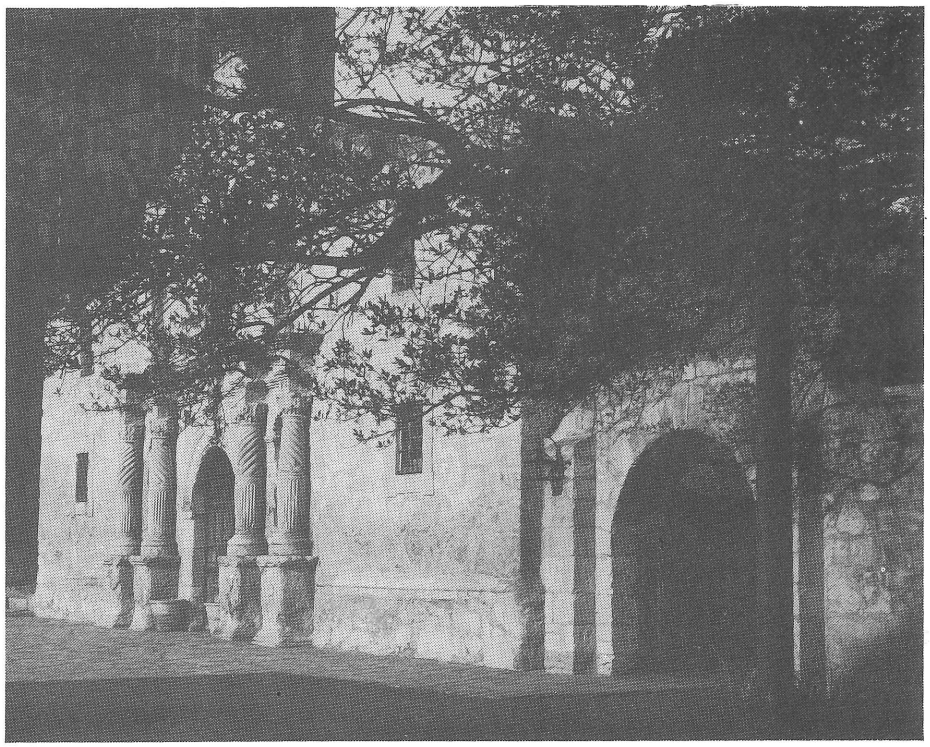
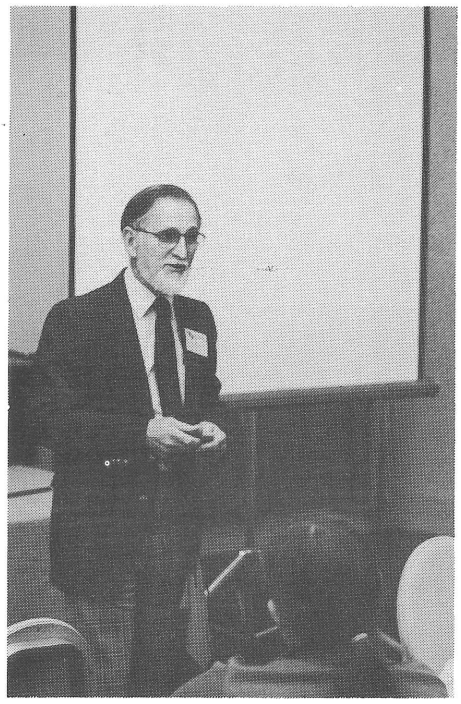
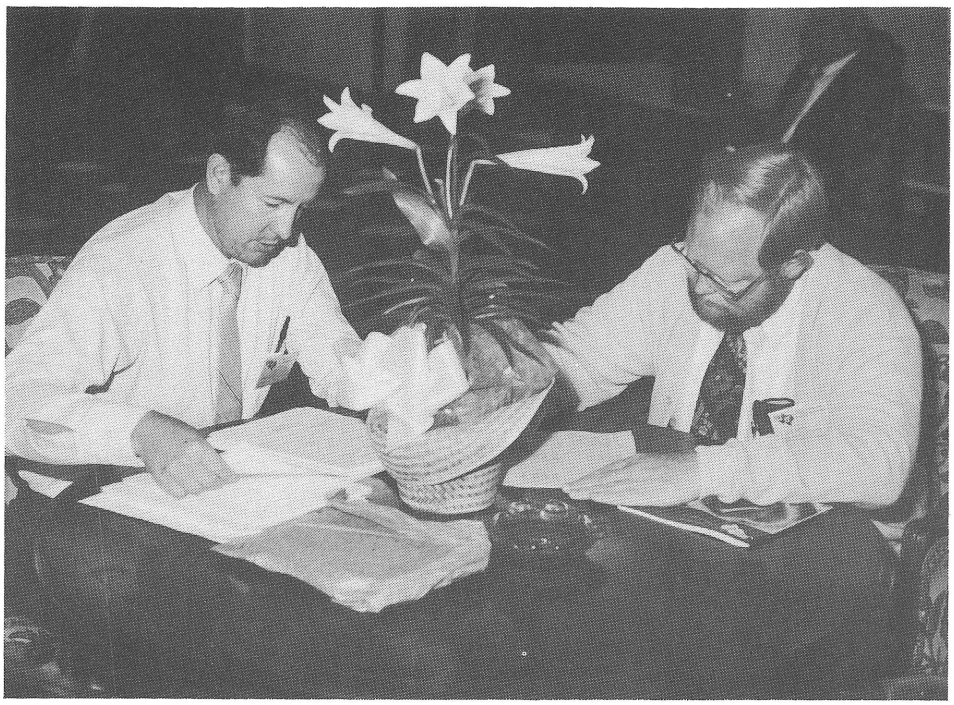
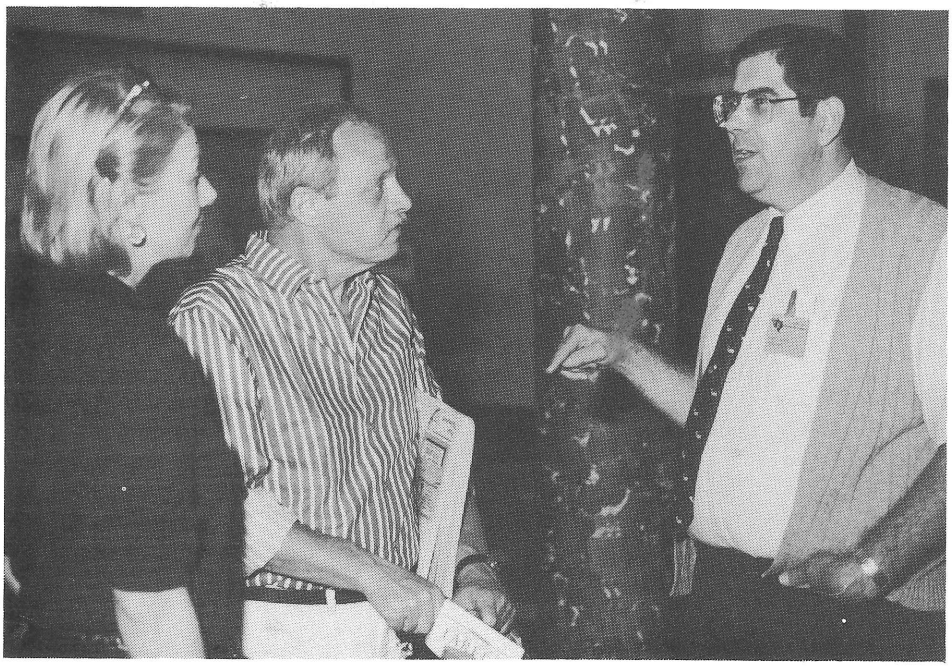
SCENES FROM THE 20th ANNIVERSARY MEETING OF THE TSEM

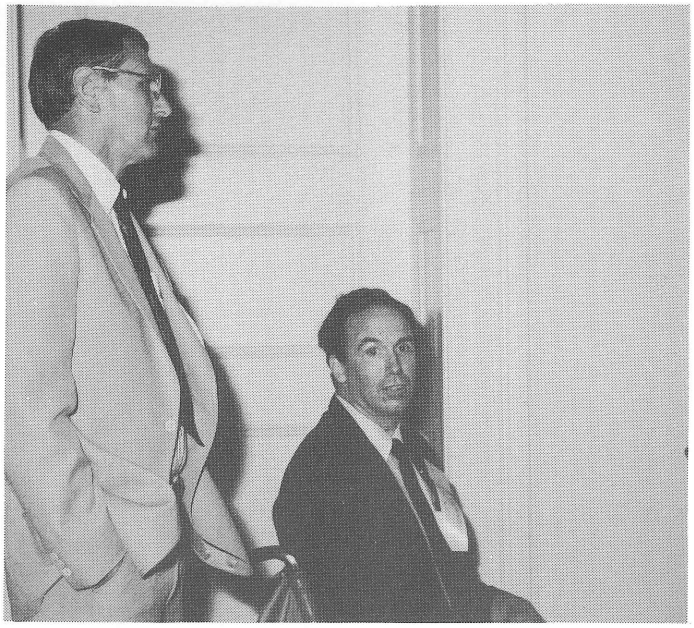
Photographs by Joiner Cartwright, Jr.

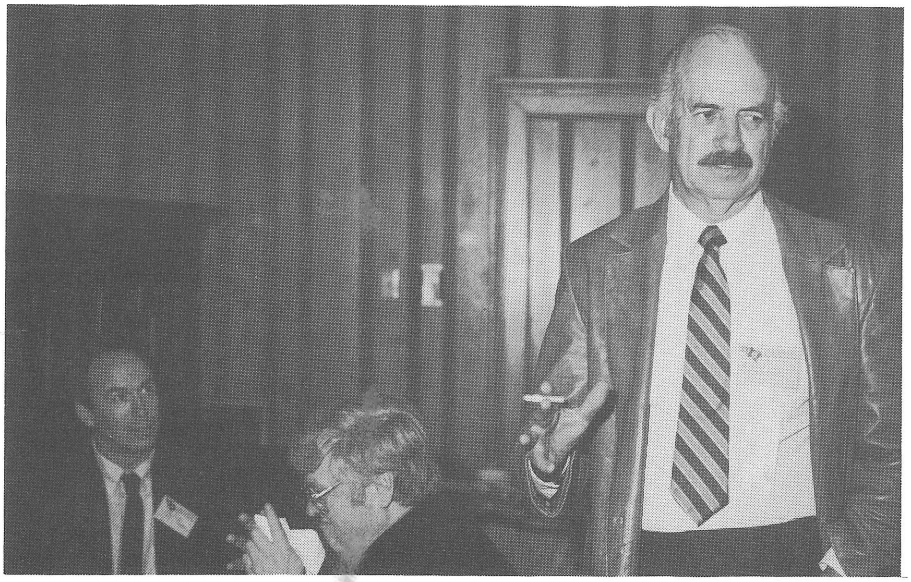


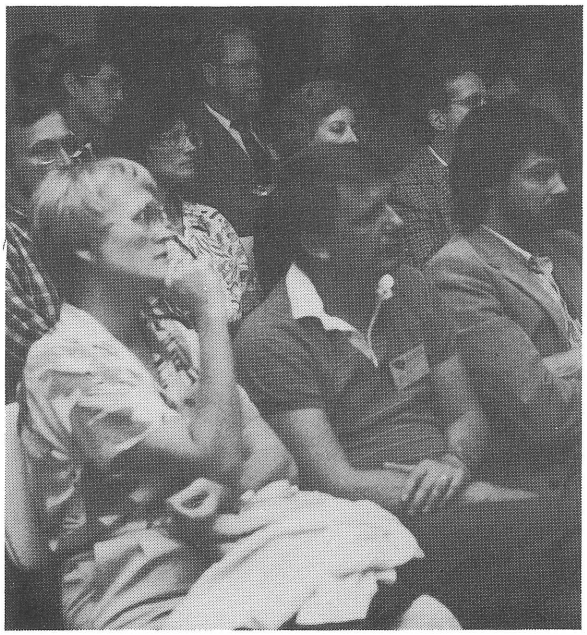


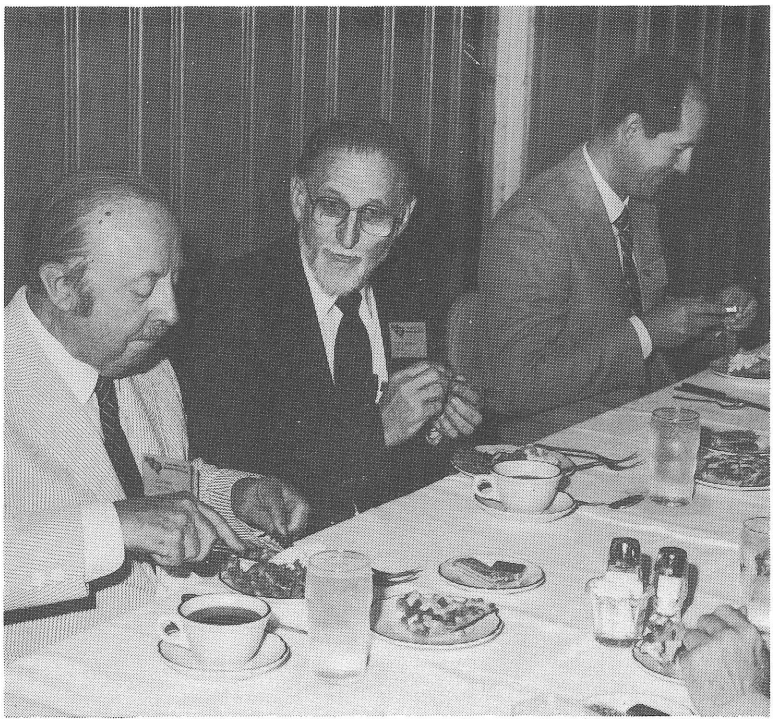












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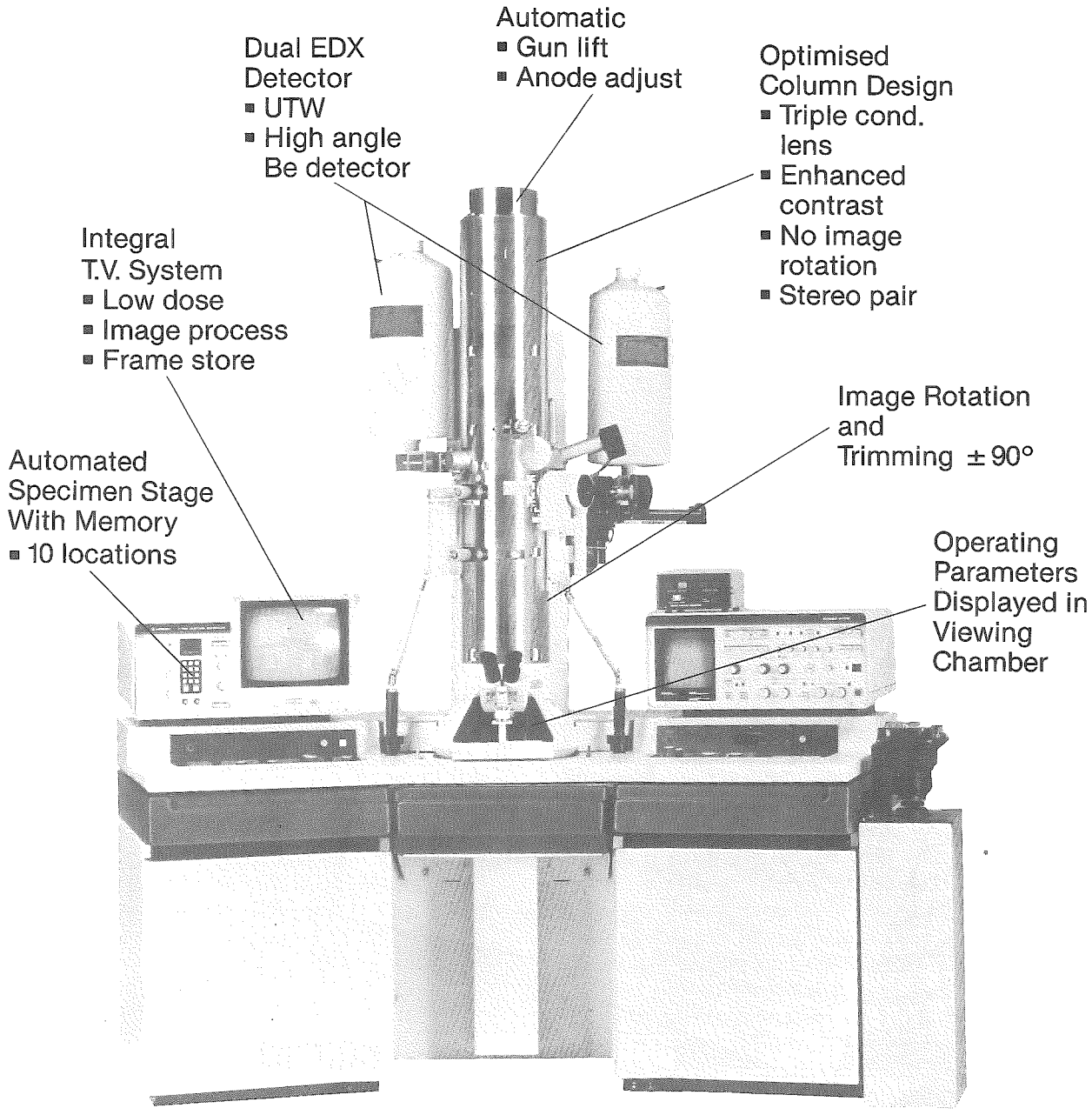

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GOLD EMBRITTLEMENT IN SOLDERING PROCESSES

By

J.L. Marshall

Motorola, Inc.
Also, Adjunct Professor, North Texas State University

James L. Marshall
(817) 232-6004
Motorola, Inc.
5555 North Beach Street
Ft. Worth, Texas 76137

ABSTRACT

When gold-plated leads are soldered with tin-lead, the brittle intermetallic of gold-tin develops. Gold-embrittled solder joints can develop into poor electrical contacts. Potentially poor electrical joints can be recognized by the presence of gold-tin at the interface of the base metal and solder. A minimum of gold should be used on plated leads to ensure maximum conductivity: thicknesses of no more than 1.5 μm of gold plating should be allowed.

INTRODUCTIONS

Gold is an excellent protective coating, not only for contacts and mating surfaces (1,2) but also for solderable surfaces (3,4). However, a layer of brittle gold-tin can form and cause mechanical weakness (3). An extensive review of different solders has been made in a search for a composition that will reduce embrittlement problems (5); however, it appears that the common tin-lead solders, which have high tensile strengths (6), give the best results.

In this study we observed that the gold embrittlement phenomenon can also result in high electrical resistances, even when mechanical (peel) strength is acceptable. This study was expanded to include a characterization of gold embrittlement by scanning electron microscopy.

Methods and Materials

Carbon-steel frames, plated with layers of nickel and then gold, were soldered. The solder process utilized International Solder Supplies 63/37 Sn/Pb solder paste, which was reflowed in a tunnel reflow device with a nitrogen blanket. The temperature excursion was 150° → 230° → 150° C over 2.5-3.0 minutes, with the peak temperature for 20-25 seconds. Temperature quench was accomplished using dry nitrogen blown over the soldered sample.

Samples were prepared both for surface and cross section examination. Cross sections were prepared using Beuhler Epo-Kwick resin and Minimet polishers, followed by gold-palladium sputtering. Samples were studied using a JEOL JSM-35CF scanning electron microscope. Energy dispersive X-ray analysis was performed using the Tracor Northern TN-4000 system. Compositional analysis was done with the Flextran Semi-quantitative Analysis software.

RESULTS AND DISCUSSION

Phases observed. There are six phases arising from the gold and solder: Au, AuSn, $AuSn_2$, $AuSn_4$, Sn, and Pb (7).* No Au-Pb intermetallics (Au_2Pb, $AuPb_2$) and no Au_6Sn were observed, as previously suggested (9).

Figure 1 displays cross sections of solder, with Sn, Pb, and the common gold-tin $AuSn_4$ phase. The three different forms of the $AuSn_4$ phases can be discerned: lamellae, prisms, and needles. In Figures 2 and 3 the less common AuSn and $AuSn_2$ intermetallics are visible, close to the gold layer and on the periphery of the solder where the tin is no longer in excess.

Surface examination. Figures 4 and 5 show a typical distribution of the bulk elements at the gold-solder interface. The tin of the solder is scavenged from the solder, causing a lead-rich area close to the interface. Close inspection of this lead-rich area (Figure 6) shows lead ovoids in a matrix of eutectic tin-laed with occasional $AuSn_4$ prisms. Futher from the gold-solder interface, an outcropping of $AuSn_4$ needles appear (Figure 7-8).

Cross section examination. Figures 1-3, already presented, show the cross section of a gold-solder interface. Where only a small amount of solder is used

**The Sn and Pb phases are not pure and accordingly are sometimes designated by "α" and "β." The tin-gold phases are also often designated as β = $AuSn_4$, γ = $AuSn_2$, and δ = AuSn; and the tin phase itself can have an α and β form (8). To avoid confusion, reference to the phases will be by formula, with the understanding that stoichiometry is rarely exact. Average percentages of the three gold-tin phases were for Au/Sn: AuSn, found 60.4/39.6, calculated 62.4/37.6; $AuSn_2$, found 45.8/54.3, calculated 45.3/54.7; $AuSn_4$, found 32.9/67.2, calculated 29.3/70.7.*

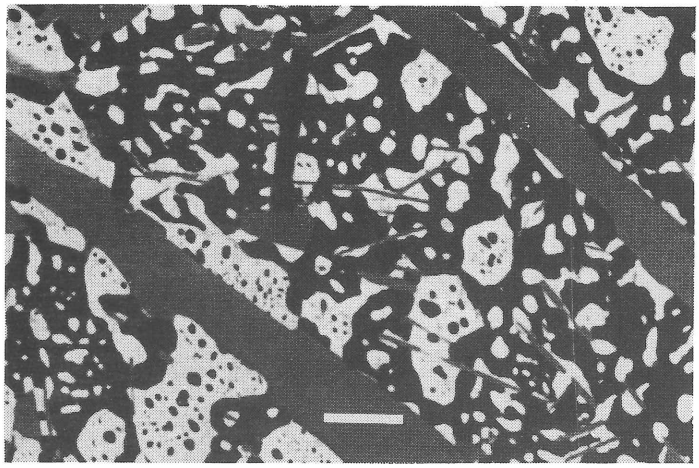


FIGURE 1: Cross section of portion of typical gold-plated joint soldered with tin-lead. The dark phases are tin, the white phases are lead, and the gray phases are the gold-tin intermetallic $AuSn_4$. This latter phase appears as a lamellae, prisms, and needles. Backscattered detection. Scale = 10 μm.

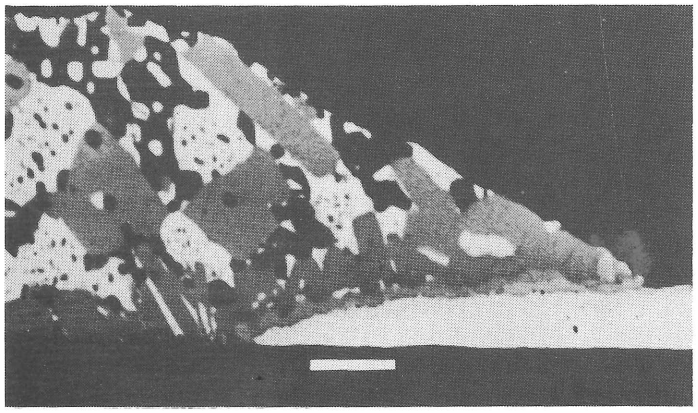
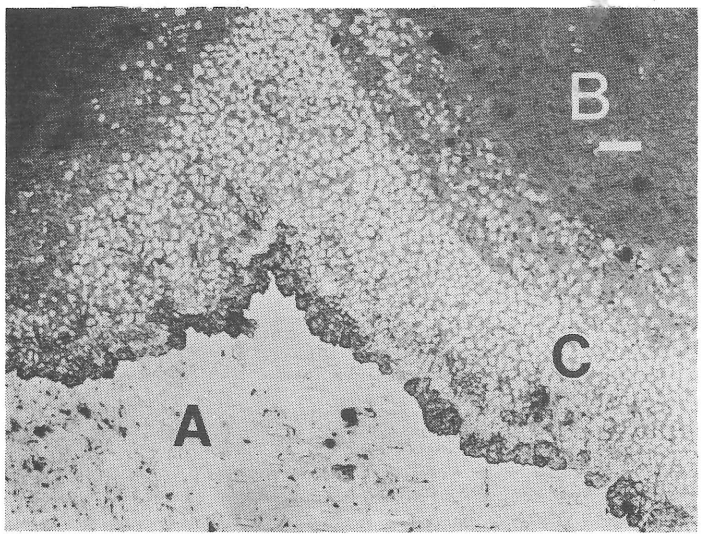


FIGURE 2: Cross section of gold-solder interface. The same phases can be detected as in Figure 1. In addition, the dark base in the nickel plate and the light tongue extending from the right is gold plating. Backscattered detection. Scale = 10 μm.



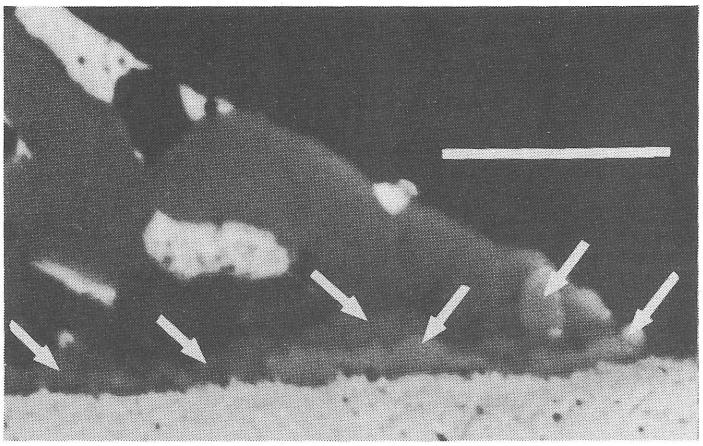


FIGURE 3: Expansion of Figure 2. Two new phases of gold-tin can be observed in the tin-poor region: intermediate shades of gray denote the presence of $AuSn_2$ (arrows from left) and AuSn (arrows from right). Backscattered detection. Scale = 10 μm.

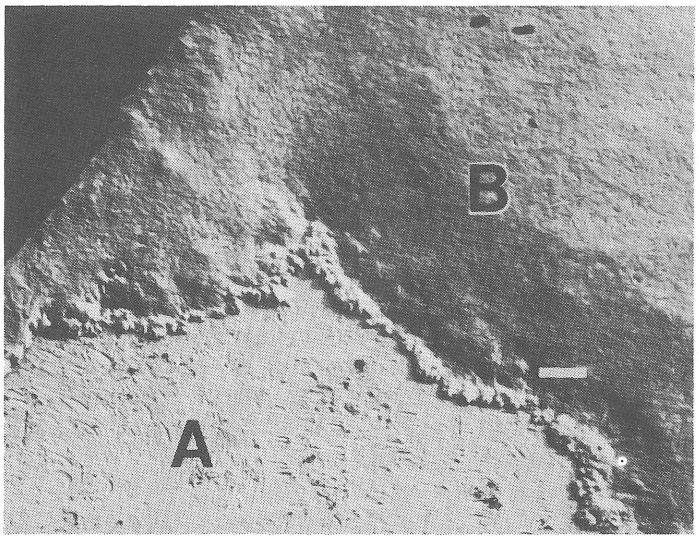


FIGURE 4: Surface view of interface between gold (A) and solder (B). Secondary detection. Scale = 100 μm.

FIGURE 5: Backscattered detection of view of Figure 4. An intermediate lead-rich region (C) is apparent, caused by scavenging of the tin by the gold. Scale = 100 μm.

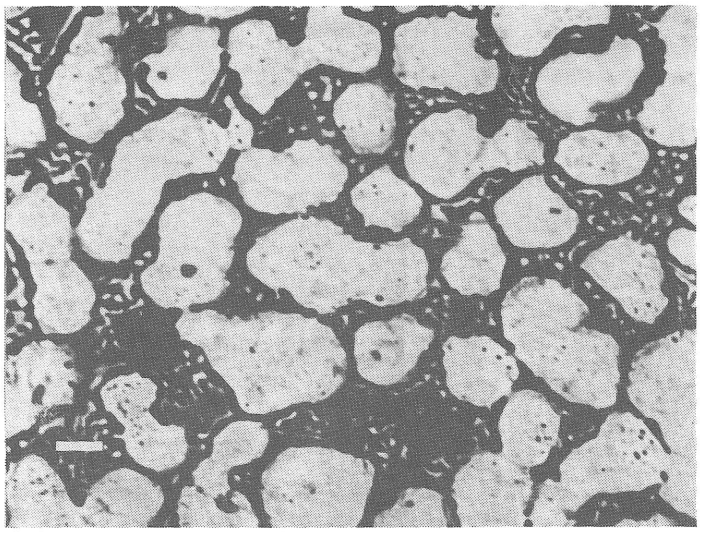


FIGURE 6: Expansion of region (C) of Figure 5. The light ovoids are lead, cemented together by the typical mosaic of eutectic solder (dark = tin, light = lead). The gray prisms are AuSn•. Backscattered detection. Scale = 5 μm.

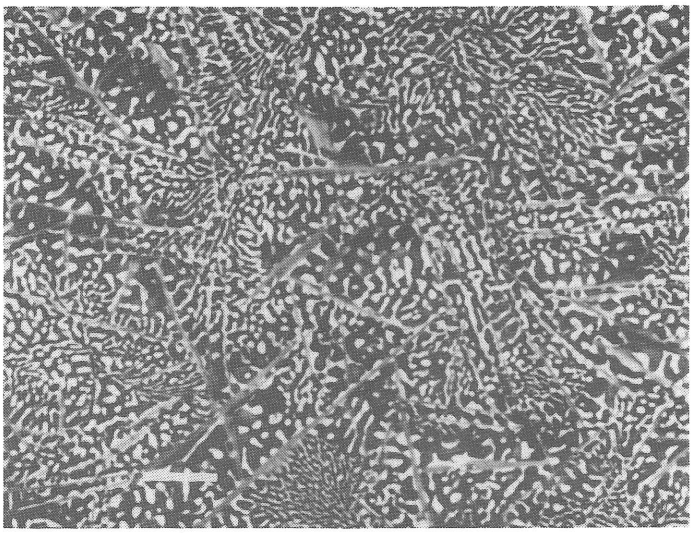


FIGURE 8: Backscattered detection of view of Figure 7. The general pattern is eutectic tin-lead solder, with AuSn• needles. Scale = 10 μm.

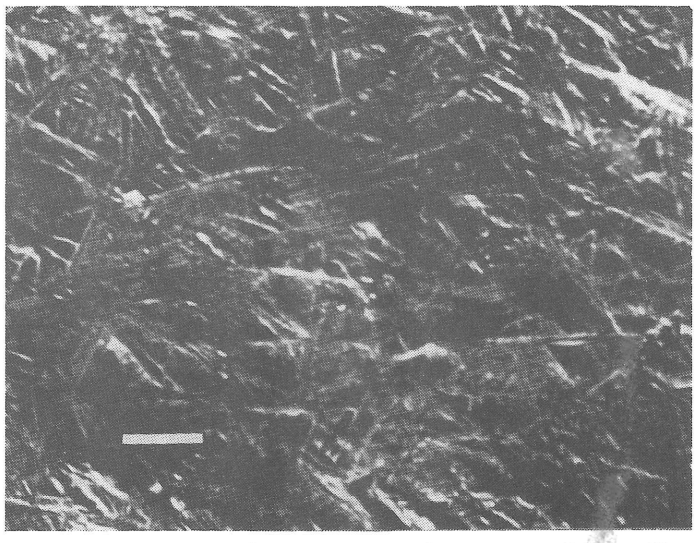


FIGURE 7: View in the (B) region of Figure 5. The needles are AuSn•. See next figure. Secondary detection. Scale = 10 μm.

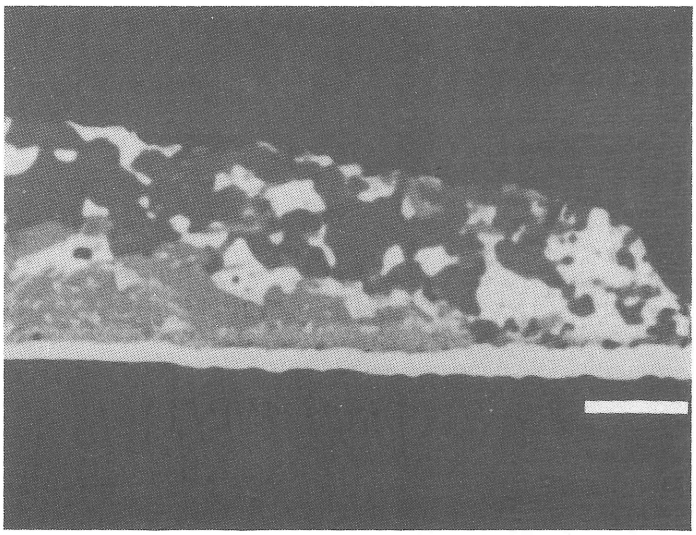


FIGURE 9: Cross section of solder joint where a small amount of solder is used on a frame with a thick plating of gold (5 μm). The phases are the same as in Figure 2. The gold layer does not completely dissolve, proving excessive gold was originally used. Extensive gold-tin intermetallic formed at the interface, causing embrittlement. This solder joint displays inferior electrical performance. Backscattered detection. Scale = 10 μm.

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on a frame with a thick layer of gold (Figure 9), the gold is not completely dissolved away and an extensive area of gold-tin intermetallic is formed, causing high resistivity and accordingly poor electrical performance. If the amount of gold-tin intermetallic is minimized, then electrical performance is improved. This reduction of gold-tin intermetallic is accomplished by using more solder, reheating the solder joint, or plating originally with a thinner layer of gold. Figure 10 shows an intermediate stage where less gold was used originally. In this example the gold is completely dissolved under the solder, but contact of the solder to the frame is still accomplished through gold-tin intermetallic, and electrical performance is not yet optimized.

Samples characterized by excellent electrical properties had both the gold and the gold-tin intermetallic scavenged from the nickel frame. Figures 11 and 12 show where this has been successfully accomplished. Careful EDX analysis shows that a thin (<1 μm) layer of nickel-tin intermetallic has formed between the nickel and solder, necessary for a good solder joint between the two metals (10-12).

The results of this study support the suggestion (3) that no more than 1.5 μm of gold over nickel should be plated. Thicknesses of 3-5 μm of gold were found to be excessive, and extra soldering steps were necessary to improve the electrical performance to acceptable levels.

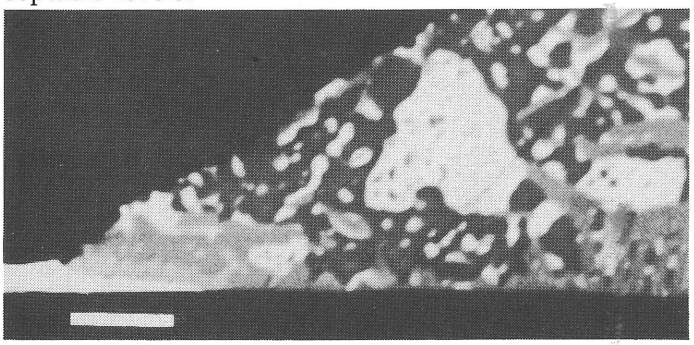

FIGURE 10: Cross section of solder joint where a greater amount of solder is used on a frame with originally a thinner layer of gold (3 μm). The gold is now completely dissolved under the solder. However, extensive gold-tin intermetallic still can be seen at the solder-nickel interface, and electrical performance is still impaired. Backscattered detection. Scale = 10 μm.

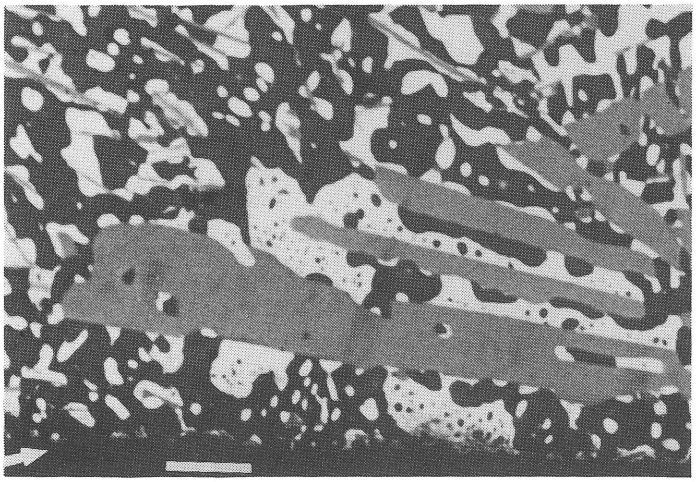

FIGURE 11: Cross section of solder interface with superior electrical properties. The dark base is the nickel plate. Throughout the entire length of the solder-nickel interface (arrow), the gold-tin intermetallic is minimal, and good contact can be made between the nickel and the tin phase of the solder. See next figure. Backscattered detection. Scale = 10 μm.

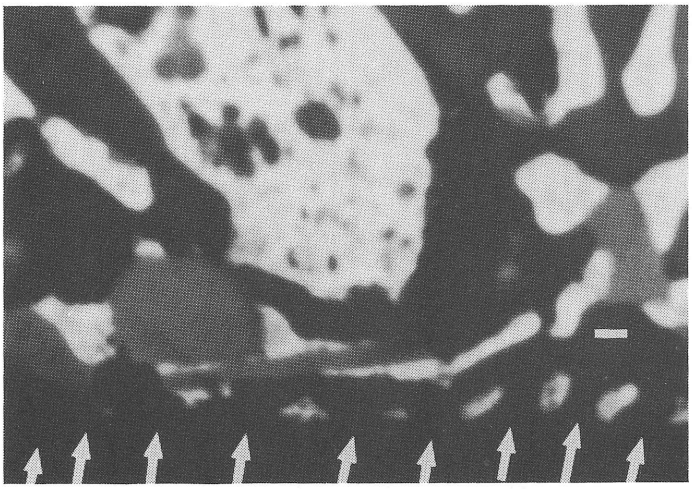

FIGURE 12: Expansion of view of Figure 11. The excellent contact between the nickel and tin can be seen (arrows). By careful energy dispersive X-ray analysis, the nickel-tin intermetallic layer necessary for good mechanical and electrical contact can be recognized (<1 μm; not discernable in figure). Backscattered detection. Scale = 1 μm.

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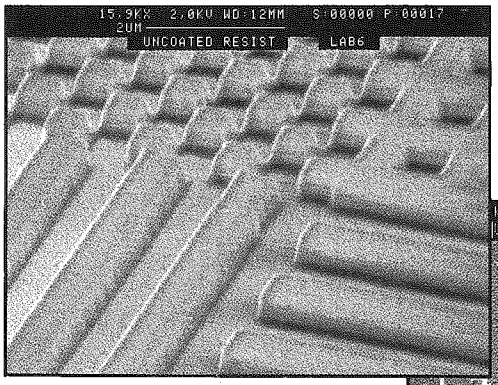

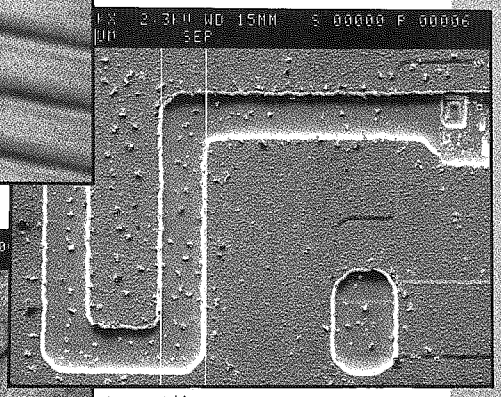

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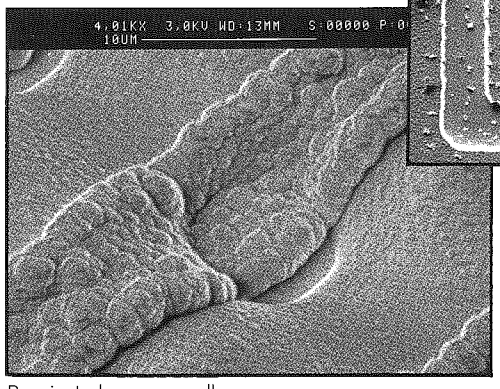

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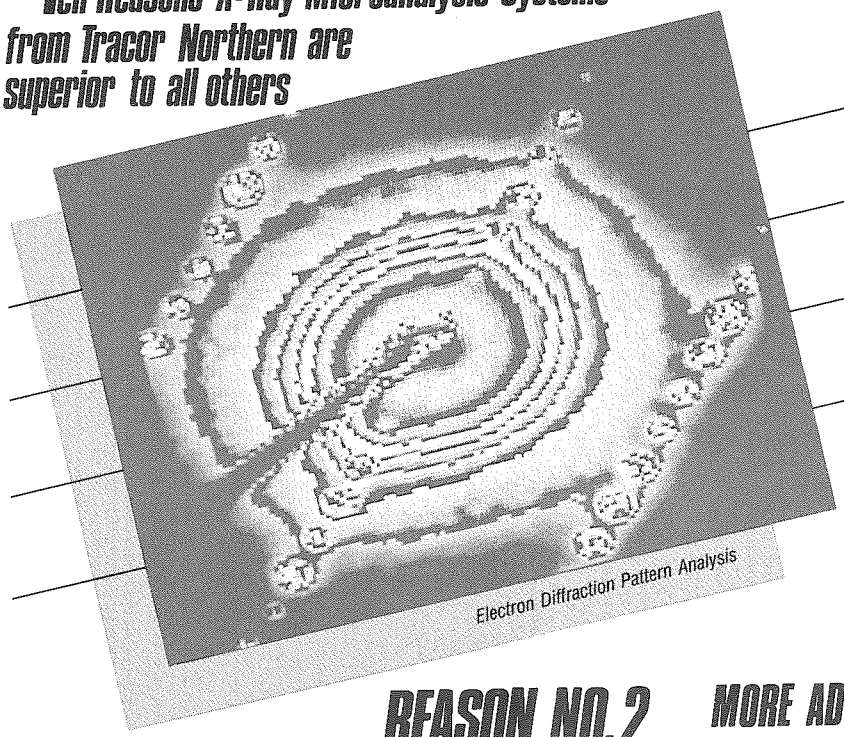

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VASCULAR CASTING: AN IDEAL WAY TO VISUALIZE THE MICROVASCULATURE THREE-DIMENSIONALLY

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ABSTRACT

An appreciation for the three-dimensional complexity of the microvasculature is difficult to obtain from two-dimensional tissue sections. Using a low viscosity methacrylate resin (Mercox), we have been able to obtain excellent corrosion casts of various vascular beds which then can be examined in the scanning electron microscope. This paper describes a protocol for using Mercox which provides corrosion casts suitable as teaching aids and research specimens.

INTRODUCTION

An appreciation for the structural complexity of the microvasculature is difficult to obtain even with the resolving power of the transmission electron microscope (TEM). This is particularly true if one wishes to examine the three-dimensional configuration of the vasculature. To some extend, an appreciation of vascular complexity can be obtained at the light microscopic level if one uses special labelling techniques such as horseradish peroxidase infusion, lead chromate precipitation, colored gelatin or latex infusion, or carbon particle labelling (1-3). Nevertheless, one often gets unsatisfactory results from incomplete filling of the smaller arterioles, venules and capillaries. Fine detail also cannot be discerned at the level of the light microscope. While TEM possesses the necessary resolving power to study the fine details of the microvasculature, tedious serial section reconstructions are necessary to gain an appreciation of its three-dimensional complexity.

Fortunately, many of these drawbacks in studying the microvasculature can be overcome by using scanning electron microscopy (SEM) of vascular casts. Corrosion casts of the blood vessels permit one to examine fine detail as well as gain insight into the three-dimensional configuration of a vascular bed. However, as with other labelling techniques, one must achieve complete filling of the small vessels and this requires a low viscosity material. In this paper, we describe a protocol of using Mercox, a low viscosity commercially available methacrylate resin, which provides excellent corrosion casts of the microvasculature.

Materials And Methods

Adult rats of either sex were anesthetized with Nembutal (40 mb/kg ip) and perfused through the heart with warm saline (38°) containing 1000 units of heparin/500 ml. When the effluent was clear, the rats were perfused with a warm fixative solution consisting of 3% glutaraldehyde in 0.1m cacodylate or 0.1 M phosphate buffer at pH 7.2. The perfusion pressure was maintained at 120 mm Hg and each animal was infused with 200 ml of fixative. Immediately following the perfusion, Mercox (Ladd Research Industries, Inc.) was prepared by combining 20 ml of resin with 1 ml of catalyst in a 50 ml syringe. This mixture was infused through the heart and into the ascending aorta in 1 min, by which time the Mercox had begun to polymerize. The infused rat was cured for 1 hr in an oven at 50°C and then the

desired tissue samples removed. Samples were macerated in 5.25% hypochlorite (household bleach) for 24-48 hrs with daily rinses in distilled water. Once clean of tissue, the casts were rinsed in water, air dried, oriented and mounted on SEM stubs, sputter-coated with gold and examined in the scanning electron microscope at 15 kV..

RESULTS AND DISCUSSION

Examples of vascular casts are shown in Figure 1-4. These representative casts demonstrate the superb filling of even the smallest capillary beds, such as those seen in the carotid body arterial chemoreceptors (Fig. 1) and the adjacent vasa vasorum of the carotid sinus baroreceptor zone (Figs. 1-2). Indentations of the endothelial cell nuclei can be observed in these casts. By means of vascular casts, a detailed overview of the complex ramifications of a vascular bed can be appreciated and, by calibrating the SEM, one can perform measurements on the extent of each vessel network. Arterioles can be distinguished from venules by their smoother contour and uniform size (Fig. 2). Vascular detail deep within a larger organ, such as the kidney or liver, can be viewed by slicing the organ into smaller slabs prior to tissue maceration (Fig. 3). The relationship between skeletal structures and the vasculature can be preserved by controlling the length of time in the hypochorite solution (Fig. 4); soft tissues are macerated more quickly than bone.

Several aspects of the protocol deserve comment. First, it is important to maintain the perfusion pressure at a level approximating that of the animal's normal pressure. One can accomplish this by directly measuring the animals' systolic blood pressure, as we did, or by consulting a chart which includes common physiologic parameters for laboratory animals based on weight, age and sex of the animal. The pressure of the perfusate then can be controlled using a gravity system and flow meter, or by actively pumping the perfusate while monitoring the pressure with a gauge. Second, heparin should be admininistered prior to the perfusion or be included in the saline flush. One may also add 1% $NaNO_2$ to the saline flush to insure that the vessels remain dilated. Third, it is important that the saline and fixative be perfused at 38°C to prevent vasoconstriction in response to cold or room temperature perfusates. Some protocols also have successfully used Mercox without prior tissue fixation. In these instances, Mercox is infused immediately after a saline or Ringer solution flush (4). Fourth, Mercox usually begins to polymerize within 4-5 min after the catalyst has been added. This time interval may be extended by decreasing the amount of the catalyst. Fifth, we found a 5.25% hypochlorite solution to be very good at macerating the tissue, but others have used anywhere from a 20-80% KOH solution with equally acceptable results (5-6). Small blood vessels and superficial vascular beds may be cleaned from the casts to gain access to deeper regions. Ultrasonic cleaning may be used if you wish to preserve only larger vascular structures (7). Finally, we have found that air drying casts is quite satisfactory. Others have suggested freezing the casts in water and then freeze-drying them or dehydrating the casts in ethanols and critical-point drying them (8). These steps probably are unnecessary.

In summary, Mercox is a commercially available low viscosity (20-30 cps at 25°C) methyacrylate resin which can produce excellent microvascular corrosion casts which may be studied using SEM. The casts retain the fine vascular detail and three-dimensional configuration of *in situ* vessels. Consequently, corrosion casts provide excellent demonstration preparations for teaching or as research specimens. Mercox polymerizes rapidly at room temperature and may be used on fresh or perfusion fixed specimens.

ACKNOWLEDGEMENTS

Supported by NIH grant HL-31320, and American Heart Association grant 83 733, with funds contributed by the Texas Affiliate. JTH is the recipient of a NIH Research Career Development Award.

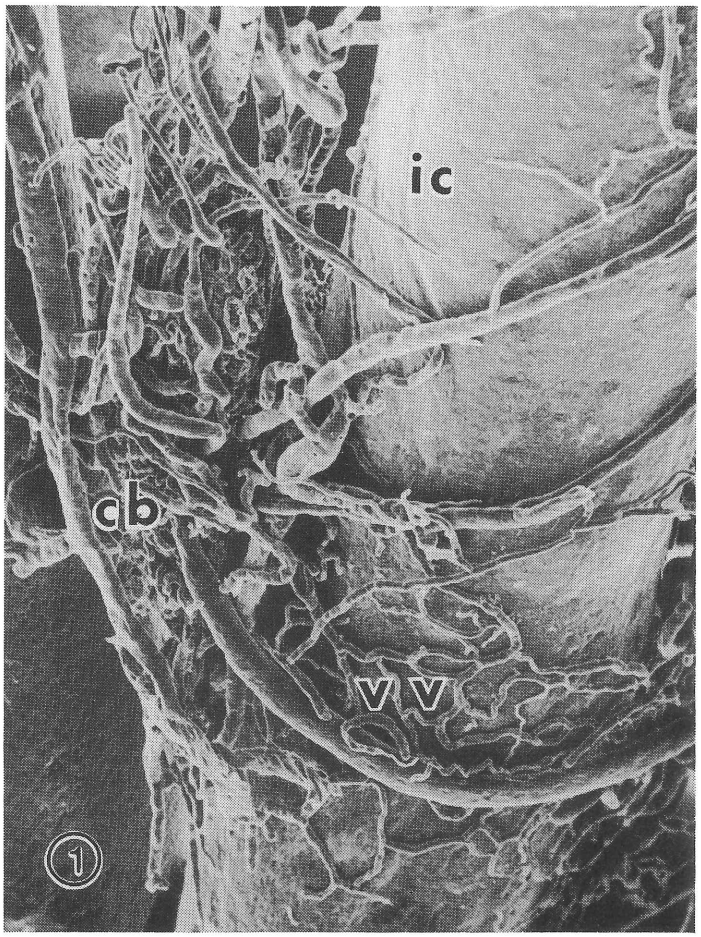

FIGURE 1: SEM of Mercox vascular cast showing the internal carotid artery (ic) and the vasa vasorum (vv) to the carotid sinus. The carotid body microvasculature also is visible (cb). x100.

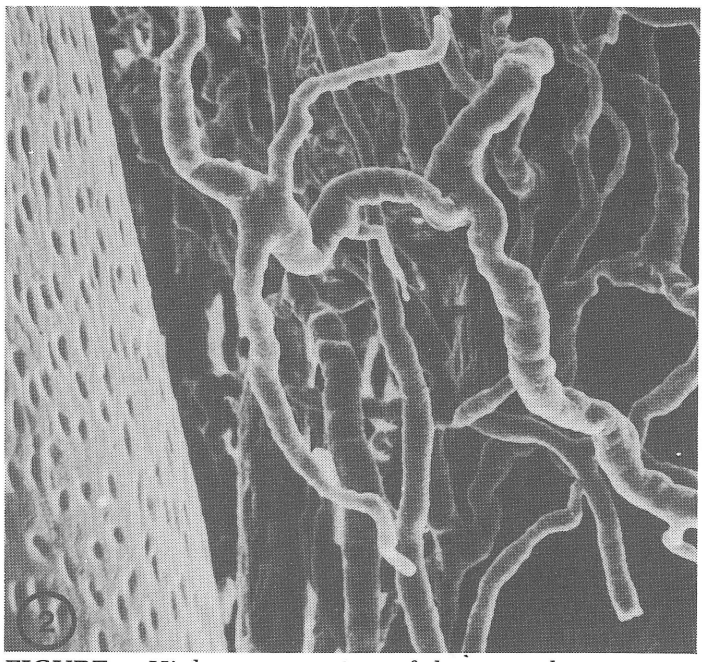

FIGURE 2: Higher power view of the carotid sinus vasa vasorum. Note the indentations of the individual endothelial cell nuclei on the internal carotid artery. The larger irregular vessels of the vasa vasorum are venules. x280.

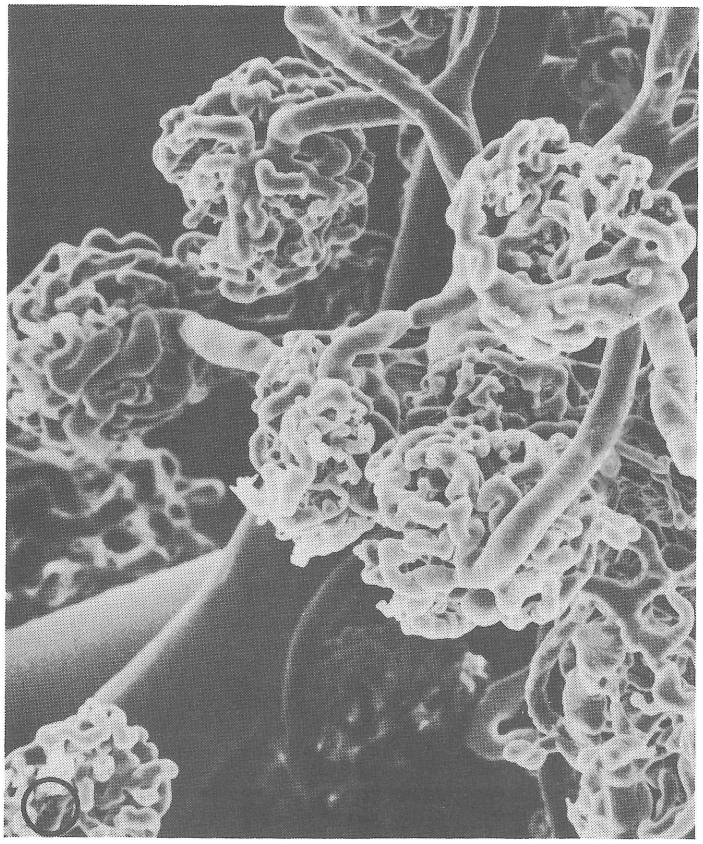

FIGURE 3: Vascular cast of the rat renal glomerulus. The afferent arteriole can be seen passing to the glomerular capillary network in each glomerulus. The microvasculature of the interior of a large organ like the kidney can be examined by slicing the preparation into slabs prior to tissue maceration. x230.

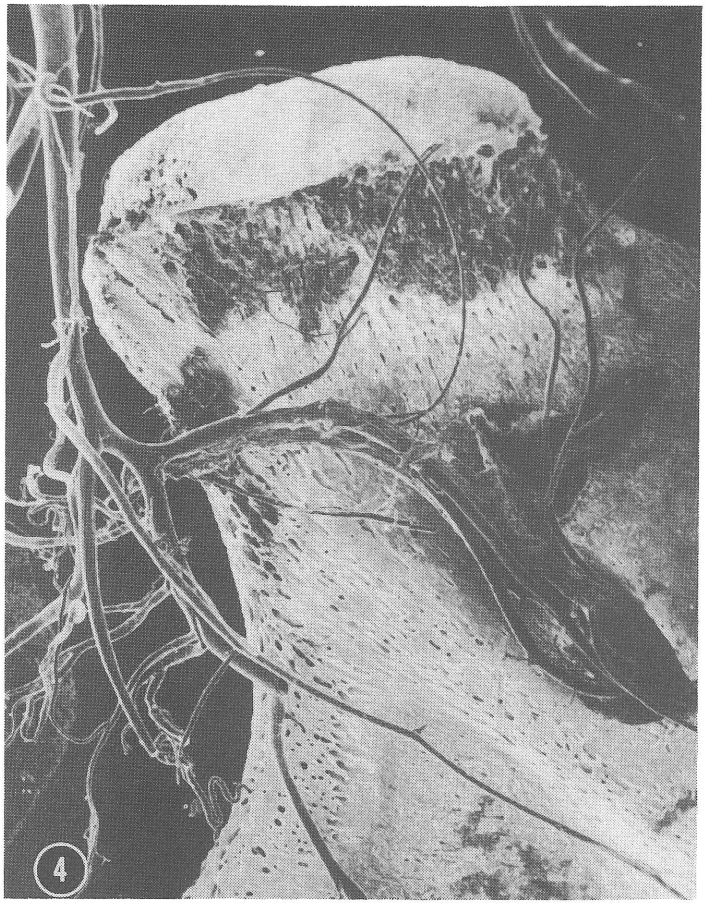

FIGURE 4: Cast of the inferior alveolar artery entering the mandibular foramen on the medial aspect of the ramus of the mandible (lower jaw). In this preparation, the relationship of the vasculature to the bone was perserved by partial maceration of the specimen. Only the soft tissues were removed. x65.

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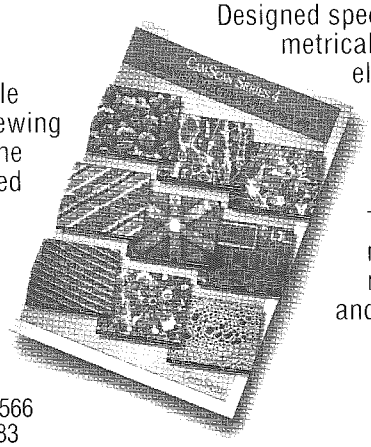

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Abstracts

FALL, 1985 MEETING BEAUMONT, TEXAS

BIOLOGICAL SCIENCES

MICROSCOPICAL STUDY OF MYCOHERBICIDE RELEASE FROM GRANULAR FORMULATIONS. W.R. Goynes and W.J, Connick, Jr., Southern Regional Research Center, New Orleans, LA 70179

Fungi that act as biological herbicides are known as mycoherbicides. These natural pathogens can be used for control of specific weeds. For efficient control it is necessary that a highly virulent form of the fungi be delivered to target weeds in effective dosages with proper timing under a variety of environmental conditions. In attempts to meet these criteria, controlled-release granules containing mycoherbicides were produced by dispensing a formulation of fungi, sodium alginate and kaolin dropwise into a solution of $CaCl_2$. After drying, the granules were 2-3 mm in diameter. A series of microscopical studies was designed to study growth, release, and regrowth of fungi on the granules. After 10 weeks storage, granules formulated with Alternaria cassiae were placed on moist filter paper in Petri dishes and exposed to light from sun lamps for 15 minutes every 24 hours for 3 days to induce sporulation. To determine the sustained release characteristics of the fungus, the granules were gently washed after 3 days to simulate the action of rain, and regrowth was observed by Scanning Electron Microscopy. Granules formulated with a pycnidium-forming Phyllosticta sp. were also studied. The fungus-containing granules were fixed with 3% gluteraldehyde in 0.05M phosphate buffer for 24 hours. Fixation was followed by dehydration in a graded water/alcohol series and critical point drying from CO_2. Scanning electron micrographs illustrate distribution of fungus within granules, emergence to the surface, and the cycles of fruiting, removal by washing, and regrowth that account for the sustained-release nature of the mycoherbicide.

RECONSTRUCTION OF THE FLAGELLAR APPARATUS IN THE DINOFLAGELLATE OXYRRHIS MARINA. K.R. Roberts, Dept. of Biology, University of Southwestern Louisiana, Lafayette, LA 70504.

The complex nature of the flagellar apparatus in the dinoflagellate Oxyrrhis marine has been determined and its three-dimensional structure is presented. The flagellar apparatus consists of several previously unknown components and component combinations. Two striated fibrous roots of differing periodicity extend to the cells left and appear to merge with one another. A large microtubuler root extends toward the posterior of cell and is proximally associated with a narrow periodicity striated fibrous component that connects the microtubules of the root with one another. Comparative dinoflagellate flagellar apparatus data are discussed.

ULTRASTRUCTURAL OBSERVATIONS OF MEIOSIS IN THE PLANT PATHOGENIC FUNGUS EXOBASIDIUM CAMELLIAE VAR. GRACILIS. E.A. Richardson and C.W. Mims, Department of Biology, Stephen F. Austin State University, Nacogdoches, TX 75962.

Members of the basidiomycetous genus Exobasidium are all plant pathogens. Traditionally most authors have placed the genus in the subclass Holobasidiomycetidae, order Exobasidiales although recently some authors have suggested that the genus should be included in the subclass Teliomycetidae. Since studies on the details of nuclear division in fungi have shown much promise in contributing to a better understanding of taxonomic and phylogenetic relationships this study of meiosis in E. camelliae var. gracilis was undertaken. Thus far various aspects of meiosis have been studied from serially sectioned basidia. The main characters of interest in this study were (1) the form of the spindle pole bodies and their behavior during nuclear division, (2) the fate of the nuclear envelope during meiosis, and (3) the nature of the spindle apparatus. Data collected thus far suggest that Exobasidium is more closely related to the Teliomycetidae than to the Holobasidiomycetidae.

ABORIGINAL UTILIZATION OF FERNS IN THE GREAT BASIN. R. G. Holloway, Dept. of Anthropology and A. J. Neumann, Dept. of Biology, Texas A&M University, College Station TX 77843.

During excavation of a late-prehistoric rockshelter, dating 900-600 years B.P., near pyramid Lake, Nevada, a number of human coprolites were recovered. These coprolites have been analyzed to determine the relative diet and nutrition of this prehistoric Indian population. The results of the analysis indicated a relatively high vegetable content in the samples. Light microscopy of the larger plant fragments suggested that they might be fragments of rhizome cortex and petiolar bases from a fern. Examination of these fragments using a scanning electron microscope revealed the general protostelic pattern and xylary elements common in many ferns of the Filicales. The identification of fern rhizomal fragments from the coprolites increases our knowledge of the aboriginal diet in the Great Basin area. This is the first documented occurrence of ferns as a component of the aboriginal diet in this region and leads to speculation as to whether it was used as food or medicine.

ELECTRON MICROSCOPIC EXAMINATION OF THE ARCHEGONIUM OF THE LIVERWORT FOSSOMBRONIA FOVEOLATA RADDI. Steven E. Ehlers and Dale M. J. Mueller. Texas A&M University, College Station, Texas 77843.

The archegonium of the liverwort undergo a remarkable cytological transformation to produce the mature egg. The archegonium consists of the venter, which surrounds the egg; the neck, the neck canal cells, which lyse to allow the passage of the sperms to reach the egg; and the lip cells, which separate to expose the neck canal. While not as spectacular as the development of the antheridium and the sperms, this process begins as a single epithelial cell and forms the second most complex structure found in the bryophytes.

MITOSIS AND CYTOKINESIS IN VEGETATIVE CELLS OF THE SUBAERIAL GREEN ALGA CEPHALEUROS PARASITICUS. R.L. Chapman and M.C. Henk, Dept. Botany, Louisiana State University, Baton Rouge LA 70803

Mitosis and cytokinesis in apical cells of actively growing cultures of Cephaleuros parasiticus Karsten sporangiate thalli were examined with transmission electron microscopy. In prophase chromosomes condense and intranuclear microtubules begin to form. Metaphase is characterized by the presence of a more or less intact nuclear envelope, a typically ordered chromosome plate, numerous apparently unorganized intranuclear microtubules and a few longitudinal perinuclear microtubules. Distinct centromeres have not been observed. Microtubule organizing centers (MTOCs) are present. In anaphase as chromosomes move apart, the nucleus elongates and the nuclear envelope appears to remain intact. Spherical daughter nuclei with large distinct nucleoli are formed early in telophase. The widely separated daughter nuclei are attached to the poles of a massive interzonal spindle at specialized sites on their envelopes. A distinct phragmoplast of interzonal spindle microtubules and vesicles of undetermined origin forms in the metaphase plain and gives rise to a thin crosswall. No involvement of the endoplasmic reticulum in plasmodesmata formation has been observed; however, some bundles of microtubules in the interzonal spindle appear to mark the sites of plasmodesmata formation. No infurrowing nor phycoplast is involved in the cytokinesis. Supported by NSF Grant BSR 83-08420 to RLC.

Various Stages of Hepatozoon canis in Dogs as Studied Using Electron Microscopy.

R. E. Droleskey, S. H. Mercer,* T. M. Craig*

U.S.D.A., Vet. Toxicol. Entomol. Res. Lab., College Station, TX 77841 and *Dep. of Micro. and Parasit., College of Vet. Med., Texas A&M Univ., College Station, Texas 77843.

Hepatozoon canis is an intracellular sporozoan parasite of dogs. More than 60 confirmed cases have been reported in Texas during the past 9 years. Dogs infected with H. canis have exhibited muscular wasting, fever, generalized pain, periosteal new bone proliferation, and extreme leucocytosis. Transmission of H. canis is via the tick Rhipicephalus sanguineus. Although the examination of blood smears and histological sections has lead to a general understanding of the life cycle of H. canis, the exact nature of each stage, and the relationship among them, has remain undetermined. In an attempt to understand the relationship among these stages, tissue samples were taken at necrospy from six dogs, with confirmed H. canis infections, for evaluation using electron microscopy. Tissues sampled included heart, liver, diaphragm, skeletal muscle, lymph nodes, and blood cells. Also sampled were in vitro cultures of H. canis infected neutrophils. The results of these evaluations will be discussed in this report.

ULTRASTRUCTURE OF BASIDIOSPORES AND CONIDIA OF THE PLANT PATHOGENIC FUNGUS EXOBASIDIUM VACCINII. C.W. Mims, Department of Biology, Stephen F. Austin State University, Nacogdoches, TX 75962.

Exobasidium vaccinii is the causal agent of red leaf disease of the lowbush blueberry Vaccinium angustifolium. Basidia of this fungus are produced in small clusters on the undersurface of infected leaves and typically bear from four to six thin-walled basidiospores. These spores are banana-shaped and measure about 4 um by 12 um. Each is initially one-celled and uninucleate but following a mitotic division a central septum develops so that the mature basidiospore is two-celled. As a result of budding, each cell of the mature basidiospore is capable of giving rise to asexual spores termed conidia. These slender, uninucleate and thin-walled spores tend to accumulate on the undersurface of the host leaf among the basidia and basidiospores. Conidia are also capable of budding to produce additional smaller conidia. As a result of the budding of basidiospores and conidia a large amount of inoculum is produced by E. vaccinii.

A Morphological Comparison of Normal and Carrier Bovine, Ovine, Murine, and Caprine Erythrocytes

R. E. Droleskey, J. R. DeLoach

U.S.D.A., Veterinary Toxicology and Entomology Research Laboratory, College Station, TX 77841

Encapsulation of compounds within erythrocytes have been performed using erythrocytes from numerous species with a variety of compounds. Carrier cells offer the unique advantage of being biodegradable, they circulate throughout the circulatory system, reduce therapeutic drug levels, prolong the systemic activity of drugs, and are useful in enzyme replacement therapy for inborn errors of metabolism. During the encapsulation process erythrocytes undergo severe stress as well as morphologic changes. The objective of encapsulation is to produce a carrier cell with as near "normal" morphology, and as high a percentage encapsulation as possible. To help determine the effect of encapsulation conditions on the production of "normal" appearing carrier cells, the scanning electron microscope was used to compare the morphology of normal and carrier bovine, ovine, murine and caprine erythrocytes. Encapsulation conditions were adjusted to produce suitable "normal" appearing carrier cells.

LOBSTER MANDIBULAR ORGAN - A HOLOCRINE SECRETORY STRUCTURE. G. D. Blaisdell, E. F. Couch (Dept. of Biology, Texas Christian University, Fort Worth, Texas 76129) and J. K. Butler (Dept. of Biology, University of Texas at Arlington, Arlington, Texas 76019). High resolution light microscopic examination of the mandibular organ of the lobster Homarus americanus shows the presence of a pattern of cell types typical of the classical picture of holocrine secretion as seen in invertebrates. This pattern involves the accumulation of characteristic secretory material in the cytoplasm followed by serial nuclear changes involving: peripheral clumping of the chromatin, the appearance of a perinuclear halo, due in part to the enlargment of the intra-cisternal space of the nuclear envelope, and contraction of the chromatin into an irregular dense mass that subsequently becomes diffuse and amorphous. Concomitantly, large masses of amorphous secretory material appear in the cytoplasm accompanied later by lattices of dense, cylindrical structures. The size of the lattice elements changes as the surrounding ground cytoplasm becomes vacuolated, disorganized, and rarified. Ultimately both the nucleus and cytoplasmic constituents become dispersed and the cell as a whole disintegrates. Electron micrographs of the regions occupied by the lattice structures are seen to contain numerous mitochondria, lipid droplets, and morphologically complex aggregates of endoplasmic reticulum surrounding lipoid masses. Previous reports implicate the mandibular organ in lipid synthesis including steroids and methyl farnesoate. The structures observed are characteristic of cells that synthesize lipids.

ENDOSYMBIOTIC BACTERIA IN THE BINUCLEATE DINOFLAGELLATE PERIDINIUM BALTICUM. J.M. Chesnick, Biology Department, Texas A&M University, College Station, Texas 77843.

Peridinium balticum is an unusual dinoflagellate that harbors a photosynthetic chrysophyte as an endosymbiont. Pienaar (1980) reported the occurrence of endosymbiotic bacteria in the cytoplasm of the dinoflagellate host as a second symbiont. A single membrane, belonging to the dinoflagellate, was observed to surround each bacterium.

TEM thin sections show these bacteria to exist as a compact unit located in the posterior or hypothecal portion of the cell. The bacteria are classified as gram positive by the ultrastructure of their cell walls. Observations of rapid freeze-fracture replicas reveal a specialization of the surrounding single membrane that occurs as a series of infoldings. Closer examination of TEM thin sections show points of contact between this membrane and the bacterial cell wall. Fibrillar extensions of the glycocalyx of the bacterial cell wall extend from the bacterial plasma membrane to the surrounding single membrane.

Endosymbiotic bacteria have been previously reported to occur in the nucleus or free in the cytoplasm of other dinoflagellate species. Some of these dinoflagellates showed an accumulation of metabolites in the cytoplasm. It was suggested that a relationship may exist between those bacteria and the accumulation of metabolites in old cells (Silva, 1967). Similar observation concerning metabolite accumulation in Peridinium balticum have not been made in this study.

CHARACTERIZATION OF THE LUMINAL SURFACES OF ATRIOVENTRICULAR (AV) ENDOCARDIAL CUSHIONS: LOCALIZATION OF SULFATED COMPLEX CARBOHYDRATES. D.A. Hay, Dept. of Biology, Stephen F. Austin State University, Nacogdoches, TX 75962.

The luminal surfaces of chick embryonic cardiac endothelium were examined for the presence and distribution of sulfated carbohydrates as demonstrated by the high iron diamine-silver proteinate (HIDSP) technique. Hearts from 4 day embryos and from cardiac endothelial monolayers cultured on collagen gels were fixed in glutaraldehyde before being stained with HIDSP. Initial results reveal a more intense reactivity for sulfated carbohydrates on the luminal than on the abluminal surfaces of virtually all cardiac endothelia. Enzyme digestion studies utilizing chondroitinase ABC (Chase) or heparitinase (Hpase) or both indicated that heparan sulfate-containing proteoglycans or heparan sulfate-containing glycoproteins were responsible for most of the HIDSP staining. Since heparan sulfate proteoglycans have been implicated in cell:substrate adhesion mechanisms, the presence of such substances on the surfaces of AV cushion endothelia may help to explain the process of cushion fusion. Supported by Grant No. G-781 from the Texas Heart Association.

DIRECT VISUALIZATION OF THE THREE DIMENSIONAL CONFIGURATION OF CELLULAR STRUCTURES. A. Cole and E.P. Armour, Phyiscs Dept., University of Texas System Cancer Center, Houston, TX 77030.

Three new techniques have been developed to directly visualize the native configurations of cellular microstructures. These include: (a) attachment and/or growth of primary or secondary cultured cells on polystyrene membrane coated stainless steel electron microscope grids; (b) selective stabilization or fixation of desired cellular structures and dissociation or digestion of unwanted or obscuring structures in intact cells attached to the polystyrene membrane; (c) visualization and recording of the total structural arrangements using a direct view stereo electron microscope system developed by A. Cole. During the past year these techniques have been used to study cell surface structures, cytoplasmic and nuclear matrices, cellular vesicles, secretory granules, and mitochondria. Both primary and secondary cell cultures from chinese hamsters, mice, and monkeys have been studied. Subtle changes related to progression through the cell cycle or to alteration induced by various physical or chemical agents have been observed. In principle, stereological features such as total organelle distributions, counts, configurations and volumes can be determined from a single stereo-pair micrograph of the whole cell.

PRECISION EMBEDDING FOR HIGH RESOLUTION LIGHT MICROSCOPY. J. K. Butler, Biology Department, The University of Texas at Arlington, Arlington, Texas 76019. A unique apparatus and method for embedding specimens in glycol methacrylate for high resolution light microscopy is described. The apparatus, consisting of a new type of open-through two part mold and an improved version of the anoxic embedding apparatus described by the author in 1984 assures uniformly dependable embedding of large numbers of specimens suitable for cytochemical and enzyme as well as morphological study. Use of the apparatus eliminates the commonest causes of tissue damage - heat and gas bubble formation during polymerization, and the commonest causes of block inhomogeneity - oxygen inhibition of polymerization and irregular water contamination of the plastic due to moisture condensation. In addition, the apparatus lends itself to the use of a unique means of tissue identification and space-saving block storage.

FINE STRUCTURE OF ASTROGLIA GROWN IN ARTIFICIAL CAPILLARY PERFUSION CULTURES. E. Tiffany-Castiglioni, T. Caceci, and K.F. Neck, Dept. Veterinary Anatomy, Texas A&M University, College Station, TX 77843.

The functional association of astroglial footplates with blood vessels is important because astrocytes may provide a channel between the blood and neurons deeper in the brain parenchyma for the passage of ions and metabolites. This hypothesized function is very difficult to study in vivo or in monolayer cultures. We have produced a 3-dimensional cell culture model of perivascular astroglia by means of an artificial capillary system. First, flat cultures of astroglia were prepared from neonatal rat cerebral hemispheres in 75 cm^2 tissue culture flasks. After 25 days, the cells were seeded in Amicon Vitafiber hollow fiber culture vessels. A culture vessel consists of a bundle of hollow, semipermeable polysulfone fibers, encased in a plastic shell. The fibers were coated with fibronectin, and astroglia were seeded on their outer surfaces. Warmed Waymouth's 705/1 medium with 10% FBS and Hepes buffer (3.68 g/l) was pumped through the lumen of the fibers (3-5 ml/min). After 13 days, the cells were fixed with 4% paraformaldehyde and examined. SEM revealed the tubes to be uniformly covered with astroglia with short processes that contacted nearby cells. TEM showed glial filaments. Gap junctions (not usually seen in monolayer cultures) were frequently located in regions where cells came together. In the brain, gap junctions are thought to facilitate ion movements between astrocytes. The cells have been hypothesized to take up K^+ discharged to the extracellular space by depolarizing neurons, and move it to areas of low concentration, i.e., to act as a K^+ spatial buffer. Our culture system should permit a direct test of this hypothesis to be made. Supported by BRSG-7-84.

NITROSAMINE AND INTERFERON PRODUCTION: BACTERIAL INDUCTION OF UROTHELIAL HYPERPLASIA AND NEOPLASIA. C.P. Davis, M.S. Cohen, M.D. Anderson, M. Gruber, M.M. Warren, University of Texas Medical Branch, Galveston, Texas.

The role of chronic bladder infections and their potential for neoplasm induction were investigated in this study. Control rats were either treated with streptomycin or had a sham operation. Experimental rats were either given repeated intraurethral inoculations of bacteria or had a stainless steel wire (nidus for infection) sutured into the bladder followed by a single intraurethral bacterial inoculation. After bladder infections (Escherichia coli, a Proteus sp. or both organisms) were established in rats for up to 24 wks., bladder tissue was examined by light and scanning electron microscopy. Rat bladder tissue and blood was examined with a plaque reduction assay for interferon production. Urine was examined after chloroform extraction by gas chromatography and mass spectrometry. Control rats showed no significant alterations in the urothelium, but infected rats showed microvillus alterations and papillomas as early as 6-8 wks. post-infection. N,N-dimethylnitrosamine, a known carcinogen, was not detected in control rat urine but was detected as early as 2 wks. post infection and found in the majority (>50%) of rats infected 12 wks. or longer. It was the only nitrosamine detected in all positive samples. In contrast, interferon was found to be either undetectable or only in trace amounts in both rat tissue or blood, although the infecting organisms could induce large amounts (up to 1000 international units) in vitro. We conclude that N,N-dimethylamine occurs in vivo in the urine from infected rats and correlates with the development of hyperplastic and early neoplastic changes in the rat urothelium.

EFFECTS OF MONENSIN ON CYTOPLASMIC CLEAVAGE IN ALLOMYCES MACROGYNUS. T. Sewall and J. Pommerville, Dept. of Biology, Texas A&M University, College Station, TX 77843.

The water mold, Allomyces macrogynus, does not possess the structural equivalent of a Golgi complex as represented by stacked cisternal elements although it does contain single smooth cisternal elements in which Golgi complex-specific enzymes have been localized. To determine if these Golgi equivalents play a role in cytoplasmsic cleavage in A. macrogynus, we used the ionophore, monensin, to probe the process of gamete formation. Treatment of cells with monensin in vitro typically causes swelling and subsequent release of dilated vesicles from the trans cisternae of the Golgi complex as well as disrupting its normal secretory function. Induction of gametogenesis in A. macrogynus with Machlis dilute salts (DS) solution containing a 10 uM or greater concentration of monensin increased the length of time required for gamete release and decreased the number of gametangia which released gametes. Addition of monensin as cleavage furrows neared completion did not inhibit release as much as in those treated at earlier stages of cleavage. Gametangia induced with 50 uM monensin in DS contained large vesicles filled with amorphous material and numerous small transparent vesicles. Cleavage furrows were greatly reduced but other events involving membrane formation appeared normal. Gametes released from monensin-treated gametangia were frequently abnormal containing numerous nuclei, flagella, and large swollen vesicles. Monensin appeared to interfer with a portion of the endomembrane system active during cleavage furrow formation. Because of the specificity of monensin's action in vitro, Golgi equivalents are implicated in the proper formation of the gamete plasma membrane in A. macrogynus.

ULTRASTRUCTURAL CHANGES OF THE LIVER FOLLOWING L-TRYPTOPHAN INGESTION. M.E. Trulson and H.W. Sampson, Dept. Anatomy, College of Medicine, Texas A&M University, College Station TX 77843.

Oral administration of L-tryptophan (250 mg/kg/day) for three consecutive days to rats produced enlarged hepatic sinusoids and vacuolated cells, many of which contained lipid. These changes persisted for at least two weeks. Since the hepatic metabolism of L-tryptophan is very similar in rats and humans, these data suggest that people who self-administer large doses of L-tryptophan for the purpose of decreasing sleep latency my be inducing hepatic pathology.

INFILTRATION AND POLYMERIZATION SCHEDULES FOR LX-112 AND POLY/BED 812 EMBEDDING MEDIA. J.A. Mascorro, Department of Anatomy, Tulane Medical School, New Orleans, LA 70112.

The epoxy resins LX-112 and Poly/Bed 812 are reliable replacements for Epon 812 in biological electron microscopy. Both media are known for their excellent sectioning and staining characteristics, and for the ability to withstand intense heat and vacuum in the microscope column. This work seeks to establish ideal infiltration and polymerization schedules in order to minimize processing time and insure that tissues are well impregnated subsequent to microtomy and microscopy. Four infiltration methods were tested: #1: 1½ total hours (½ hr. 1:1, 1 hr. full resin); #2: 3½ total hours (½ hr. 1:1, 3 hrs. full resin); #3: 6½ total hours (½ hr. 1:1, 6 hrs. full resin); #4: 21½ total hours (½ hr. 1:1, 21 hrs. full resin). Tissues were polymerized for 15-72 hrs. @ 70°C. All schedules used a very short (30 min.) initial infiltration period in 1:1 propylene oxide and resin followed by a final infiltration for 1, 3, 6 or 21 hours. Except for Schedule #1, all others produced results ranging from good to excellent. In general, the longer periods produced the best results in terms of ease of sectioning and block hardness. Both LX-112 and Poly/Bed 812 were prepared according to 3A:7B hard ratio. Because Schedule #3 could be completed easily during the course of a working day, and since it gave very acceptable results, it was preferred over Schedules #2 or #4. Polymerization for 24 hours at 70°C was sufficient to yield very firm blocks from the 3:7 ratio embedding media, although the blocks were sectionable after only overnight hardening.

ELECTRON MICROSCOPIC FINDINGS IN AMIODARONE TOXICITY
Robert S. Zirl, M.D., Mattie J. Bossart, Ph.D., and Jerry DePriest, M.D., Dept of Pathology, St. Luke's Episcopal Hospital, 6720 Bertner, Houston, TX 77030

Electron microscopy has become an indespensible tool in identifying structural targets of injury by exogenous pharmacologic agents. Amiodarone hydrochloride, a benzofuran derivative, has had long term use in Europe to treat refractory supraventricular arrhythmias. There have been few reports of toxicity. This investigational drug is under study in the U.S. for use in the treatment of arrhythmias that are unresponsive to conventional therapies. Toxic pulmonary reactions, identified by abnormal chest radiographs, are serious side effects that appear to be dose dependent and reversible.

We report on electron microscopic studies of patients with presumed amiodarone associated pulmonary disease who underwent open lung biopsy or fine needle aspiration as diagnostic procedures. One case has radiographic changes of pulmonary fibrosis, with histologic confirmation of marked interstitial fibrosis accompanied by large areas of necrosis. The second case has radiographic findings of multiple nodular lesions with central cavitation. Aspiration biopsy is diagnostic of squamous cell carcinoma.

Distinct membrane-bound and non-membrane bound cytoplasmic inclusions are present in both cases. These inclusions consist of homogeneous electron-dense material and lamellar bodies or combinations of both. The structures are present in Type II pneumocytes, endothelial cells, smooth muscle cells, fibroblasts, macrophages, neutrophils and tumor cells. The possibility that this is a drug-induced phospholipidosis will be discussed.

MATERIAL SCIENCES

STEREOIMAGING TECHNIQUE FOR MAPPING CRACK TIP STRAIN FIELDS IN GRAPHITE/EPOXY COMPOSITES. M.F. Hibbs, Mechanical Engineering, Texas A&M University, College Station, TX

Stereoimaging techniques are used to examine the strain fields associated with delamination in graphite/epoxy composites. Stereo-photographs of a delamination crack tip in both mode I (opening) and mode II (pure shear) loading conditions are made using a specially adapted tensile stage within a scanning electron microscope (SEM).

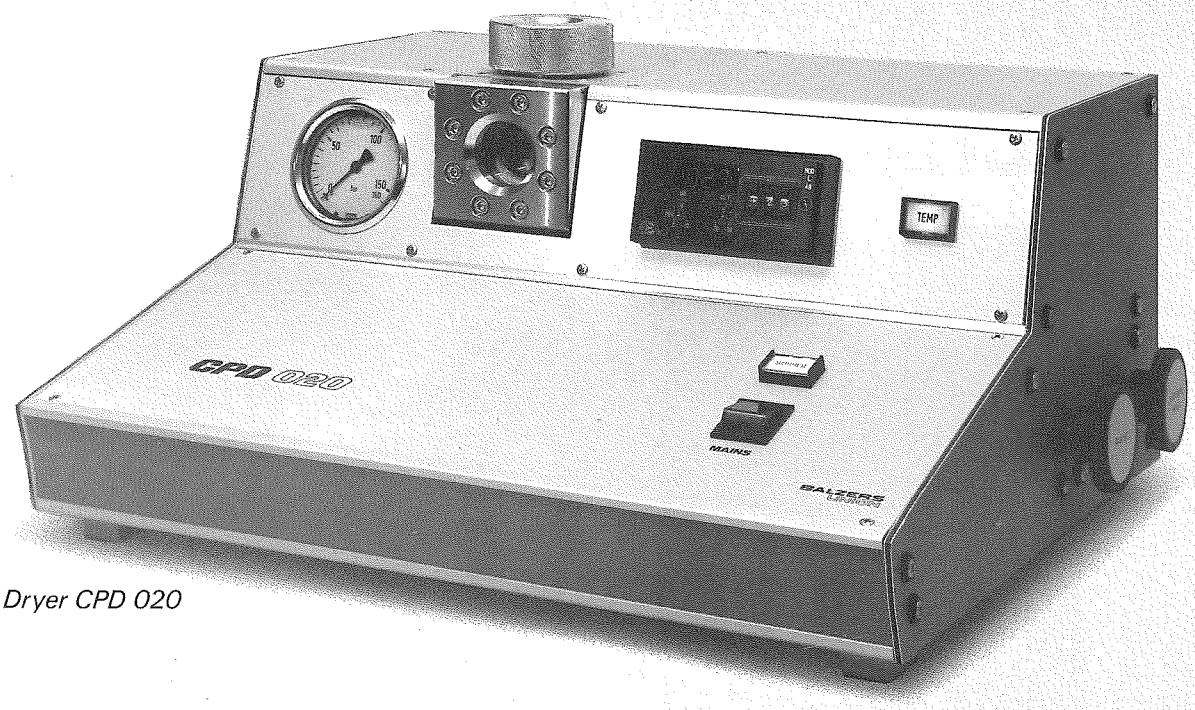
The advantage of stereoimaging is that high-resultion strain field maps can be made without prior knowledge of the stresses. By photographing the specimen before and after deformation, the relative displacements are directly measured by viewing the photographs through a stereoscope. The in-plane strains are then calculated by differentiating this displacement field.

From the displacement and strain field maps generated, the effects of macroscopic loading as well as the role of fiber constraint on the deformation and resin yielding in the region of the delamination crack tip are examined.

SEM ANALYSIS OF THE STRUCTURAL STATE OF UNDEFORMED AND DEFORMED OCEANIC BASALTS. K. R. Wilks, Center for Tectonophysics, Texas A&M University, College Station, TX 77843

Microstructures of three layer 2A basalts from sub-basement depths of 138-142 m cored from DSDP Site 483B (Tamayo Fracture Zone) have been examined in secondary and backscattered electron modes to augment research into the mechanical behavior of oceanic basalt at high temperatures and pressures. Analyses were undertaken to understand further the structural state of undeformed and deformed basalts.

SEM analyses of polished surfaces of undeformed basalt reveal the presence of pores and cavities in all three samples, however, pore sizes and abundances vary considerably over the 6 m interval investigated. Hydrothermal alteration minerals, identified as zeolites and smectites (mixed-layer clays) line pores in each basalt, however their absence from other pores shows not all pore space is interconnected. Backscattered electron images of polished optical thin sections show that alteration is restricted to grain boundary areas between primary phase minerals and that crack densities in deformed basalts are greater than in undeformed basalts. Deformed samples are characterized by absence of positive relief and the presence of pores that survive high temperature and pressure deformation. Pores are flattened perpendicular to the maximum compressive stress, and some contain undeformed mineral grains. Throughgoing cracks are observed in pores suggesting crack-pore interactions occur during deformation. Variations in porosity are correlated with mechanical behavior as the basalt with the lowest porosity is consistently stronger than the more porous basalt.



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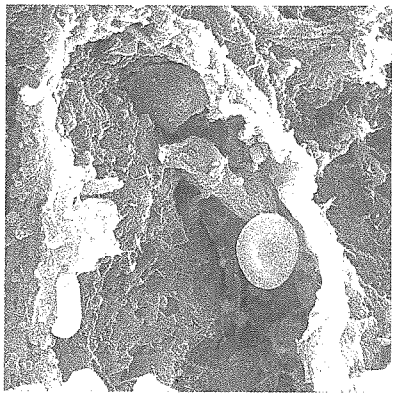

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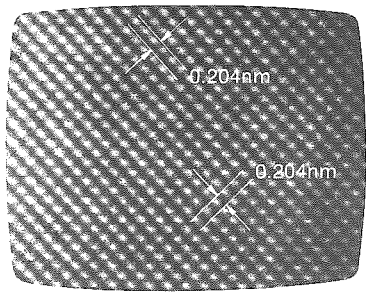
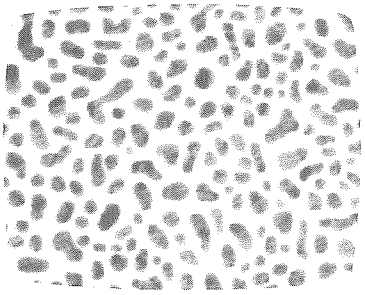


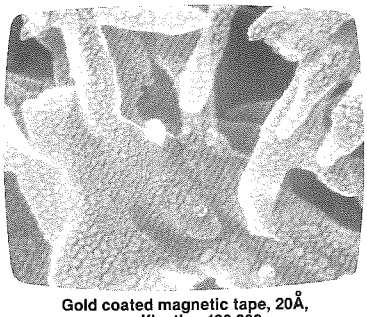
Crystal lattice (200) of gold, 2.04Å

STEM Mode



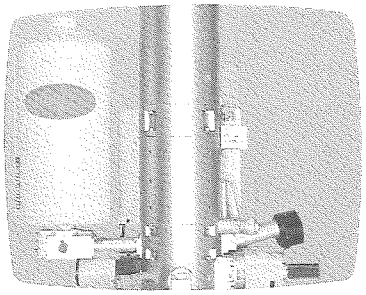
Evaporated gold, 10Å, magnification 300,000x

SEM Mode



Gold coated magnetic tape, 20Å, magnification 100,000x

X-RAY



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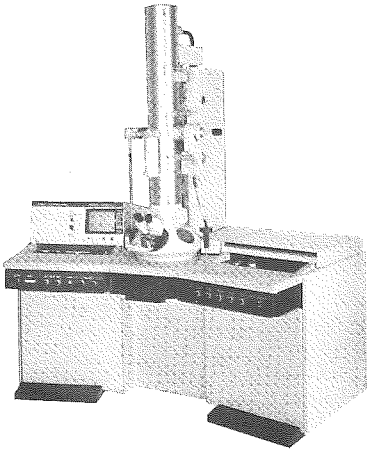

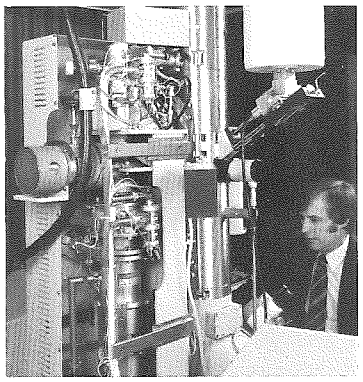

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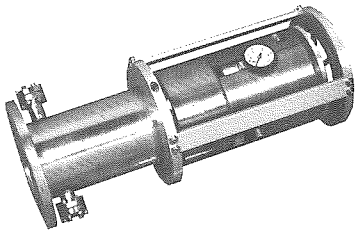

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5. Diamond knives
6. Serial sectioning
7. SEM specimen preparation
   a. Critical point drying
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11. Rapid freezing
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15. Cryultramicrotomy
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17. Microincineration
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22. Other: specify _____

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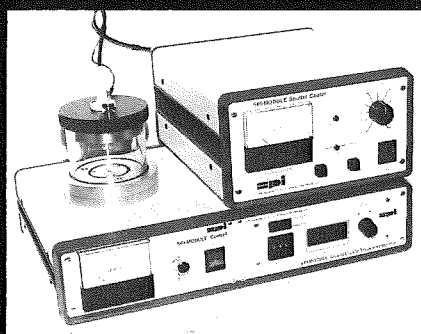

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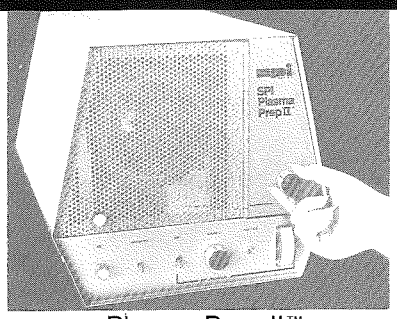

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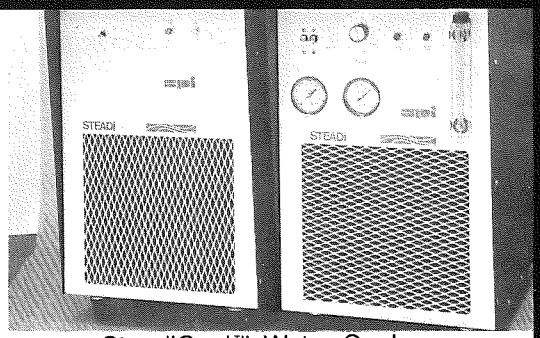

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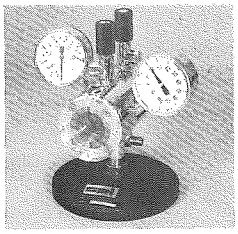

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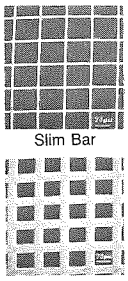

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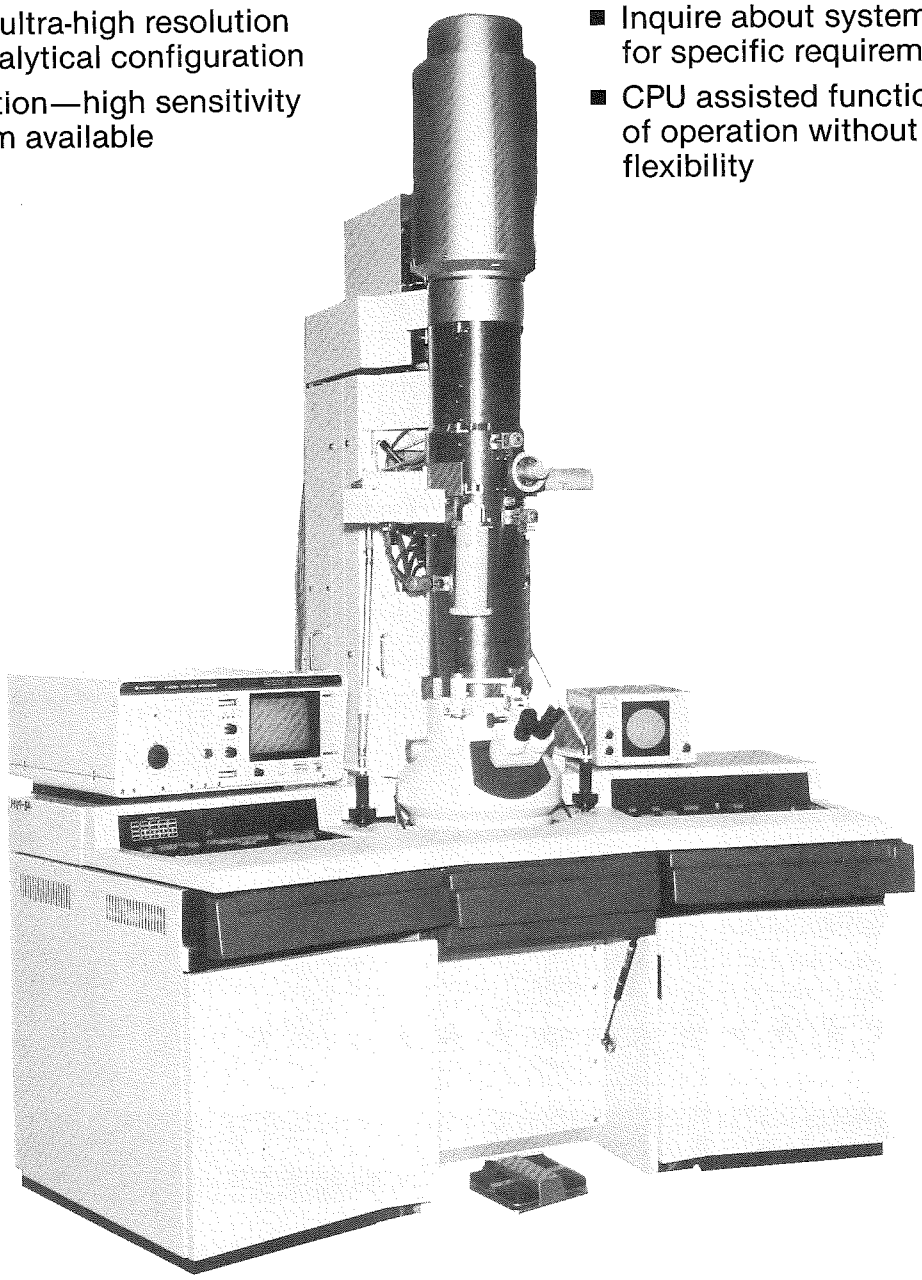

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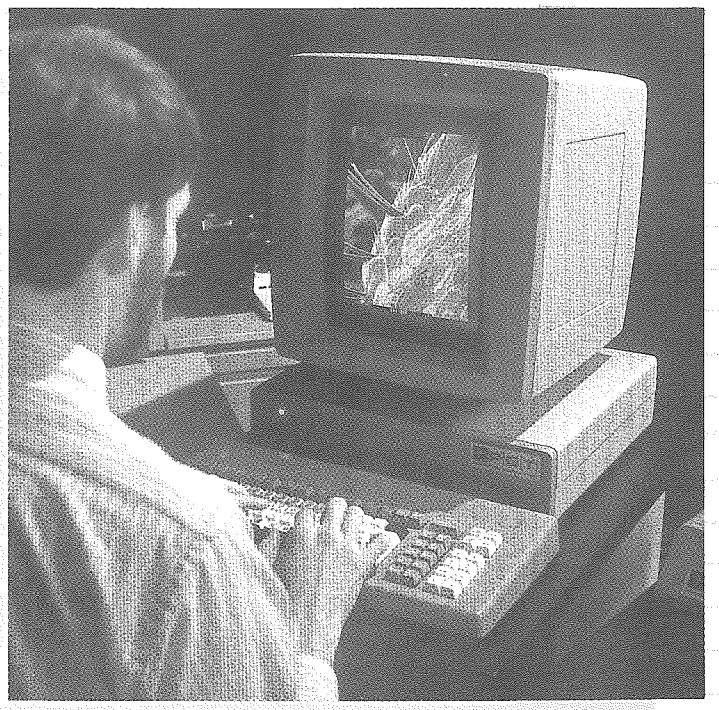

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