ELECTRON MICROSCOPE UNIT

ANNUAL REPORT

2009

Permanent Staff

Director	**B.T. Sewell**
Principal Technical Officer (Part Time)	**J. Duncan**
Chief Technical Officer	**M. A. Jaffer**
Chief Scientific Officer	**B.W. Weber**
Chief Technical Officer	**M.E. Waldron**
Technical Assistant	**S. Karriem**

HIGHLIGHTS OF 2009

TECNAI T20 TEM

An application to the National Equipment Programme for a state-of-the-art system was successful. It is expected that this will be delivered in January 2010, and the room wher it was to be installed was redecorated, had new air conditioning installed, had new floor tiles installed and had acoustic tiles and curtails installed

APPLICATION FOR FEGSEM

The Wolfson Foundation, a British science funding charity, agreed to accept an application from UCT for a new FEGSEM, after an approach from the Vice-Chancellor. The application was submitted on 1 March 2009 and the outcome was the award of GBP200k. SASOL also agreed to donate R1.5m towards a new FEGSEM. In January Prof Sewell visited the Zeiss factory in order to evaluate their range of SEMs, the same factory was visited by Prof Claeys in April. Profs Knutsen and Claeys also visited Jeol UK for a demo on their machines in July 2009. The Zeiss machine was eliminated due to the unacceptable imaging performance. In September, Prof Sewell visited FEI in Eindhoven for an extended demo on their Nova NanoSEM and in October, final quotes from FEI and Jeol were submitted. Although both instruments had similar configurations and were similar prices, a committee decided that the FEI instrument would suit our needs better and the quote was accepted. The instrument was expected to be delivered in February 2010, the room it was to be installed in was redecorated which involved new floor tiles, new curtains the removal of an old bench and installation of a prep table..

Jeol 1200EX

In order to make room for the new Tecnai T20 TEM, the Jeol 1200EX TEM had to be moved. After making enquiries in local universities and research institutions, a home was found for the TEM at the University of Stellenbosch and it was transported to its new home in September 2009

S200 dismantled

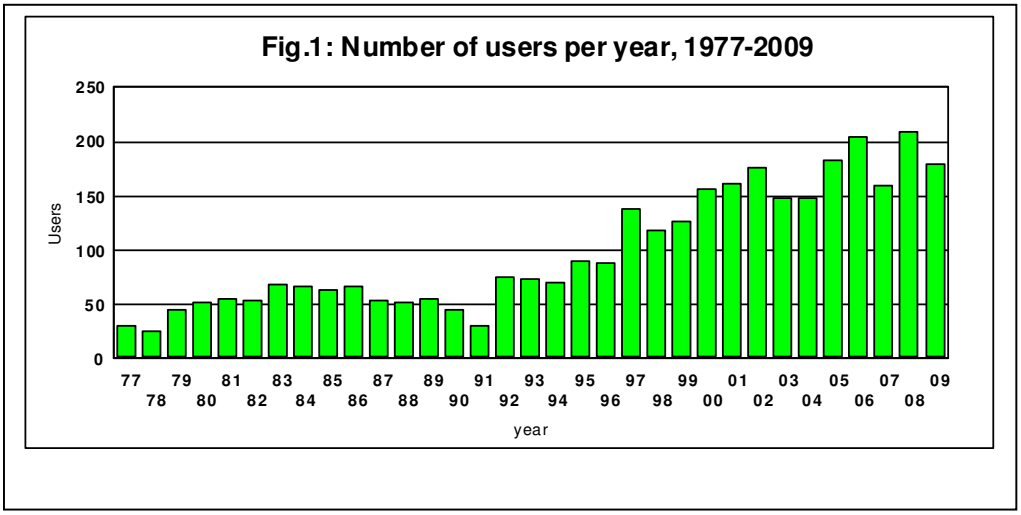
The Cambridge S200 SEM also had to be dismantled and moved in order to make way for the new SEM. The Department of Physics, University of Stellenbosch, acquired the columns from the S200 and the spare SEM we used for parts. The rest of the machines were sent for scrap. Both the SEM and the TEM rooms underwent site surveys by FEI to make sure the rooms were suitable for the new instruments

UPS - continuous power

After major power disruptions in the past couple of years, the R.W.James building had a generator installed and all the major instruments were put on to a UPS system, thus ensuring continuous power during power failures. The system was operational by July 2009.

USE OF THE UNIT

Services provided by the Unit during 2009 are listed in Table 1. Frequent usage was made of all key services of the Unit. 179 people made use of the microscopy services of the Electron Microscope Unit in 2009. This is a slight decrease from 2008, 22 further users utilized services other than those related to microscopy, notably critical point drying and liquid nitrogen collection. The names and departments of the users are listed in Table 2.



Total time spent using the Unit's microscopes was 1327 hours in 2009, similar to the microscope hours in 2008 (Fig 2.). The S200 SEM and the 1200CX TEM were both decommissioned in 2009 and the S440 SEM and the 912 TEM were the most used instruments.

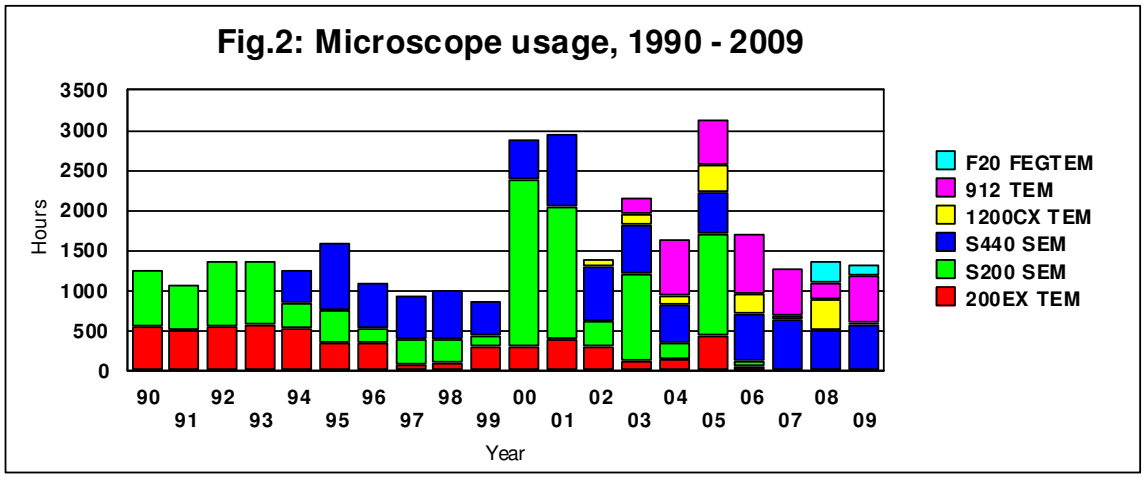
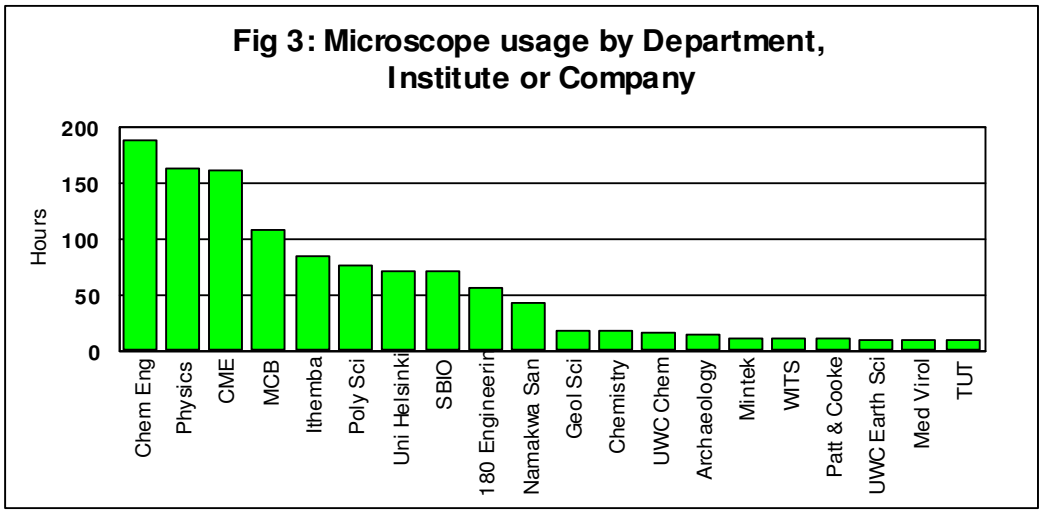


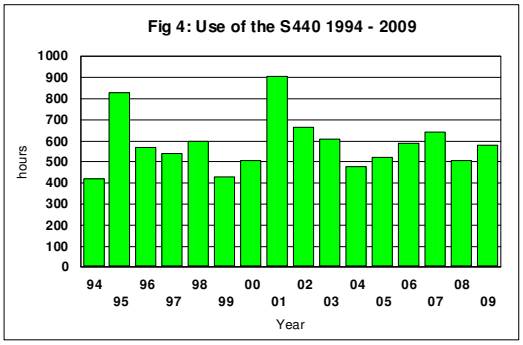
Fig 3 shows the top 20 departments, institutes and companies who used the electron microscopes during 2009. Chemical Engineering used the instruments for 189 hours, the next department being Physics with 163.5 hours, through to Tswane University of Technology, using the instruments for 10 hours. The hours used by Chemical Engineering were notched up by several

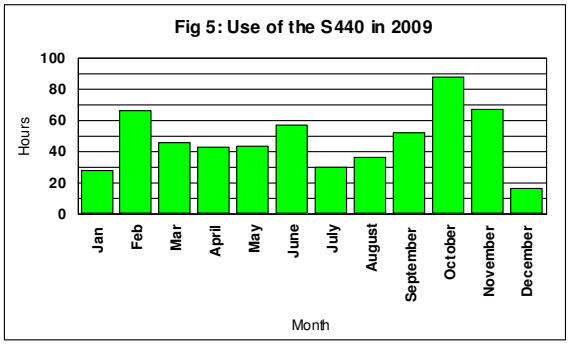


different students, whereas although Physics and CME had several users, the majority of hours notched up by these departments were from one main user in each department.

ELECTRON MICROSCOPES AND ASSOCIATED EQUIPMENT

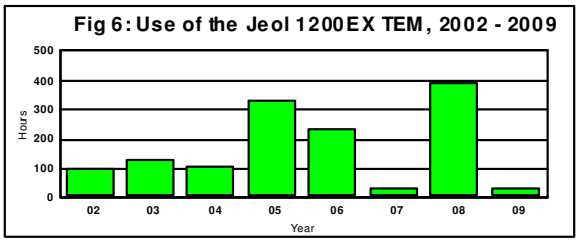
LEO STEREOSCAN S440 SEM





The S440 was used for a total of 577 hours which is an increase on the usage in 2008. Fig. 4 shows the hours the instrument was used since its installation in 1994. The highest years of use were 1995 (the first full year of operation) and 2001 (which followed a period of intense marketing and increased user support). 57 people from UCT made use of the instrument in 2009 and there were 66 outside users, the first time more people from outside UCT have used the instrument. In February the Unit experienced power spikes which resulted in the power tripping 3 or 4 times a day, this affected the S440 and the Leo912. In order to be able to continue to operate the S440, the tungsten filament was used until the problem was sorted out, which turned out to be caused by a faulty air conditioning unit. The microscope was down for 1 day whilst the UPS was being installed and for another 2 days with vacuum pumping problems. The S440 is showing signs of age / obsolescence but UCT have had 15 productive years of use of this instrument. It was the first modern, fully digital SEM in South Africa and has proved to be an extraordinarily robust, reliable and popular design.

JEOL 1200EXII TEM



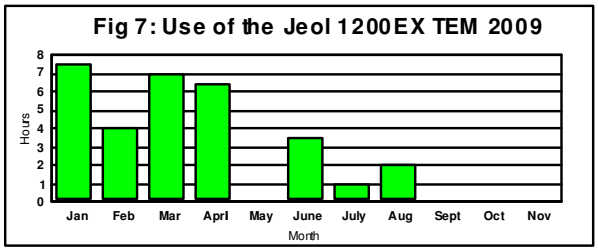
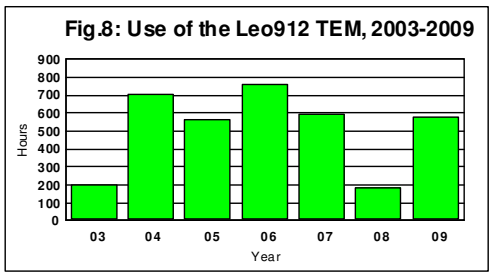
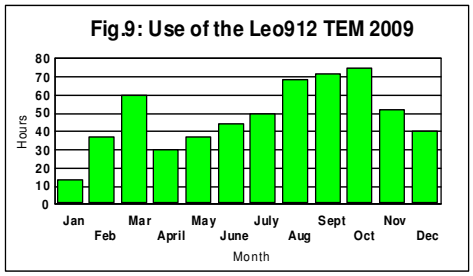


Fig. 6 shows the use of the Jeol 1200EXII since 2002, the instrument was decommissioned in September 2009, but was still used for a total of 31.5 hours, (Fig.7).Only 6 people from UCT and 1 from other universities or institutes used the instrument in 2009.

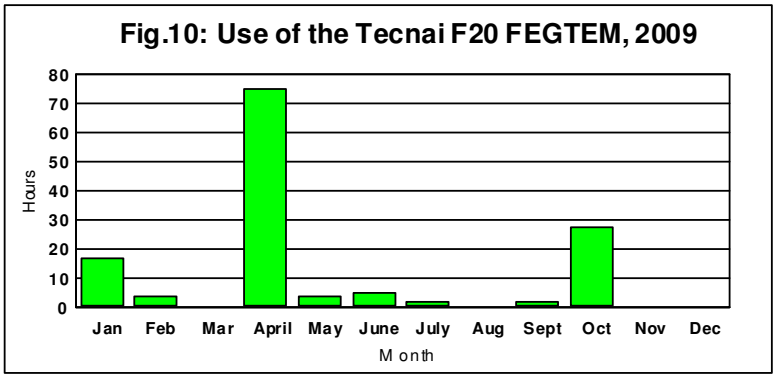
LEO 912 TEM





In 2008, the Leo912 TEM was affected by power cuts and was down for nearly 6 months. Apart from the power problems mentioned above, things were back to normal in 2009 (figs. 8 & 9) and the 912 was used for 581 hours. 36 people from UCT and 31 from other universities or institutes used the instrument.

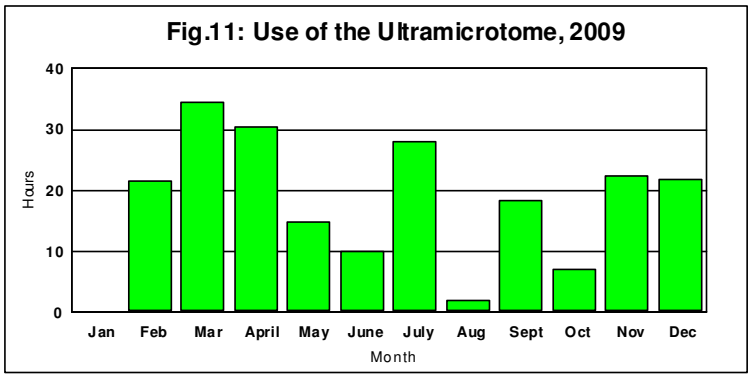
TECNAI F20 FEGTEM



This microscope used for a total of 137.5 hours, by 5 users from UCT and 3 from other universities. The users from UCT were predominantly from the Structural Biology group, 2 of the

outside users were from the University of Helsinki and used the instrument extensively in April in a collaborative study with UNW and Onderstepoort, which will result in a better understanding of African Horse Sickness Virus.

ULTRAMICROTOME



Use of the ultramicrotome in 2009 was 211 hours, a slight decrease from last year. Cryomicrotome facilities were used by:

LIGHT MICROSCOPY

All the light microscopes and the Zeiss Axiocam continued to be used throughout the year, by a variety of students, a few of whom used them in conjunction with SEM projects.

IMAGING CENTRE

The imaging centre has sophisticated software capability aimed at image enhancement and three dimensional reconstruction. Photographic negative digitization using the Nikon LS4500 and Leafscan scanners is the basis of data analysis.

TEACHING AND EXTENSION

INDIVIDUAL TRAINING

Leo 912 TEM

Jeol 1200EX TEM

Tecnai F20 TEM

Leo S440 SEM

L. Jansen, Marine and Coastal Management
S. Maduray, Marine and Coastal Management

T. Samaai, Marine and Coastal Management
S. Petersen, Centre for Materials Engineering
M. Kotze, Zoology
W. Archer, Archaeology
S. Jones, Physics
B. Sone, Ithemba labs
M van Niekerk, Geological Sciences
K. Singh, Botany
L. Monfal, Physics,
D. Kiewets, Namakwa Sands

Ultramicrotome

SCHOOL VISITS

There were fewer school visits than usual in 2009: Bishops High School visited the unit in February and a student from St Josephs College did a work shadow at the Unit in May.

MICROSCOPY FOR BIOLOGISTS

The Microscopy for Biologists course was held in February and attended by 19 MCB honours students.

RESEARCH ACTIVITY

SA STRUCTURAL BIOLOGY INITIATIVE

The director applied to the DST for a grant to establish a process which was intended to lead to the creation of a National Programme in Structural Biology. The grant which was administered by the Cape Biotech Trust provided for a series of meetings to establish a consensus on how such a programe would operate. After three meetings which were attended by all University and CSIR personnel that declared an interest in the discipline, a proposal calling for the establishment of a National Centre for Structural Biology was presented to Dr Boni Mehlomekhulu. The proposal was rejected and the committee was called upon to propose an alternative structure. The constraints were that no new equipment would be available to the programme and a grant administered by the NRF should be applied for by researchers wishing to do structural biology.

After a further meeting the committee proposed that the programme comprise the following key components:

(1) Essential equipment for the programme comprises a cryo_FEGTEM (UCT), X-ray diffractometers (UWC and Wits), NMR spectrometers (Stellenbosch, Rhodes and UFS) and computers (CHPC, CSIR and UP)

(2) Five technical staff would be funded in order to provide access to these resources for those who were funded within the programme

(3) The NRF would call for proposals totaling R6m per year to fund projects the purpose of which was to gain insight into biological systems through a study of their structure.

(4) The masters programme at the University of the Western Cape would be supported through student grants, resources for student projects and infrastructure to support the programme.

It is anticipated that the above structure will be approved in 2009 and that it will be implemented in 2010. Funds have been allocated to maintain the Masters programme at UWC during 2010 in anticipation of the broader programme.

SOUTH AFRICAN SYNCHROTRON INITIATIVE

The objectives of the synchrotron initiative are to secure ready access to synchrotron beamlines for structural work arising from all disciplines ranging from Paleontology on the macroscopic scale to chemistry on the atomic scale. There are approximately fifty synchrotrons located world-wide. An early decision was taken not to push for a synchrotron in South Africa at this time – but rather to create a roadmap that would progressively empower South African researchers. Initially researchers would use existing resources, then South African researchers would build and maintain one or more beamlines at a synchrotron and ultimately, if justified by circumstances build a synchrotron in South Africa.

The initiative has involved regular meetings between the key players: Professor Simon Connell, Professor Giovanni Hearne and the Director of the EMU with people at the DST, including the Director General. Funds for a second Science at Synchrotrons meeting were obtained from the DST and a delegation from the French synchrotron (Soleil) and the European Synchrotron Radiation Facility (ESRF) were invited to attend. The director and post-docs Jennifer van Wyk and Jean Watermeyer, PhD students: Joni Frederick and Serah Kimani and MSc student, Simon Broadley, presented at the conference which was held in January 2009.

The last session of the meeting entailed the presentation of a memorandum of understanding enabling South Africans to use Soleil under the same conditions as French scientists. Essentially the French would fund the use of the Synchrotron for refereed projects and the South Africans would pay to get to Soleil. The scheme would be administered by the NRF. Subsequent to the meeting the MoU was signed by Dr Albert van Jaarsveld and Dr Michel van der Rest.

BIG STRUCTURES WORKSHOP SERIES

Funds were obtained from the NRF Knowledge Interchange Programme to promote Structural Biology, especially cryo-electron microscopy and more particularly to hold workshops that will enable PhD students and from all South Africa to learn from established world experts.

Date	**Presenter**	**Topic**	**Location**
30th August – 10th September 2009	Dr Michael Lawrence	Advanced Topics in the structures of receptors	Cape Town
8th – 20th December 2009	Professor Pawel Penczek	Advanced Topics in EM reconstruction and computing	Cape Town

Project	Collaborators	Students	Technique	Status	Database ID's	Publication
CynD from *Pseudomonas stutzeri*	Michael Benedik (A&M), Paul Meyers (UCT)	Mark Berman	EM / modeling	published	1050	Sewell, B.T., Berman, M., Meyers, P.R.,Jandhyala, D., and Benedik, M.J. (2003). The Cyanide Degrading Nitrilase from *Pseudomonas stutzeri* AK61 Is a Two-Fold Symmetric, 14-Subunit Spiral, Structure, **11**, 1413-1422. Citations 13. Impact 5.7
CynD from *Bacillus pumilus*	Michael Benedik (A&M), Paul Meyers (UCT)	Mark Berman Margot Scheffer Leandri van der Vyver Alistair Gray	EM / modeling	Published, work continues		Jandhyala, D., Berman, M., Meyers, P.R., Sewell, B.T., Willson, R.T.,and Benedik, M.J. (2003). CynD, the Cyanide Dihydratase from *Bacillus pumilus:* gene cloning and structural studies. Appl. Environ. Microbiol. **69**, 4794-4805. Citations 11. Impact 3.5 Jandhyala,DM, Willson,RC, Sewell,BT and Benedik,MJ.(2005). Analysis of Three Microbial Cyanide Degrading Enzymes. Applied Microbiology and Biotechnology. **68**, 327-335. Citations 3, Impact 2.4 Sewell, BT, Thuku RN, Zhang X, Benedik M. (2005) The oligomeric structure of nitrilases: the effect of mutating interfacial residues on activity. Annals of the New York Academy of Sciences, **1056**, 153–159. Citations 4. Impact 1.13
Cynanide Hydratase from *Gloecercospora sorghi*	Michael Benedik (A&M) Jean Watermeyer (UCT) Andreas Hoenger (Colorado)	Paul Chang Margot Scheffer Jeremy Woodward	EM /modeling	Published, work continues		Woodward, J.D., Weber, B.W., Scheffer, M.P., Benedik, M.J., Hoenger, A., Sewell, B.T.(2008) Helical Structure of Unidirectionally Shadowed Fibres of Cyanide Hydratase from *Gloeocercospora sorghi.* Journal of Structural Biology, **161**, 111-119. Citations 1. Impact 3.5
Cyanide Hydratase from *Neurospora crassa*	Michael Benedik (A&M) Jean Watermeyer (UCT)	Kyle Dent, Lacy Basile	EM / modeling	Two papers published Work continues		Basile, L.J., Willson, R.C. Sewell, B.T., Benedik, M.J. (2008) Genome mining of cyanide degrading nitrilases from filamentous fungi, Applied Microbiology and Biotechnology Dent, K.C., Weber, B.W., Benedik, M.J. and Sewell, B.T. (2009) The cyanide hydratase from

						Neurospora crassa forms a helix that has a dimeric repeat, Applied Microbiology and Biotechnology **82**, 271-278
Nitrilase from Rhodoccus rhodochrous J1	Dean Brady (CSIR)	Robert Ndoria Thuku	EM / modeling	Published	1313	Thuku, RN, Weber, BW, Varsani, A and Sewell, BT (2007) Post-translational cleavage of recombinantly expressed nitrilase from *Rhodococcus rhodochrous* J1 yields a stable, active helical form. FEBS Journal, **274,** 2099-2108. Citations 4. Impact 3.0 Thuku, R.N., Brady, D., Benedik, M.J., Sewell, B.T. (2009) Microbial nitrilases: versatile, spiral forming, industrial enzymes, Journal of Applied Microbiology **106**, 703-727
Glutathione S transferase	Heinrich Dirr (Wits)	Diane Kuhnert, Nicole Kinsley	X-ray crystallography, modeling,	Published	1ydk, 2r3x	Kuhnert DC, Sayed Y, Mosebi S, Sayed M, Sewell BT, Dirr HW. (2005) Tertiary interactions stabilise the C-terminal region of human glutathione transferase A1-1: a crystallographic and calorimetric study. J Mol Biol. **349**, 825-38. Impact 4.9
Glutamine synthetase type III from *Bacteroides thetaiotamicton*	Jennifer van Wyk (UCT) Val Abratt (UCT)			Cloned, expressed		
Glutamine synthetase type III from *Bacteroides fragilis*	Val Abratt (UCT)	Jason van Rooyen	EM / modeling / x-ray crystallography	Published EM structure of native enzyme. Work on METSOX – GS complex by EM and X-ray crystallography continues.	1205, 1205	Van Rooyen JM, Abratt, VR, Sewell BT (2006) Three-dimensional Structure of a Type III Glutamine Synthetase by Single-particle Reconstruction. Journal of Molecular Biology, **361**; 796-810. Citations 4. Impact 4.9
Angiotensin converting enzyme from human testis	Ed Sturrock (UCT)	Jean Watermeyer, Itai Chitapi	X-ray crystallography	2 xMSc, 1x PhD, 3 x papers. Mutant solved	2iul, 2iux, 2oc2, 3bkk,3bkl	Watermeyer, J., Sewell, BT, Schwager, SL, Natesh, R, Corradi, HR, Acharya, KR and Edward D. Sturrock, ED (2006) Structure of

				at 2.0A resolution. Co-crystallised with potential drugs		testis ACE glycosylation mutants and evidence for conserved domain movement. Biochemistry, **45**; 12654-12663. Citations 5. Impact 3.6 Corradi, H., Chitapi, I., Sewell, B.T, Georgiadis, D., Dive, V., Sturrock, E, Acharya, K. R. (2007) The Structure of Testis ACE in Complex with the C Domain-Specific Inhibitor RXPA380, Biochemistry, **46**, 5473-5478. Citations 2. Impact 3.6 Watermeyer JM, Kröger WL, O'Neill HG, Sewell BT, Sturrock ED. (2008) Probing the basis of domain-dependent inhibition using novel ketone inhibitors of Angiotensin-converting enzyme. Biochemistry, **47**, 5942-50.
Nitrilase from Rhodococcus rhodochrous ATCC BAA870	Dean Brady (CSIR)	Joni Frederick	EM	Cloned, expressed, characterized by EM, apparently identical to J1 nitrilase		
Nitrile hydratase from Rhodococcus rhodochrous ATCC BAA870	Dean Brady (CSIR)	Joni Frederick	x-ray crystallography	Cloned		
Nitrile Hydratase from *Geobacillus pallidus* - thermostability	Donald Cowan (UWC) Muhammed Sayed (UWC)	Jennifer van Wyk	x-ray crystallography	Five structures of different mutants solved at 1.2 – 1.5 A resolution		
Nitrile Hydratase from *Geobacillus pallidus* - structure	Donald Cowan (UWC) Muhammed Sayed (UWC) Ozlem Tastan Bishop (UWC)	Tsepo Tsekoa	x-ray crystallography	Four structures of different mutants solved at 2.5-2.8A, 2 papers, 1 PhD	2dpp	Tsekoa, T.L., Sayed, M.F., Cameron, R.A., Sewell, B.T. and Cowan D.A. (2004) Purification, crystallization and preliminary X-ray diffraction analysis of thermostable nitrile hydratase. South African Journal of Science, **100**, 488-491. Tastan Bishop AO, Sewell T (2006) A new approach to possible substrate binding mechanisms for nitrile hydratase. Biochem Biophys Res Commun. **343**,319-25.

Nitrile Hydratase from *Geobacillus pallidus* - mechanism	Donald Cowan (UWC) Jennifer van Wyk (UCT)	Tsepo Tsekoa Parikshant Kowellesur Samuel Kwofie Ndlelenhle Khanyile Clive Mketsu William Mavengere	x-ray crystallography	2 MSc's, Relevant mutants made, one crystal structure at 2.5A. Illustration of rational enzyme design.		
Nitrilase from*Geobacillus pallidus*	Donald Cowan (UWC) Rory Cameron (UWC)	Kyle Dent Johann van Heerden Dael Williamson Joni Frederick	Electron microscopy, biochemistry, bioinformatics	Pre-structural paper in final stages prior to submission		
Amidase from *Geobacillus pallidus*	Donald Cowan (UWC) Muhammed Sayed (UWC) Stephanie Burton (CPUT) Arvind Varsani (UCT)	Serah Kimani, Steven Makhongela	X-ray crystallography, biochemisry	Structure complete at 1.8 A, 2x MSc, 3 papers	2plq	Agarkar VB , Kimani, SW, Cowan, DA, Sayed, MF-R., Sewell, BT (2006) The quaternary structure of the amidase from *Geobacillus pallidus* RAPc8 is revealed by its crystal packing, Acta Cryst. F62 , 1174-1178. Citations 5. Impact 0.6 Makhongela HS, Glowacka, A., Agarkar VB, Sewell, BT, Weber, B, Cameron RA., Cowan DA, and Burton SG (2007) Molecular characterization and immobilization of D-specific amidase from *Geobacillus pallidus*. Applied Microbiology and Biotechnology, **75**, 801-11. Citations 1. Impact 2.4 Kimani, SW, Agarkar,VB Cowan, DA, Sayed, MF-R and Sewell, BT (2007) Structure of an aliphatic amidase from *Geobacillus pallidus* RAPc8 Acta Cryst. **D63**, 1048–1058. Citations 1. Impact 1.7
Amidase from *Geobacillus*	Brandon Weber (UCT)	Serah Kimani	X-ray crystallography,	Structure of mutant solved at		

pallidus E142L			Mass spectroscopy	1.5A resolution. Paper in process		
Amidase from *Nesterenkonia* sp.	Donald Cowan	Andrew Nel	X-ray crystallography	Structure solved and deposited in database. Paper in progress	3hkx	
HcRNAV	Arvind Varsani (UCT)	Jennifer Miller	EM	MSc, preparing for publication, handedness currently unknown		
Maize streak virus	Arvind Varsani (UCT)	Kyle Dent	EM / modeling	Structure of two forms solved		
African Horse Sickness virus	Albie van Dijk (UNW) Pasi Laurinmaki (University of Helsinki)	Violetta Manole	EM	Reconstruction complete		
Old yellow Enzyme from *Thermus scotoductus*	Esta van Heerden (UFS)	Dirk Opperman	X-ray crystallography	Two different crystal forms collected and solved. Model building in process.		Paper submitted to Journal of Molecular Biology.
Thioredoxin reductase from *Thermus scotoductus*	Esta van Heerden (UFS)	Armand Bester	X-ray crystallography	Protein prepared, crystal trials successful		
Histone octamer tubular crystals	Hugh Patterton (UFS)	Timothy Frouws	EM / modeling	MSc, published, Project terminated	1469, 3c9k	Frouws, T.D., Patterton, H.-G., Sewell, B.T. (2009) Histone Octamer Helical Tubes Suggest an Internucleosomal 4-Helix Bundle Stabilizes the Chromatin Fiber, Biophysical Journal, **96**, 3363-3371
NAD+ synthetase from *Mycobacterium tuberculosis*	James Sacchettini (A&M)	Serah Kimani	EM / modeling / x-ray crystallography	Project discontinued for the present as planned work was published by researchers from the University of		

				Maryland.		
GroEL from E. coli E461K	Helen Saibil (Birkbeck)	Robert Best	EM / modeling	Completed, published	1095	Sewell,BT, Best,RB, Chen,S, Roseman,AM, Farr,GW, Horwich,AL & Saibil,HR (2004). A mutant chaperonin with rearranged inter-ring electrostatic contacts, defective cooperativity, and temperature-sensitive dissociation of rings, Nature Structural and Molecular Biology, **11**,1128-1133. Citations 14. Impact 11.5
Malarial glutamine synthetase	Amit Sharma (ICGEB) Jennifer van Wyk (UCT) Sandra Meredith (UCT)			WT and synthetic genes cloned. Expression		
Malarial NAD+ syntetase	Amit Sharma (ICGEB) Jennifer van Wyk (UCT) Sandra Meredith (UCT)			WT and synthetic genes cloned. No expression		
Malarial hypoxanthine ribosyl transferase	David McIntosh,	Simon Broadley	X-ray crystallography	Protein prepared / novel drug analogue identified / crystallization trials in process		
HIV gp120 & aptamer	Shooz Khati (CSIR) Marisa Baron (CSIR)	Elinor Cave	EM / modelling	Microscopy in process. Paper at an advanced stage.		
Mtb Mycothiol reductase and MshB	Daan Steenkamp (UCT) David Gammon (UCT) Brandon Weber (UCT) Anwar Jardine (UCT)	Dael Williamson	X-ray crystallography	Protein expressed and purified. In crystallization trials		
Cytochrome P450 with unusual activity	Martie Smit (UFS)		X-ray crystallography	Grant refused, licking wounds!		
Plant nitrilases (4)	Markus Pitrowski (Bochum)	Jeremy Woodward	EM	All four proteins cloned, expressed and purified. Reconstructions of three		

				complete. One in crystallization trials.		
Three dimensional reconstruction of metal coated surfaces	Roger Wepf (ETH, Zurich)	Jeremy Woodward	EM	Paper published, work continues		Woodward, J.D., Wepf, R., Sewell, B.T. (2009) Three-Dimensional Reconstruction of Biological Macromolecular Complexes from Scanning Electron Micrographs, Journal of Microscopy (in press)

2009 Grants

Grant Name	Source	Amount
Solving Structures for Drug Discovery*	NRF/UCT	256,667
Key International Science Capacity Award*	NRF	70,000
CCD Camera package	NRF	2,500,000
KIC Travel grant	NRF	15,000
Tecnai T20 TEM	DOE	6,800,000
Nova NanoSEM	SASOL	1,550,000
Nova NanoSEM	Wolfson Foundation	£200,000
ICGEB Collaborative programme	ICGEB	€15,000
Science Faculty Block Grant	UCT	13,664

*Break down of IRDP Money:
Post Doc 90,000
Lab Expenses 166,667

*KISC award is R70,000 per year for 3 years, this just reflects the amount awarded in 2009

PUBLICATIONS

Publications for 2009 that resulted from research in which the EM Unit staff have been directly involved are listed:-

Dent, K.C., Weber, B.W., Benedik, M.J. and Sewell, B.T. (2009) The cyanide hydratase from *Neurospora crassa* forms a helix that has a dimeric repeat, Applied Microbiology and Biotechnology **82**, 271-278

Frouws, T.D., Patterton, H.-G., Sewell, B.T. (2009) Histone Octamer Helical Tubes Suggest an Internucleosomal 4-Helix Bundle Stabilizes the Chromatin Fiber, Biophysical Journal 96(8), 3363-3371

Kinfe H. H., Chhiba, V.. Frederick J, Bode M. L., Mathiba M., Steenkamp P. A. and Brady D.. (2009) Enantioselective hydrolysis of □-hydroxy nitriles using the whole cell biocatalyst *Rhodococcus rhodochrous* ATCC BAA-870. J. Mol. Catal. B: Enzymatic. 59, 231-236.

Thuku, R.N., Brady, D., Benedik, M and Sewell, B.T. (2009). Microbial nitrilases: versatile, spiral forming, industrial enzymes. Journal of Applied Microbiology. **106** 703-727

Varsani, A. Shepherd, D., Deny, K.C., Monjane, A.L. Rybicki, E and Martin, D.P. (2009). A highly divergent South African geminivirus species illuminates the ancient evolutionary history of this family. Virology Journal. **6:36** 1-12

Watermeyer, J., Kroger, W.L and Sturrock, E.D. (2009). Angiotensin-Converting Enzyme - New Insights into Structure, Biological Significance and Prospects for Domain Selective Inhibitors. Current Enzyme Inhibition. **5**. 134-147

PUBLICATIONS BY USERS OF THE UNIT

The following list includes those papers given to the Unit by users. It is unfortunately not a complete list of published work that has been conducted in the Unit. A great deal of the work done by users is published only as conference proceedings, such work is not reflected here.

Bathori, N and Bourne, S (2009) Crystal Structure of D(−)-amino-(4-hydroxyphenyl)acetate, the Zwitter Ionic Form of Biologically Active D(−)-4-hydroxyphenylglycine. Journal of Chemical Crystallography, **39** 539-543

Britton, D, Ayodele, O.E, Girma, G.G., Ohieku, J.E. and Harting, M (2009). Size distribution and surface characteristics of silicon nanoparticles. Journal of Applied Crystallography **42** 448-456

Farrant, J., Arnaud, L. and Cooper, K. (2009). Desiccation tolerance in the vegetative tissues of the fern *Mohria caffrorum* is seasonally regulated. Plant Journal **57** 65-79

Gwagwa, X.Y. and van Steen, E (2009). Migration of potassium in an Fe₂O₃/H-ZSM-5 composite catalyst. Chemical Engineering and Technology. **32** 826-829

Hedderson, N, Balsamo, R, Cooper, K & Farrant, J (2009) Leaf tensile properties of resurrection plants differ among species in their response to drying. South African Journal of Botany, 75. 8-15

Hockman, D, Picker, M., Klass, K & Pretorius, L (2009). Postembryonic development of the unique antenna of Mantophasmatodea (Insecta). Arthropod Structure & Development 38(2) 125-133

Hove, M., van Hille, R.P and Lewis, A. (2009). The effect of different types of seeds on the oxidation and precipatation of iron. Hydrometallurgy **97** 180-184

Scriba, M., Britton, D., Arendse, A., van Stasen, M and Harting, M (2009) Composition and crystallinity of silicon nanoparticles synthesised by hot wire thermal catalytic pyrolysis at different temperatures. Thin solid films, **517** 3484-3487

Topic, M., Pineda-Vargas, C., Bucher, R., du Plessis, H., Breedt, B., Pischedda, V and Silethelwe, N (2009). High temperature study on thin aluminium coatings deposited onto thick platinum substrates. Surface and coatings Technology. **203** 3044-3048

PHD THESES

Al-Farsi, A, Botany. Molecular, morphological and biogeographic studies of the stapeliads with emphasis on the genus Caralluma s.l. (Apocynacae-Asclepiadoideae)

Matahwa, H. Chemical Engineering: Chemical modification of polysaccharides with hydrophilic polymers for $CaCO_3$ crystal growth modification and filler retention, for paper applications.

Odo, A, Physics. Synthesis, characterisation and device application of silicon nanoparticles produced by mechanical attrition

Samakande, A. Chemical Engineering: Use of the RAFT technique as an efficient method to synthesise well-defined polymer-clay nanocomposites with improved properties.

Theka, T, Chemistry: Studies of genetic, gastro-intestinal, renal and dietry factirs in white and black South African subjects as a possible key to understanding the relative absence of calcium oxalate kidney stones disease in the black population

van den Dungen, E. Polymer Science: Self-healing coatings based on thio-ene chemistry

van der Merwe, E. Human and Cell Biology: A structural and developmental study of the postrabecular aqueous outflow pathway of the mouse eye.

Zhuang, Y. Chemical Engineering. The performance of structured cobalt catalysts in Fischer-Tropsch synthesis

MSc Theses

Chirowodza, H. Synthesis and characterization of cationically and anionically modified poly(vinyl alcohol) microfibrils.

Chihoro, L. Effects of oxygen diffusion hardening on fatigue resistance in T-i-6Al-4V alloy
Solana, O, Production and characterisation of nanoparticulate silicon photovoltaic devices

Zengeni, E. Poly(acrylonitrile/methyl acrylate) copolymers and clay nanocomposites: Structural and property relationships.

User Projects in 2009

Students	PI	Project	Technique
Archaeology			
Archer, W	Dr Braun	Functional and raw-material economy of bifacial technology at Elandsfontein, Western Cape.	SEM
Galimberti, M	Prof Sealy, Prof Lee-Thorp	Oxygen isotope analysis of Middle Stone Age shellfish.	SEM
Hinkman, A	Prof Sealy	Investigating the use of bone density fractionation as a morphological tool in stable light isotope analysis	SEM
Botany			
Singh, K	Dr Klak	Revision of *Oscularia* (Aizoaceae)	SEM
Chemical Engineering			
Amod, M	Prof. van Steen	Potassium promotion on nano-sized iron crystallites in the Fischer-tropsch process	TEM
Case, J Julies, M Van Heerden, T	Prof. van Steen	An Investigation of ion exchange as a method for preparation of gold catalysts for use in ethylene glycol oxidation.	TEM
Chonco, Z	Prof. van Steen	Promotion of Fe-based FT-catalysts by group II metals.	TEM
Fisher, N	Prof M Claeys	Nanometre and Angstrom sized cobalt ensembles and their performance in the Fischer-Tropsch synthesis.	TEM
Gertenbach, R	Dr Van Berg,	SASOL catalyst, R2.2	TEM
McFadzeen, B	Dr Parolis,	Floatation and comminution of industrial minerals.	SEM
Na, K Mabala, P Paradza, N	Prof. A Lewis	Control, of particle characteristics in precipitation process	SEM TEM
Mungwe, N	Prof. M Claeys	Fischer-Tropsch Synthesis based on Rhodium crystallites and clusters of different sizes.	TEM
Mangere, M	Prof. A Lewis	Continuous precipitation using the replication model system.	TEM
Reddy, T	Prof. A Lewis	Eutectic freeze crystallisation	SEM
Rendani, R	Prof. A Lewis	Phosphate removal from waste water.	SEM
Stoddart, F	Prof. van Steen	Low pressure cobalt catalyzed ammonia oxidation	TEM
Tsoenyane, M	Prof van Steen	The inveastiagtion of a novel cobalt catalyst preparation of cobalt based Fischer-Tropsch synthesis	TEM

Wright, C	Prof Fletcher	Synthesis of medium pore zeolites for the alkylation of M-cresol with isopropaniol.	SEM
Chemistry			
Banothile, A	Dr Hutton, Dr Smith,	Synthesis of polymer supported transition metal complexes as potential catalysts in anti-cancer agents	SEM
Botha, S		Preparation of mono-, bi- or tri-metallic nanocatalysts for CO oxidation	TEM
Teleke, V	Prof A Rodgers	Investigation of the effect of dietary oxalate dosages in urinary risk factor for calcium oxalate kidney stone in black and white subjects.	SEM
CME			
Finkelstein, L Miller, D	Prof C Lang	Novel ordering of platinum alloys	SEM
George, S	Prof Knutsen		SEM
Leteba, G	Prof C Lang	Catalytic properties of pt-based near surface alloys	TEM
Mias, K	Prof. C Lang	Super-black Platinum.	SEM
Petersen, S	Prof. C Lang	Investigation of the hot deformation if sintered titanuim compacts produced from direct reduction powder.	SEM
Geological Sciences			
Dreyer, T	Diener, J	Investigation of artifacts from Naukluft thrust	SEM
Viglietti, P Faber, C	Dr Rowe	Deformation textures in ther Naukluft thrust – dolocalaclasites to calcmylonites	SEM
Molecular and Cell Biology			
Beckett, M	Prof Farrant	The role of abscicic acid in stomalta regulation in Xerophytea Humilis	SEM, TEM
Botha S	Dr V Coyne	The investigation of pathogenesis-related proteins produced by *G. gracilis* in response to infection .	SEM
K Cooper, M Burger, Pineo, C	Prof Farrant	Biochemical, genetic, physiological and cellular research in dessication plants.	SEM, TEM
Rholand, J	Dr P Meyers	Identification of novel antibiotics.	SEM
Mbewana, S Mortimer, E	Prof E Rybicki	Vaccine development against avian influenza.	TEM
Mechanical Engineering			
Lakhi. F	Prof Ramesh	Development of a pulsed power supply for micro EDM	SEM
Thorpe, K Carulei, O	Prof Kuppuswamy	A new innovative drill proposed to minimize burr when drilling cross holes through aluminum	SEM
Physics			
Mongalo, L Nsengiyumva, S	Prof Britton	Investigation of copper metal matrix nano composite	SEM
Nzikou, M	Prof D. Britton, Prof M. Harting	Plasma-sprayed ceramic coatings.	SEM, LM
Unigibe, D Ramanandraitsiory, I	Prof D. Britton, Prof M. Harting	Application of Ni-Si semiconductor material.	SEM, TEM, LM
Structural Biology			
Broadley, S	Dr B.T.Sewell	Purification and co-crystallisation of Plasmodium falciparum HGXPRT with a chalcone inhibitor	TEM
Frederick, J	Dr B.T.Sewell	Structural elucidation and characterization of nitrile-metabolizing enzymes from *Rhodococcus rhodochrous* ATCC BAA-870.	TEM

Kimani, S	Dr B.T.Sewell	Structural characterization of the active sites of amidases from the nitrilase superfamily; with emphasis on the amidase domain from the Mycobacterium tuberculosis NAD+ synthetase	TEM
Thuku, N	Dr B.T.Sewell	Structure of the nitrilase *Rhodococcus rhodochrous* J1: Homology modelling and three-dimensional reconstruction.	TEM
Van Wyk, J	Dr B.T.Sewell	The basis of thermostability in the Co containing nitrile hydratases.	X-ray
Van Rooyen, J	Dr V Abratt Dr B.T.Sewell	The structure of the type III Glutamine Synthetase from *Bacterioides fragilis* determined by combining electron microscopic and X-ray data.	TEM
Watermeyer, J	Dr B.T.Sewell	Investigating the mechanism of helix formation in microbial nitrilases by three-dimensional negative stain and cryo-electron microscopy	TEM
Williamson, D	Dr B.T.Sewell	Characterisation of the active site pocket with respect to substrate specificity of nitrilase enzymes	TEM
Woodward, J	Dr B.T.Sewell	The feasibility of high resolution, three-dimensional reconstruction of metal-coated surfaces in structural biology.	TEM/SEM
Zoology			
De Ponte M		Pelican distribution by tagging	SEM
Kotze, M	Prof Hoffman	The role of antiexnosis in gauling patterns by *Dasineura Dielsi* on alien *Acacia* species	SEM
Cape Peninsula University of Technology			
Malang, K	Prof Hendry/ C Greyling	Water treatment using non-woven nano alumina filters	SEM
CSIR			
Khati, S, Joubert, M	Scriba, M	Synthesis of silicon nanoparticles.	SEM, TEM
iThemba labs			
Abiona, A	Dr M Maaza	Preparation and characterization of electrospun nanifibers of polymeric materials	
Balla, N, Menovar, D	Dr M Maaza	Modification and charaterization of materials using nanotechnology and thin film physics.	SEM
Khamilch, S Sithole, J	Dr M Maaza	FeSi and FeSi doped with Co for superconductiong properties.	SEM
Khumalo, Z Sone, B	Dr M Maaza	ZnO nano-structures for gas sensing applications	SEM
University of Stellenbosch			
Alkaabi, K	Dr van Reenen	Synthesis and charatcerization of selected binder systems.	TEM
Bailly, N	Dr P Hartman	Synthesis of complex polymer architectures in mini emulsions.	TEM
Bayley, G	Dr R Sanderson	Synthesis and charaterisation of organic-inorganic hybrid block co-polymers.	TEM
Lasper, Q	Prof Aldrich	Sonochemical synthesis of gold nanoparticles in microemulsion	TEM
D'Aguiar, D	Prof Sanderson	Flourescentkly labeled surface hydrolysed polyvinyl pivalate (core shell) particles	TEM

Greesh, N	Dr R Sanderson	Preparation of polymer-clay nanocomposites using emulsion polymerization : influence of clay modifiers on the final nanocomposites morphology.	TEM
Hendricks, K	Dr Mallon	Elexctrospinning of polyisocyamides	TEM
Jobse, P	Prof Lana	Size and stability of PVP stabilized ZnS particles particles	TEM
Kotze, I	Prof Koch	The self and hetero-association of [Pt(phen)CRSO]Ce and fluorine through Ti-cation interaction	TEM
Lakay, E	Prof Koch	Synthesis and characterisation of magnetic nanoparticles.	TEM
Pound, G	Prof Klumperman	Zinc sulphide particles investigation.	TEM
Rose, J	Dr Callanan	Optimizing the process for the hydroxylation of 2-methylmaphthalene to 2-mrthyl-1,4-napthoquinone	SEM
Smit, E Cronje, L	Dr R Sanderson	Investigating PVOH nanofibers as materials for humidity and other sensor applications.	SEM
Sutherland, A	Dr Mallon	The study of free volume of hybrid polymers.	TEM
Zengeni, E	Dr R Sanderson	Ca-clay nanocomposites.	TEM
University of the Western Cape			
Adekola, S	Dr Akinula	Integrated approach to solving reservoir problems using stratigraphy and diagenesis	SEM
Akinyemi, S Olufunke, O	L Petrik, Dr Akinula	Minerology and chemical mobility in flyash from different disposal systems.	SEM
Arotiba, O Hendricks, N	Prof. Iwuoha	Development of electrochemical DNA biosensors.	SEM/TEM
Greyling, C	Dr L Petrik	Nanotechnology for water treatment nanofiltration	SEM
Lukusa, K	Dr Marla	Isolation and genetic characterization of bacteriophages from Antarctic cold desert environments	TEM
Oluwaseun, F	Dr Akinlua	Facies, depositional environment and reservoir properties of the alsian age gas bearing sandstone of Ibhube oilfield, Orange basin, offshore South Africa	SEM
Topley, E	Prof Davison	Honybee black queen cell virus expression in heterologous boculovirus expression system	TEM
Van Onsen, T	Dr Pound-Lana	The study of modified PVP grafted on lime sulphide nanoparticles for potential applications as an in vivo contact agent for medical imaging	TEM
University of Fort Hare			
Greyling, C Gogwana, P	Tichagwa, L	Development of clay/cellulose nanocomposites	SEM

FINANCE

Details of the Unit's accounts are presented in Table 3.

OTHER MATTERS

MSSA 2009

The annual meeting of the Microscopy Society of Southern Africa was held in Kwa Zulu Natal at the University of Kwa Zulu Natal, Durban Campus. The director was responsible for editing the Proceedings as well as inviting Dr Pavel Penczek to give a special plenary lecture. Dr Penczek is from the Department of Biochemistry and Molecular Biology, University of Texas and his trip was sponsored by the NRF. The conference was attended by Serah Kimani, Jean Watermeyer, Simon Broadley and Miranda Waldron.

LEAVE BY THE DIRECTOR

Zeiss factory January
USA in June
Eindhoven Sept

SERVICE TO INDUSTRIAL AND OTHER EXTERNAL USERS

The Unit offers its facilities on an ad hoc basis to external users. Clients exploiting these services during 2009 were: Automotive Quality, Chemical Investigations, Fascination Wigs, Industricon, Iziko Museum, Jeffaries and Green, Metsep, Mintek, Namakwa Sands, NBI, NECSA, One eighty Degrees, ONFO, Origen, Patterson and Cooke, Precision Press, SASOL, Shimoda Biotech and TF Design. These clients almost exclusively use the S440 SEM and the 912 TEM and together accounted for 91 hours instrument time.

VISITORS TO THE UNIT

Dr Michael Lawrence, Walter and Eliza Hall Institute, Melbourne Australia.
Dr Pavel Penczek, Department of Biochemistry and Molecular Biology, University of Texas

REPLACEMENTS AND REPAIRS IN 2009

Instruvac Rotary pump
Gilson iongetter pump
Iongetter pump power supply
Ceiling in west corridor
Return air supply
Air conditioning in TEM and SEM rooms
Software from Tecnai

SUMMARY

Prepared by: Associate Professor B.T. Sewell and Mrs. Miranda Waldron

TABLE 1

Services Offered by the Unit during 2009

Service	Comment
Access to 1200EX TEM	Used by 7 people
Access to S440 SEM	Used by 123 people
Access to 912 TEM	Used by 67 people
Access to F20 FEGTEM	Used by 8 people
Training on the 1200EX TEM	new users were trained
Training on S440 SEM	12 new users were trained
Training on the 912 TEM	new users were trained
Training on the F20FEG TEM	new users were trained
Access to Ultracut S Ultramicrotome	Used by people
Training on Ultracut S	new users were trained
Cryo-microtomy and immunolabelling	Well used
Sectioning of blocks supplied by the user	Well used
Embedding of biological specimens in methacrylate and epoxy	Well used
Freeze substitution	Used
Sputter Coating of specimens supplied by user	Very popular service
Critical point drying of specimens supplied by the user	Very popular service
Printing of EM films	Service used
Access to optical microscopy facilities	Used
Access to Image Processing and Analysis (Analysys)	Used
Element analysis by EDS	Well used.
"Introduction to EM for Biologists"	This course was held once.
Access to specimen polisher	Used
Access to high vacuum coating plant and accessories	Adequately used
Store of EM consumables	Used by most users
Access to prep lab	Well used
Collection of books and journals on microscopy	Used
Vacuum Leak Detection	Used
Production of CD ROMS	Well used
Digitization of transparent media on LS4500	Used
Digitization of transparent media on Leafscan	Used
High quality ink-jet printer	Very popular
Flat bed scanner	Well Used

Table 2

2009 User List

Archaeology	
Hinkman, A	
Galimberti, M	PhD
Archer, W	PhD
Botany	
Singh, K	MSc
Cardiovascular	
Human, P	
Chemical Engineering	
Amod, M	MSc
Case, J	MSc
Chonco, Z	PhD
Fisher, N	PhD
Gertenbach, R	Staff
Hsu, P	PhD
Julies, M	PhD
Lindiswe	MSc
Mangere, M	MSc
McEwan,L	MSc
McFadzeen, B	Staff
Mungwe, N	PhD
Na, K	Hons
Naglio, D	MSc
Paradza, N	MSc
Premesh	PhD
Reddy, T	MSc
Rendani, R	MSc
Sinethemba	Staff
Stoddart, F	MSc
Tsoenyane, M	MSc
Van Heerden, T	PhD
Viljoen, A	
Wright, C	PhD
Chemistry	
Banothjile, A	PhD
Botha, S	Staff
Egan, T	Staff
Teleke, V	Staff
Centre for Materials Engineering	
Finkelstien, L	MSc
George, S	PhD
Knutsen, R	Staff
Letaba, G	PhD
Matthews, L	MSc
Mias, K	MSc
Miller, D	Staff
Noma, P	Hons
Petersen, S	MSc
Williams, R	Hons
Fine Art	
Buffler, F	Staff
Geological Sciences	
Diener, J	Staff
Dreyer, T	MSc
Rowe, C	Staff
Thakani, T	Hons
Viglietti, P	MSc
IIDMM	
Tiedt, F	Staff
Molecular and Cell Biology	
Beckett, M	PhD
Botha, S	PhD
Burger, M	
Cooper, K	Staff
Cross, B	PhD
Mbewana, S	Staff
Meyers, P	Staff
Mortimer, E	Staff
Mutepfa, D	PhD
Pineo, C	
Rholand, J	Staff
Rybicki, E	Staff
Mechanical Engineering	
Lakhi, F	Hons
Ramesh, R	Hons
Tait, B	Staff
Thorpe, K	MSc
Carulei, O	MSc
Medical Virology	
Van Niekerk, M	
Physics	
Jones, S	PhD
Magunje, B	MSc
Nzikou, M	PhD
Monfal, L	MSc
Ramanandraitsiory, I	MSc
Schadrack	PhD
Unugibe, D	PhD
Structural Biology	
Kimani, S	PhD
Thuku, N	PhD
Van Rooyen, J	PhD
Varsani, A	Staff
Watermeyer, J	Staff
Woodward, J	PhD
Zoology	
De Ponte, M	MSc
Kotze, M	PhD

Other Universities and Technicons

Cape Peninsula University of Technology	
Fester, V	Staff
Malange, K	MSc
CSIR	
Khati, S	Staff
Joubert M	Staff
Ithemba Labs	
Abiona, A	MSc
Ajao B	MSc
Masina, C	MSc
Mbuyisa, P	MSc
Menovar, D	Post Doc
Mlungi, P	MSc
Mtshali, S	PhD
Nanky, P	MSc
Nechaev, A	Staff
Ngoma, B	MSc
Nolwashizoli, N	PhD
Khamich, S	MSc
Sam	
Stihomeni, J	PhD
Sone, B	PhD
Topic, M	Staff
Tswane University of Technology	
Sheku, K	
University of Helsinki	
Verimaki, P	Visitor
Violetta	Visitor
University of Mozambique	
Guambe	PhD
University of Stellenbosch	
Biochemistry	
Vd Merwe	
Chemistry	
Lakay, E	PhD
Kotze, I	PhD
Microbiology	
Smith, J	
Polymer Science	
Aboubaker, A	PhD
Baily N	PhD
Basson, N	PhD
Bayley, G	MSc
Cronje, L	Staff
D'Aguilar	PhD
Geesh, N	PhD
Koen, H	
Malherbe, I	
Pound, G	Post Doc
Smit, E	Staff
Sutherland, A	MSc
Zengeni, E	MSc
Process Engineering	
Lasper, Q	
Rose, J	
Wine Biotechnology	
Moore, J	Staff
University of the Western Cape	
Chemistry	
Adeniyi, O	PhD
Greyling, C	Staff
Hendricks, N	PhD
Majeed, A	PhD
Martinovic	
Murithii, G	PhD
Muyosa, N	PhD
Olushola, A	MSc
Earth Sciences	
Adekola, S	PhD
Falipe, O	MSc
Olufunke, F	PhD
Madzivipe, G	PhD
Oluwaseun, F	PhD
IMBM	
Lukasa, L	PhD
WITS	
Gale, B	MSc

Commercial Users:

180 Engineering	
Cotton, J	Staff
Opperman, M	Staff
Automotive Quality	
Buss, D	Staff
Chemical Investigations	
Norton, S	Staff
Fascination wigs	
Berry, C	Staff
Industricon	
Wepener, P	Staff
Iziko Museum	
Hosford, J	Staff
McMillan, I	Staff
Jeffaries & Green	
Bone	Staff
METSEP	
van Wyk C	Staff
Mintek	
McPhearson, J	Staff
Mokoena, L	Staff

Namakwa Sands	
Kiewets, D	Staff
Philander, C	Staff
National Botanical Inst	
Boatwright, S	Satff
Snjman, D	Staff
NECSA	
Patience	Staff
ONFO	
Fitton, J	Staff
Origen	
Jones, J	Staff
Paterson & Cooke	
Malloch, R	Staff
Wickens, J	Staff
Zengeni, E	Staff
Precision Press	
Ledgerwood, J	Staff
Sasol	
Savage, N	Staff
Savati	
Renrie, A	Staff
Shimoda Biotech	
Swart, H	Staff
TF Design	
Halimer, G	Staff

TABLE 3