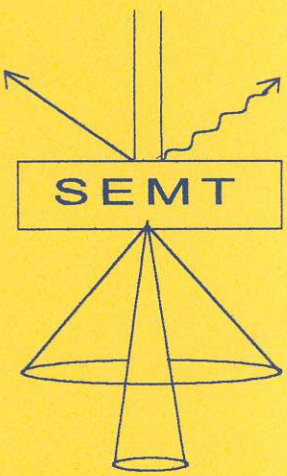
SEMT

ONE DAY MEETING

Wednesday 25 March 1998

London School of Pharmacy
Brunswick Square
London WC 1

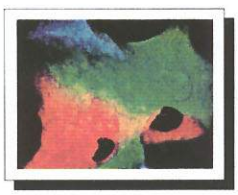


Society of Electron Microscope Technology

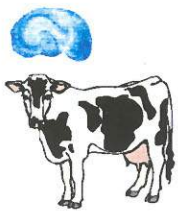
One - Day Meeting Wednesday 25th March

The School of Pharmacy, 29-39 Brunswick Square London WC1N 1AX

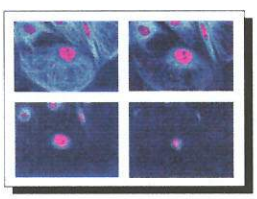
Speakers



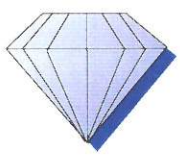
Professor A.Boyde (Anatomy dept.UCL)
Special methods for Hard Tissue Microscopy



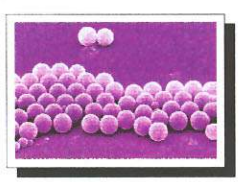
B. Cooley (Central Veterinary Labs, Newhall, Surrey)
The Use of Electron Microscopy for the Diagnosis of Transmissible Spongiform Encephalopathies; an Update



A.Todd (Glasgow University)
Combined Confocal and Electron Microscopical Techniques used in Multiple Labelling by Immunocytochemistry in the Central Nervous System



R. Hough (Planetary Sciences Research Institute, Open University)
Electron Microscopy of Diamonds and Silicon Carbides formed by Meteoric Impact



A.Wildeman (Coates Lorilleux ltd; Orpington)
Microscopy Application in Printing Ink Technology

D.Samuel (Institute of Archeology, UCL)
Scanning Electron Microscopy and Ancient Egyptian Beer

Registration and applications for poster presentations
to the Hon Secretary Dr G M Lewis, 19, Bellfield Avenue Harrow , Middx HA3 6ST
0181 428 4264 (Phone, Answerphone, Fax)

Registration fees (including abstracts & lunch)

Members	£15
Non- members	£20
Students	£10

THE SOCIETY OF ELECTRON MICROSCOPE TECHNOLOGY

Prospective members should obtain an application form from
Jill Lewis (Hon Secretary), 19 Bellfield Avenue, Harrow Weald, Middx, HA3 6ST .
The annual subscription is £10.
Current committee members are listed below and are available for further information.

ACKNOWLEDGMENTS

The SEMT wishes to express special thanks to :-

The London School of Pharmacy as hosts
Emitech Ltd. for sponsorship

and to the following companies for supporting the Trade Exhibition -

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Oxford Instruments
Taab Equipment Ltd.

FUTURE PROGRAMME

Wed 20 May	2.00 p.m.	**The Future of Imaging**	School of Pharmacy
Wed 10 June		Visit to Open University with Leica Demonstration	
Wed 14 Oct		Visit to EMU at Natural History Museum	
Wed 9 Dec	2.00 p.m.	**3D from 2D**	School of Pharmacy

WELCOME TO THE ONE DAY MEETING

PROGRAMME

9.15 **Registration**
Coffee - Trade Exhibition

10.00 **Microscopy applications in printing ink technology**
Alison Wildman (Coates Lorilleux Ltd., Orpington, Kent)

10.40 **Special methods for hard tissue microscopy**
Prof Alan Boyde (Dept Anatomy, University College London)

11.20 **Scanning electron microscopy and ancient Egyptian beer**
Delwen Samuel (Inst of Archaeology, Univ Coll London)

12.00 **Combined confocal & EM techniques used in multiple labelling by immunocytochemistry in the central nervous system.**
Andrew Todd (Dept of Anatomy, Univ of Glasgow)

12.40 **LUNCH**
Trade Exhibition and posters

2.40 **EM of diamonds and silicon carbide formed by meteoric impact**
Rob Hough (Planetary Science Research Inst., Open University)

3.20 **The use of EM for the diagnosis of transmissible spongiform encephalopathies: an update.**
Bill Cooley (Central Veterinary Labs., New Haw, Surrey)

4.00 TEA and Close

S.E.M.T. 25 March 1998 One-Day Meeting

Microscopy applications in printing ink

Alison Wildman
Coates Lorileux Ltd, Orpington, Kent

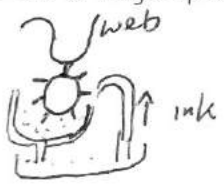
Printing ink must:
- have required visual characteristics - colour, gloss, etc
- print by a given process
- dry under specified conditons
- adhere to the given material
- have specific resistance properties.

The screen-printing process is the only one which can be used for funny-shape surfaces.

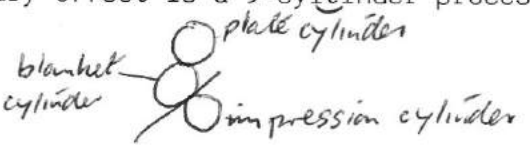
Flexograph involves 4 rollers, and is used for long strips, e.g. labels,cartons.

Recycled paper has a lot of short fibres which build up and clog the dot gaps between the dots on the roller

Gravure is a high-quality process; because of the cost of making the plates, it is only used for long runs.



Rotary offset is a 3-cylinder process



Dots of different colours can be printed on top of each other; each dot is 20 - 80 μm diametere

Ink is composed of a colourant, a vehicle (= resin + solvent), & additives.
Dyes bleed easily, and are only found in over-lacquers.
Pigments are found in most inks; they are organic or inorganic, plus extenders

The performance of an ink depends on the degree of dispersion - gloss, etc.
Some pigments do change under the electron beam.

The laboratory uses TEM, optical microscopy, image analysis, and many other techniques. Polarised light is particularly useful.
During shrink-wrapping, a glossy surface may wrinkle, as the supporting paper shrinks.

Special methods for hard tissue microscopy

Prof. Alan Boyde
Dept. of Anatomy, University college London

Polarised light gives a lot of ultrastructural inforation. For linearly-polarised light, birefringence etc, two $\frac{1}{2}\lambda$ plates are used; it is now possible to use circularly-polarised light, with an additional two $\frac{1}{4}\lambda$ plates.

The signal from collagen remains constant, even with the plates.
Polaristion colours show the loading in compression or tension.

The auto-fluorescence is proportional to the degree of mineralisaiton, in bone matrix.
Formaldehyde fixation causes fluorescence everywhere.
Staining with brilliant sulphaflavin is in inverse proportion to the degree of mineralisation. Osteoid is intensely stained by brilliant sulphaflavin.

The secondary electron signal is strongly slope-dependent, so stereological studies are oftne necessary.
The back-scattered signal has directional dependency.

Scanning electron microscopy & anciant Egyptian beer

Delwyn Samual
Inst. of Archaeology, University College London

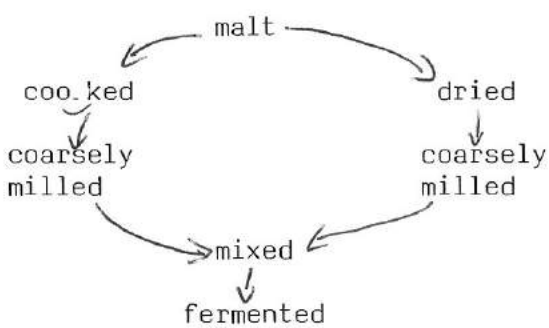
Her work centres on the interactions of humans & plants, e.g. for food.
Barley was used a lot by the ancient Egyptians; also Emmer wheat, in which the husk does not separate readily from the kernal during winnowing.

A group of ancient pottery figures was shown; a man pounding with a pestle, two others with containers and vat; women with sieve, oven, and milling.

The initial theory was that bread was lightly baked to preserve the yeasts; then crumbled into water and fermented. The exact ingrediants are unknown, so it difficult to test.
Funerary evidence is preserved in the desart areas, not in the flood plain.
Barley has been identified, broken coarsely by milling.

The action of dry heat on grain causes the protein to shrink away from the starch granules. If a small amount of water is added, the starch begines to change, and depending on the amount of water, will become fused and glossy.
Amylases make pits on the surface of the starch granules, then hollow out the granule.

Yeasts can be distinguished easily from the starch granules, by the presence of the bud scars.
Tests show that the grain must have been sprouted before it was exposed to the heat.
When modern yeasts are air-dried, they collapse and the bud scars cannot even be seen.
EM showed that the starch in the ancient beers was a bit distorted but not pitted; so there had been no attack by amylase, and so bread had not been used in making the beer.
Bread can sometimes be made with malt.



The dried malt has active enzymes. Cooked malt has denatured starch, and so is more susceptible to active enzymes, to produce sugars.
The yeast is strained from the chaff.
Emmer wheat is rare today, because it is more trouble to separate the chaff.

The Scottish & Newcastle brewery has produced Tutankhamun beer, in a limited edition of 1000 bottles, following a recipe as close as possible to the proposed one. It was rather cloudy. The beer had been made in a Victorian maltery; and flavoured with coriander seeds and juniper berries.
They intend to try again, using replicas of the ancient tools.

Combined confocal & EM techniques used in multiple labelling by immunocytochemistry in the central nervous system.
Andrew Todd
Dept. of Anatomy, University of Glasgow

He used confocal microscopy on antibodies to four two neuronal peptides; searching for contacts between synapses and dendrites.
Antibodies from different species were used for different antigens.

Confocal microscopy
- can detect up to 3 antibodies easily
- sampling gives rapid detection of contacts
- 3-D reconstruction is possible
But Z- resolution is less good than X & Y resolution; so it is not so easy to be sure that contacts are synapses.

Usual microscopy
fixed 4% formaldehyde
section 60 µM with Vibrotome
apply immuno-cocktail of primary antibodies (usually with Triton detergent)
fluorescence-labelled species-specific secondary antibodies (+ Triton detergent)
Mount in glycerol-based medium
View with confocal microscope.

Confocal / EM
- fixative + 0.2% glut
- for immunocytochemistry, avoid detergent
  (50% ethanol can be used to enhance penetration)
- additionally: - biotinylated secondary antibodies (can bind despite the presence of fluorescent secondary antibodies)
  - avidin-HRP / DAB / osmium

--> dehydrate and embed for EM

Because the earlier stage was mounted in glycerol, it is possible to float the coverslip off and re-stain with biotinylated antibodies for EM.
It is essential to get all the required confocal images before starting to process for EM.
It is not necessary to use distinct EM labels; the distinctions can be made on the basis of the fluorescence results.
The two fluorescent markers together give a white blob where they co-incide.

EM of diamonds & silicon carbide formed by meteorit impact

Rob Hough
Planetary Science Research Inst, Open Univerdsity

The distribution of known meteoric craters correlates with access and where research is possible; many are known in eastern USA, Scandivia, Australia. He has studied especially the Chicxlub crater in Mexico, from 40,000 years ago, and the Ries crater in Germany.
Craters can be identified especially by the presence of excess iridium, and of shocked quartz.

The Ries crater is particularly well-preserved; the town is situated within it, built of the swaleite stone; this contains diamonds up to 300 μm diameter, hexagonal, seen by cathodoluminescence; and silicon carbide. There is some inter-growth of the silicon carbide and diamond.

Matter from the Chicxlub crater was ejected a long way.

The Ries church and town are built of the swaleite; local dignitaries were concerned that diamond hunters would come and chip bits off, not realising how small the diamond fragments are - TEM needed to see them - and low concentration.
The iridium is a relic of the actual meteorite.

EM for the diagnosis of transmissible spongiform encephalopathies: an update

Bill Cooley
Central Vetinary Labs, New Haw, Surrey

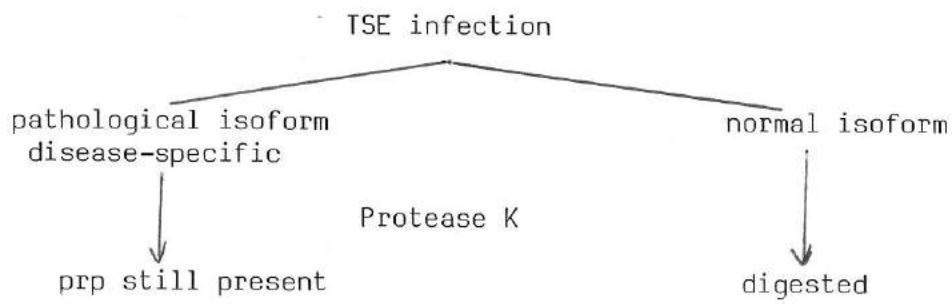
Scrapie has not been reported from Australia, New Zealand, or some countries of Europe and South America.

Transmissible Spongiform Encephalopathies are always fatal; there is a long incubation period, neurodegeneration of the Central Nervous System; no immune or inflammatory response; resistance to heat, radiation and formalin. Nucleases do not affect the infectivity.

In scrapie, the animals scrape off their own fur.

Theories:
- unconventional virus
or - virino - a core of non-translated nucleic acid associated with other proteins
or - prion - small proteinaceous infectious particles.
The prion is now considered to be the most likely.

Prions are small virus-like particles, 10-12 nm wide; seen as rods, tubules etc since 1974. In scrapie, they have been shown to be a proteinase-resistant protein, i.e. a normal cellular glycoprotein.



Scrapie-associated fibrils show linearity, occasionally bending; amyloid-like. Single pairs are 40-60 nm long; double pairs are 100-120 nm long. There are significant differences between different species, and even different breeds, of sheep. The fibril and cross-over lengths have now been shown to be two or three times the lengths indicated by previous studies.

PrP^{SC} has been seen in lymph nodes, spleen. Scrapie-associated fibrils have been seen in all of the brain and spinal cord, but NOT in sciatic nerve.; also in spleen & lymph nodes, and occasionally in other tissues.

The infectious matter is ingested, and from the gut it passes to the visceral lymph nodes, Peyer's patches and spleen; thence to the nerves, then to the Central nervous System.

The likelihood of detecting fibrils depends on the region of brain from which the sample has been taken; cervical spinal cord is OK for detection.

Detection of fibrils is possible on fixed material, and even archival wax-embeded tissue blocks.
Western Blot is better and more sensitive for detecting infection, than depending on detection of scrapie-assiciated fibrils.

Vacuoles are seen only in the Central Nervous System, not in the spleen.

The tissue must be handled in safety cabinets.

Abstracts of papers

Microscopy applications in printing ink technology

Alison Wildman (Coates Lorrileux Internatl., Orpington, Kent)

How many of you, I wonder, wander around your local Sainsbury's or Tesco's, studying the packaging of the goods you buy? Most people do not think about this aspect of their weekly shopping, but I can assure you that there is as much, if not more, technology in the wrapper as there is in the contents! Have you ever thought about the way your newspaper or magazine is produced? Or considered how much of your everyday world is influenced by printed material - posters, advertising, clothes, wallpapers, labels - even the laminated worktops in your kitchen are likely to have been printed!

As one of the world's largest ink manufacturers, Coates Lorilleux produce printing inks for all of these and many other applications. We are part of the Speciality Chemicals Division of the TOTAL Petrochemicals business, a privately owned French Company, and have wholly-owned subsidiaries in many countries worldwide.

The purpose of this talk is to introduce you to the technology of printing ink in general and then to demonstrate the usefulness of microscopy in problem-solving and research into current and future products. Coates Lorilleux set up a dedicated microscopy facility at our Research Laboratories in Kent around 6 years ago. The primary objective was to study pigments and dispersion to further our own understanding of our products. However, the ability to look at both our own and our customer's problems under the microscopes has enhanced the technical support we offer. Three main techniques will be discussed:- optical microscopy, with FT-IR microspectrometry, scanning EM and transmission EM.

Optical microscopy is an essential "first-look" for all our samples. Most print or ink problems, such as "hickies" on a print, or contaminants or poor dispersion in an ink, are easily seen with the resolution offered by this technique. The types of sample which will be discussed include cross-sections through packaging materials to study film and varnish thicknesses and samples of printed material and inks relating to various printing processes.

Scanning EM is the real work-horse of the section. The sample preparation is relatively quick and the high depth of field and resolution give excellent images. The use of X-ray microanalysis provides an added dimension of analytical capability to complement our other analytical techniques.

Transmission EM is really a research tool for us. The sample preparation methods available to us are limited to the study of dispersion and homogeneity at the microscopic level. Some cross-section studies have been undertaken, but the time required to prepare samples usually prohibits its use in a problem-solving situation.

Special methods for hard tissue microscopy

Prof Alan Boyde (Dept Anatomy, University College London)

Scanning electron microscopy and ancient Egyptian beer

Delwen Samuel (Inst of Archaeology, University College, London)

Beer was a staple item of diet for the ancient Egyptians. We know from documents that it played a major role in daily life, in the economy, and was an essential part of rituals. Egyptologists have studied ancient brewing for many years, but have relied mainly on artistic depictions. As a result, many aspects of ancient Egyptian brewing have remained unresolved. Recently, new data about brewing has come to light using scanning electron microscopy of desiccated beer residues. The arid climate of Egypt has preserved food remains in excellent condition, both on a macroscopic level and at the microscopic scale. As a result, starch granules, yeast cells and other materials have been observed. The structure of the starch in particular has allowed a reconstruction of ancient Egyptian techniques, which differs substantially from previous ideas. Ancient Egyptian brewing involved a two-part cereal preparation process, malting, cooking and filtration, as well as fermentation. It appears to resemble traditional African brewing methods which are widely used today.

Combined confocal and EM techniques used in multiple labelling by immunocytochemistry in the central nervous system

Andrew Todd (Dept of Anatomy, University of Glasgow)

One of the major goals of neuroanatomical research is to identify synaptic connections between neurons in the CNS. Because of our improved knowledge of the chemistry of neuronal populations together with an increasing range of antibodies against neuronal antigens, immunocytochemical techniques are proving extremely useful tools in studies of this kind. The combination of immunofluorescence and confocal microscopy has revolutionised studies of neuronal connectivity, both because of the high spatial resolution of the confocal microscope and its ability to generate 3-dimensional images, and because two or three different antigens can be reliably distinguished with different fluorescent dyes. Although appositions between neurons can be recognised with the confocal microscope, EM is needed to confirm that synapses are present at points of contact. We have developed a method by which Vibratome sections of CNS can be reacted with two or three antibodies, examined with a confocal microscope and subsequently be prepared for electron microscopy. The method is based on the sequential use of fluorescent secondary antibodies (for confocal microscopy) followed by biotinylated antibodies and immunoperoxidase staining (for EM). This approach combines the advantages of confocal microscopy (including the possibility of sampling large amounts of tissue rapidly) with that of EM (the ability to identify synapses).

Electron microscopy of diamonds and silicon carbide formed by meteoric impact

Rob Hough (Planetary Science Research Institute, Open University)

There are approximately 150 known meteorite impact craters; they are recognised initially by a circular structure either exposed at the surface or buried and therefore imaged using geophysical techniques. A search then takes place for chemical and mineralogical signatures of impact processes. These include shock structures in quartz and zircon and also high pressure polymorphs of certain minerals. We have searched the ejecta from the Chicxulub crater in Mexico and the Ries crater in Germany for the high pressure polymorph of carbon, namely diamond. Using acid digestion, a resistant residue is formed which is subsequently investigated using petrological and electron microscope techniques. In a residue of the ejecta from the Chicxulub crater using TEM combined with EDS and SAED we identified cubic diamonds up to 10 nm in size occurring as clusters; they occur at locations in New Mexico, Colorado and Montana. Closer to the crater, i.e. in N.E.Mexico at a site named Mimbral using SEM (again of a residue) we found much larger polycrystalline diamonds up to 30 μm in size; TEM identified them also as cubic. In a residue of impact rock from the Ries crater in Germany, a petrological microscope study identified diamonds up to 300 μm in size, again polycrystalline with a pseudo-hexagonal form. Also present, however, were silicon carbide grains up to 100 μm in size and fine grained skeletal laths. These laths were studied using TEM and EELS and found to be diamond, silicon carbide, and in one case the two minerals apparently intergrown. The large diamonds are thought to have been formed by direct shock transformation of graphite from the target rocks whereas the fine laths are interpreted to be condensation products from the impact produced fireball and would have been formed in a similar process to chemical vapour deposition.

The use of electron microscopy for the diagnosis of transmissible spongiform encephalopathies: an update

Bill Cooley (Veterinary Laboratories Agency, New Haw, Addlestone, Surrey)

The transmissible spongiform encephalopathies (TSEs), or prion diseases are a complex group of chronic, fatal neurodegenerative disorders. The animal TSEs include scrapie, bovine spongiform encephalopathy (BSE), chronic wasting disease of mule deer and elk, transmissible mink encephalopathy and feline spongiform encephalopathy. The TSEs of man are Creutzfelt-Jakob disease, Gerstmann-Straussler-Scheinker disease and kuru. They are characterized pathologically by vacuolation and astrocytosis, which gives the classical spongiform change within the brain. Two additional diagnostic criteria for the TSEs, the detection of disease-specific fibrils by TEM and the detection of the main constituent of the fibrils, an abnormal protease-resistant neuronal membrane glycoprotein (termed PrP^{Sc}) by Western immunoblotting have also been used.

This presentation will mainly concentrate on our involvement in research and diagnosis of the TSE's using EM detection of scrapie-associated fibrils (SAFs) These disease-specific structures were first identified by TEM in brain extracts from affected mice and hamsters experimentally infected with scrapie (Merz 1981 & 1984). Following these publications, the EM Unit began work on scrapie of sheep and goats (the archetype of the TSEs). This work involved the detection of SAF according to the method of Hilmert and Diringer (1984) as an aid to the diagnosis of natural sheep scrapie. This work was quickly expanded following the recognition of BSE, the scrapie-like or prion disease of domestic cattle (Wells 1987). Since then the EM Unit has developed and modified the extraction procedure in order to develop a diagnostic technique for both scrapie and BSE. This procedure requires the use of unfixed brain material (1 g of CNS tissue: the grey matter from spinal cord plus caudal medulla is ideal) The CNS tissue is then subjected to detergent extraction with N-lauroyl sarcosine, followed by various centrifugation steps at low and high sedimentable forces to pellet the abnormal protein, before treatment with the enzyme Proteinase K. The "surviving" sedimental and Proteinase K resistant protein PrP^{Sc} can then be visualised as SAF following negative staining with 2% phosphotungstic acid in the EM.

A brief historical review of other EM investigations of TSEs and their findings will also be given, which will highlight the findings of small virus-like particles and tubular vesicular structures from TSE-affected animals. A summary of recent published work comparing the sensitivities of SAF detection with histopathology and Western immunoblotting, the recovery of SAFs from autolysed, chemically fixed and peripheral tissues as well as the effects on SAF recovery after long term storage of brain tissue will also be discussed.

Hilmert, H and Diringer,H (1984) *Biosciences Reports* **4**, 165-170

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Merz, P.A., Rohwer, R.G., Kascsak, R, Wisniewski, H.M., Somerville, R.A., Gibbs, C.J.R and Gajdusek, D.C. (1984) *Science* **225**, 437-440

Wells, G.A.H., Scott, A.C., Johnson, C.T., Gunning, R.F., Hancock, R.D., Jeffrey, M,, Dawson, M and Bradley, R (1987) *Veterinary Record* ***121***, *419-420*

S.E.E.C. Romania

Anne Drewe (Dept of Microbiology, ICSTM, Charing Cross Hospital)

An update of our work and contacts with Romania.

Synaptic and neuronal morphometry in the hippocampus of apolipoprotein (apoE) knock-out mice.

K.A.Cambon[1], M.G.Stewart[1], H.A.Davies[1], C.Large[2] and G.Higgins[2] ([1]Dept of Biology, Open University, Milton Keynes and [2]Glaxo-Wellcome, Stevenage)

In Alzheimer's disease (AD), synaptic pathology is a major lesion that has been correlated to cognitive impairment. The molecular layer of the dentate gyrus in the hippocampal formation is one of the first regions affected by synaptic loss. The three major forms of human apolipoprotein E (apoE2, E3 and E4) are associated with differences in the age of onset of AD. However, little is known of the role that apoE plays in normal brain function and pathology and whether there is any relationship between the 3 isoforms of AD and synaptic or neuronal structure in the hippocampus. One approach is to study apoE in the brain using apoE transgenic mice.

As a first step we have undertaken an electron microscope study of the hippocampus of 8 months old apoE deficient mice (knockouts KO) and wild type controls. ApoE KO were Maeda derived (UNC, North Carolina), backcrossed at least 6 times into C57/BL6 mice. Synapse and neuronal density was estimated in several subregions of the hippocampus using unbiased counting methods.

Our results show that total synapse density in the middle molecular layer of the Dentate Gyrus and neuron density of the granule cell layer do not differ between wild type and ApoE KO mice. In the region CA1, shaft synapse density in the ApoE KO group is 40 and 50% greater on the basal and apical dendrites respectively compared to the wild-type group. Spine density in the ApoE KO mice is 5% smaller on the basal dendrites but 15% greater on the apical dendrites compared to wild-type mice. However, in all cases, these differences do not reach a level of statistical significance.

Further studies will be necessary in aged apoE deficient mice to investigate the role of apoE in synaptic plasticity.

Supported by MRC foresight grant G9535597

SEMT Meeting 25 March 1998

List of Registrants

Chris Andrews	School of Pharmacy
Paul Ansell	Hitachi Scientific Instruments
Don Ashcroft	SEM Tech Ltd; Wirksworth, Derbyshire
Alan Boswell	ISS, Withington, Manchester
Tony Brain	EMU, Kings College, London W8
John Bredl	Royal Veterinary College
Judith Brock	Oxford Instruments, Abingdon, Oxon
Jackie Brown	Biol Dept., Open University
Karine Cambon	Biology Dept., Open University, Milton Keynes
Melanie Chaplin	Central Veterinary Laboratories, New Haw, Surrey
Dave Chapman	Leica Microsystems Ltd., Milton Keynes
Bill Clarke	Agar Scientific Ltd, Stanstead, Essex
Allyson Clelland	BVS; Royal Veterinary College
Terry Cooper	Taab Laboratories, Aldermaston, Berks.
John Critchell	Jeol (UK) Ltd., Welwyn Garden City
Heather Davies	EMU, Biol. Sci., Open University, Milton Keynes
Sheila Davis	CAMR, Porton Down
Anne Drewe	Microbiology, Charing Cross & Westminster Med School
Barry Dowsett	CAMR, Porton Down, Wilts.
Peter Flood	Royal Veterinary College
Richard Frost	D & A Technology, Waterlooville, Hants.
Don Fry	Electron Optical Services, Urmston, Manchester
Colin Gagg	Dept of Materials, Open University
Alan Gray	London Hospital Medical College
Gisele Hodges	Queens University, Belfast
Kevin Jennings	SmithKline Beecham Pharmaceuticals, Harlow, Essex
Lynne Joyce	Agar Scientific Ltd, Stanstead, Essex
Claire Kendal	Biology Dept., Open University, Milton Keynes
Nicola Kessel	Coates Lorilleux, Orpington, Kent
Gill Lewis	EMU, Eastman Dental Inst
Trish Lovell	Histopathology, Royal Marsden Hospital, SW3
David McCarthy	EMU, School of Pharmacy
Kevin Meade	Leo EM, Cambridge
David Michell	Edge Scientific Instrument Co., Milton Keynes
Nicky Mordan	EMU, Eastman Dental Hospital
David Robinson	Emitech Ltd., Ashford, Kent
Phil Salmon	Royal Veterinary College, London
Padmini Sarathchandra	EM, Surgical Research, NPIMR, Harrow
Lee Scott	Leica Microsystems Ltd., Milton Keynes
Arthur Smith	Bemax Ltd, Milton Keynes
Tony Tamo	Eastman Dental Hospital
Julie Tierney	Coates Lorilleux, Orpington, Kent
Wendy Tynan	Cortecs Research Lab., School of Pharmacy
Berina Waights	Biol Dept, Open University
Chris Walker	Philips Analytical, Cambridge
Bob Whitenstall	Dept of Materials, Queen Mary & Westfield College
Neil Wilkinson	Gatan Ltd, Corby, Northants.
Naomi Williams	Dept of Material Engineering, Open University
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