

**A Study of the Effects of Mobile-Phone Type
Signals on Calcium Ion Levels Within A Human
Leukaemic T Cell Line.**

**A thesis submitted for the degree
of Doctor of Philosophy**

**by Charles G. Cranfield
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Declaration

I declare that this thesis contains no material that has been previously submitted for a degree or award at any other university or educational institution. To the best of my knowledge this thesis contains no material that has been previously published, or written by another person, except where due reference has been given. Where the work was based on joint research or publications, the relative contributions of the respective workers or authors has been disclosed.

Signed

Charles Gordon Cranfield

Table of contents

Acknowledgments	ii
Financial Acknowledgments	iii
Declaration	iv
Table of contents	v
List of Figures	ix
List of Tables	xiii
List of Abbreviations used	xv
Abstract	xviii
1.0 Review of literature	1
1.1 Introduction	1
1.1.1 Some Definitions	3
1.1.2 Epidemiological studies into the health affects of mobile phones	5
1.1.3 <i>In Vivo</i> studies into the affects of RF radiation of mobile phones	6
1.1.4 <i>In Vitro</i> studies into the affects of RF radiation of mobile phones	7
1.1.5 Athermal RF research <i>in vitro</i>	9
1.2 Physiology of T cells	11
1.2.1 The Jurkat E6.1 T lymphocyte.....	12
1.2.2 Activation of T lymphocytes	13
1.2.3 Calcium fluctuations in T lymphocytes	17
1.2.4 Calcium oscillations in T lymphocytes.....	20
1.3 How RF might interact with biological calcium ion levels.	22
1.3.1 RF/Cancer link hypothesis.....	23
1.3.2 Models for EMF influences on intracellular calcium oscillations	27
1.4 Imaging Jurkat T Lymphocytes whilst exposing them to RF energy.	32
1.4.1 Artificial stimulators of cytoplasm calcium influx for positive control.....	33
1.4.2 Confocal Microscopy.....	33
1.4.3 The Exposure Chamber	36
1.4.4 Calcium Fluorescence Imaging.....	37
1.5 Aims of this research.	39
2.0 Materials and Methods	41

2.1 Materials.....	41
2.1.1 Coaxial RF exposure device.....	41
2.1.2 List of RF exposure generating equipment.....	46
2.1.3 Tissue sample.....	47
2.1.4 $[Ca^{++}]_i$ assay technique:.....	48
2.1.5 Imaging system.....	48
2.1.6 Temperature monitoring.....	49
2.1.7 Media perfusion equipment.....	49
2.2 Methods.....	50
2.2.1 Preparation of the in vitro RF exposure device.....	50
2.2.2 Harvesting of T cells.....	52
2.2.3 Loading of Fluo-3 fluorescent dye into the Jurkat cells.....	52
2.2.4 Cell viability test.....	53
2.2.5 Loading the cell suspension into the exposure chamber.....	53
2.2.6 Imaging of cells inside chamber.....	53
2.2.7 RF Exposure.....	55
2.2.8 Experimental time-lines.....	59
2.2.9 Data analysis.....	60
2.3 Cell Monitoring Software.....	61
2.3.1 Cell monitoring software – procedures.....	62
2.4 Experimental naming protocol.....	72
2.5 Statistics.....	72
3.0 Results of preliminary experiments.....	75
3.1 Imaging calcium in Jurkat cells over time.....	75
3.1.1 The effects of photobleaching.....	75
3.1.2 Spontaneous Oscillations.....	77
3.1.3 Mitogen stimulation.....	79
3.1.4 Ionophore stimulation.....	82
3.1.5 Fluorescent dye leakage into cell organelles.....	84
3.1.6 Multi-photon imaging.....	85
3.2 Experiments in the RF exposure chamber.....	86
3.2.1 Imaging through the shim.....	86
3.2.2 Temperature control inside chamber.....	88
4.0 Results from RF exposure experiments.....	91

4.1 Flow rates	91
4.2 Temperature Control	91
4.3 RF Exposure control	94
4.4 Changes in mean fluorescence and mean calculated [Ca²⁺] levels.....	96
4.5 Changes in calcium measured as a mean change in slope.....	101
4.5.1 Regression slope data for experiments.	102
4.5.2 Tables of mean slopes.....	108
4.6 Changes in the average frequency of intracellular calcium oscillations.	110
4.6.1 Average frequency changes for 1.5W/kg exposures.....	110
4.6.2 Average frequency changes for 7.5W/kg exposures.....	113
4.6.3 Table for mean average frequency data	115
4.6.4 Comparisons between factors in average frequency data	116
4.6.5 Summary of average frequency data.....	118
4.7 Calcium oscillations as a result of RF energy.....	119
5.0 Discussion of experimental procedures.....	120
5.1 Athermal experiments.....	120
5.2 Determination of calcium concentrations.....	121
5.3 Imaging quality.....	122
5.3.1 Refractive index mismatch	122
5.3.2 Noise.....	123
5.3.3 Possible confounding factors	125
5.4 Exposure measurements	126
5.5 Improving experimental techniques	126
5.5.1 Improved fluorescent probes and imaging.....	126
5.5.2 Improved tissue selection.....	127
6.0 Discussion of results.....	130
6.1 Change in average fluorescence and calcium ion concentration.	130
6.2 Change in slope data	132
6.3 Change in average frequency	133

6.4 Oscillation patterns	137
6.5 RF/Cancer link hypothesis.....	138
6.6 Conclusions	139
References.....	141
Appendix A: Monitoring cell growth and viability	163
A.1 Counting cells in a haemocytometer	163
A.2 Cell viability check.....	164
Appendix B: Personal Communication concerning dye compartmentalisation.	
.....	166
B.2 Reply from Donald O’Malley	166
B.3 Reply from Pedro Camello.....	167
B.4 Reply from Steven Cody.....	167
B.5 Donald O’Malley in response to Steven Cody	168
B.6 Steven Cody in response to Donald O’Malley	170
Appendix C: Data sheets	172
C.1 900MHz Continuous Wave 1.5W/kg. Cells not activated.....	172
C.2 900MHz Continuous Wave 1.5W/kg. Cells activated by PHA.....	174
C.3 900MHz Pulse Modulated at 217Hz, 1.5W/kg. Cells not activated.	176
C.4 900MHz Pulse Modulated 217Hz, 1.5W/kg. Cells activated by PHA.....	178
C.5 900MHz Continuous Wave 7.5W/kg. Cells not activated.....	180
Appendix D: Design specifications for heating collar	181
Appendix E: Diagram of T cell receptor signalling pathways	182
Appendix F: Average SAR calculation above viewing hole	183
Appendix G: Example of cell fluorescence readings before and after background subtraction.	185
Personal Publications	192

List of Figures

- Figure 1.1** Diagram outlining of the electromagnetic spectrum highlighting regions of ionising radiation and non-ionising radiation. Mobile telephone communications fall into the non-ionising part of the spectrum. 2
- Figure 1.2** A major signalling pathway in typical T lymphocytes after stimulation with a mitogenic lectin such as phytohemagglutinin (PHA). Activation of the T cell receptor complex eventually leads to the formation of 1,4,5-trisphosphate (IP₃) and diacylglycerol (DAG) from phosphatidylinositol 4,5-bisphosphate (PIP₂) via various enzymatic processes. 14
- Figure 1.3** Intracellular signalling cascade in T lymphocytes after the production of IP₃. IP₃ leads to the release of calcium from intracellular stores, which in turn opens plasma membrane calcium channels eventually leading to the activation of transcription factors such as NFAT. 15
- Figure 1.4** The signalling pathway in T lymphocytes after production of DAG. DAG activates protein kinase C (PKC), which in turn activates a series of other kinases leading to the activation of the transcription factor NF_κB. 17
- Figure 1.5** Two plots showing two types of typical intracellular calcium ion concentration oscillatory activity in T lymphocytes. The top plot shows spiking from a baseline, whilst the bottom plot is a representation of spiking on a raised plateau. 21
- Figure 1.6** A model of EMF interaction on calcium oscillations formulated by Eichwald and Walleczek (1996), where EMF would affect the enzyme activation by the mitogen (PHA in this case). Dashed line (1) indicates that the activation of the enzyme is dependent on cell stimulation, and dashed line (2) indicates where the enzyme in turn acts on the feedback control of the signal pathway. In their model EMFs would effect the kinetics of the enzyme which would then effect calcium signalling. 29
- Figure 1.7** Diagram outlining the light path from the laser to the sample on the microscope stage via an optical fibre using an Optiscan™ confocal microscope. Returning fluorescent light is diverted through special dichroic mirrors and filters to a photomultiplier tube. Source: www.optiscan.com.au 35
- Figure 1.8** Diagram of a Crawford Cell. This device has often been used to expose biological samples to RF energy. Source www.ifi.com 36
- Figure 1.9** Calcium ion dependent fluorescence emission spectra for Fluo-3 when excited with 488nm laser light. Source: Haugland, 1996. 39
- Figure 2.1** Cross-sectional schematic diagram of the RF exposure device. The cells were located on a 0.15mm thick coverglass at the bottom of the cell chamber. The cell chamber itself was perfused with RPMI 1640 medium. Cells were imaged using a 60x, oil immersion objective on an

inverted stage microscope through a central 1.0mm hole in a thin stainless steel shim. The device disassembles for cleaning at the horizontal dashed lines.	42
Figure 2.2 Picture of outside of the exposure device showing perfusion ports for cell media and chemical reagent addition, with an extra port for the insertion of a temperature probe. On top of device is an “N” type connector for attachment to coaxial cable.	43
Figure 2.3 Exposure device unscrewed at midline showing top section with teflon™ filler with stainless steel core, and bottom section sealed teflon chamber.	44
Figure 2.4 Bottom section of RF exposure devise unscrewed from the shim attachment. Teflon™ chamber attaches to bottom shim section with coverglass sandwiched between. Imaging from microscope is done through 1mm viewing hole.	45
Figure 2.5 Plan view of access ports to the cell chamber in the RF exposure device.	45
Figure 2.6 Picture of RF generating equipment showing signal generator, amplifier, power meters pulse generator and oscilloscope.	47
Figure 2.7 Picture of the Luxtron 790 fluoroptic thermometer with fluoroptic probe.	49
Figure 2.8 Schematic diagram of nutrient medium perfusion circuit for RF exposure devise.	51
Figure 2.9 Picture of custom made microscope stage with micropositioners to guide objective under central viewing hole.	54
Figure 2.10 Schematic RF circuit diagram.	56
Figure 2.11 Chart showing variations in Specific Absorption rates over the central viewing hole in shim. Ordinate: SAR values (in W/kg) on the coverglass above the 1mm hole in the stainless steel shim. Abscissa: radial distance (in mm) from the center of symmetry.	58
Figure 2.12 Image from cell monitoring software, highlighting regions of interest around cells.	62
Figure 2.13 Cell monitoring software menu commands for loading multi-image TIFF images for analysis.	63
Figure 2.14 Data window in cell monitoring software requesting a number which corresponds to an image (or frame) in a sequence of images to be opened following the <i>Read Cell Image</i> command.	63
Figure 2.15 Image of cells in cell monitoring software with a rectangle highlighting average cell dimensions for software algorithms to use in cell selections.	65
Figure 2.16 Image of cells after a command typed in text box that adds a colour palette to the cell images.	66
Figure 2.17 Command used to select circular regions of interest around cells of radius 25 pixels. One circular region was selected for background fluorescence measurements.	67
Figure 2.18 Data-window requesting a file name for image fluorescence data to be saved as. The <i>track cells</i> option can also be toggled in this window.	69
Figure 2.19 Display of cell coordinate data after cell selection. Every row represents a different cell’s coordinates. The first 2 columns are the pixel coordinates of where the cell image was selected along the X and Y-axis respectively. The next 2 columns are the X and Y coordinates of the centre of the circular regions of interest created. These 2 columns are almost always virtually	

identical to the first 2 columns. The next column is the circular region of interest's radius in pixels, which is followed by the area of that region in pixels. The last column shows the average fluorescence intensity measured in that region of interest. The last row, with the low fluorescence intensity, is the background region. This region was always selected last in experiments for ease of identification later.....	70
Figure 3.1 Gradual photobleaching of Fluo-3 in Jurkat cells with laser power at 100% and maximum current setting.	76
Figure 3.2 Graph showing typical spontaneous calcium oscillation spikes in a Jurkat T lymphocyte loaded with the fluorescent probe Fluo-3.....	78
Figure 3.3 Graph showing typical spontaneous calcium ion spiking on a raised plateau in a Jurkat T lymphocyte.....	79
Figure 3.4 Jurkat cells imaged before the addition of PHA (left), and 15min after 6.5µg/ml of PHA was added (right). About 10 min following the addition of PHA, cells can be seen attaching together making single cell analysis difficult.....	80
Figure 3.5 Comparison of oscillating cells stimulated with PHA and those not stimulated.....	82
Figure 3.6 Graph showing change in fluorescence in a single Jurkat T lymphocyte as a result of ionophore (A23187, 3.8µM) stimulation.	83
Figure 3.7 Jurkat cells imaged before the addition of A23187 (left), and after 3.8µM of A23187 was added (right). An increase in intracellular calcium as a result of ionophore stimulation is responsible for an increase in fluorescence in cells.	83
Figure 3.8 Two-photon fluorescence microscopy image of Jurkat cells loaded with leakage-resistant Indo-1 PE3. Brighter regions highlight areas of high cytoplasmic calcium ion concentrations. The darker intracellular region is the large T lymphocyte nucleus where the fluorescent dye had not sequestered.....	85
Figure 3.9 Diagram of coverglass and shim hole showing cone of acceptance. Source: Anderson 2001.	87
Figure 3.10 Pictures of a special heating collar designed to fit over the 60x oil objective (also pictured). The heating collar was warmed by a perfusion of heated water through tubing from a water-heater. Temperature could be controlled in the collar by adjusting the flow rate through the tubing.	89
Figure 4.1 Five typical temperature plots from 1.5W/kg experiments show a steady rise in temperature that then forms a plateau.	93
Figure 4.2 Mean change in fluorescence (Period B - Period A) between RF exposed and sham exposed cells for 1.5W/kg exposures.....	99
Figure 4.3 Mean change in fluorescence (Period B - Period A) between RF exposed and sham exposed cells for 7.5W/kg exposures.....	100
Figure 4.4 Diagram showing a representative example of regression slope for fluorescence data for the control period (Period A) and the RF/sham exposure period (Period B).	102

Figure 4.5 Comparison of mean regression slopes for control periods (Period A) to the 1.5W/kg RF exposure periods (Period B) for all conditions.....	104
Figure 4.6 Comparison of mean regression slopes for control periods (Period A) to the 1.5W/kg sham exposure periods (Period B) for all conditions.....	105
Figure 4.7 A comparison in the <i>change</i> in mean regression slopes (Period B - Period A) for RF exposed cells and sham exposed cells.....	105
Figure 4.8 Comparison of mean regression slopes for control periods (Period A) to the 7.5W/kg RF exposure periods (Period B) for cells not activated by PHA.....	106
Figure 4.9 Comparison of mean regression slopes for control periods (Period A) to the 7.5W/kg sham exposure periods (Period B) for cells not activated by PHA.....	106
Figure 4.10 Mean change in regression slopes (Period B- Period A) for cells exposed to 900MHz continuous wave RF energy 7.5W/kg, and those sham exposed. Cells were not activated by PHA.	107
Figure 4.11 Changes in average frequencies between RF exposed and sham exposed cells.	112
Figure 4.12 Overall average change in mean frequencies between control period (Period A) and RF exposed (1.5W/kg)/sham periods (Period B) for all cells analysed.	113
Figure 4.13 Change in average mean frequencies (Period B -Period A) in CW 900MHz RF (7.5W/kg SAR) energy exposed and sham exposed cells, not activated by PHA.	114
Figure 4.14 Change in average frequency (Period A - Period B) for PHA activated and unactivated cells exposed to 1.5W/kg RF energy.....	116
Figure 4.15 Change in average frequency (Period A - Period B) for cells exposed to 1.5W/kg continuous wave 900MHz RF and 1.5W/kg 217Hz pulse modulated 900MHz RF.	117
Figure 5.1 Comparison of 3D image reconstructions that would occur with a refractive match for the coverglass, objective and media, and a refractive index mismatch.....	123
Figure 5.2 Image of Jurkat cells inside exposure chamber, with banding interference in the form of curved lines altering the image to a small degree.	124
Figure A.1 Grid shape similar to that found on most haemocytometer slides. The region labelled 5 here is typically for counting red blood cells, whilst regions 1-4 are for counting white blood cells.	164
Figure F.1 Diagram of viewing region divided into annuli.	183

List of Tables

Table 2.1 Experimental time-line for unactivated Jurkat T lymphocytes.	59
Table 2.2 Experimental time-line for PHA activated Jurkat T lymphocytes.	59
Table 3.1 Number of unactivated and PHA activated cells judged to be showing calcium ion spikes..	81
Table 3.2 Changes in relative permittivity and conductivity at 900MHz exposure of RPMI 1640 solution (supplemented with 10% Fetal Bovine Serum) as a result of increased temperature. Source; Anderson (2001), submitted.....	90
Table 3.3 Comparisons of relative permittivity and conductivity values of RPMI-1640 exposed to 900 MHz at 37°C, both with and without 10% Foetal Bovine Sera. Source: Anderson (2001), submitted.....	90
Table 4.1 Mean change in temperature versus time regression slopes for 1.5W/kg experiments and 7.5W/kg experiments.	92
Table 4.2 Forward and backward power readings for experiments that were continuous wave (900MHz) 1.5W/kg exposures. Cells not activated by PHA.....	94
Table 4.3 Forward and backward power readings for experiments that were continuous wave (900MHz) 1.5W/kg exposures. Cells activated by PHA.....	94
Table 4.4 Forward and backward power readings for experiments that were pulsed modulated (900MHz, pulsed 217Hz) 1.5W/kg exposures. Cells not activated by PHA.....	95
Table 4.5 Forward and backward power readings for experiments that were pulsed modulated (900MHz, pulsed 217Hz) 1.5W/kg exposures. Cells activated by PHA.....	95
Table 4.6 Forward and backward power readings for experiments that were continuous wave (900MHz) 7.5W/kg exposures. Cells not activated by PHA.....	95
Table 4.7 Mean fluorescence measurements for control Periods A and RF/sham Periods B for all conditions. n refers to the number of cells analysed.	96
Table 4.8 Mean Calcium ion concentration measurements for control Periods A and RF/sham Periods B for all conditions.....	97
Table 4.9 Change in mean fluorescence between the control period (Period A) and the RF/sham exposure period (Period B) for all conditions.	98
Table 4.10 Changes in mean calcium ion concentrations between the control period (Period A) and the RF/sham exposure period (Period B) for all conditions.....	100
Table 4.11 Mean slopes for the control period (Period A) and the RF exposed period (Period B) in all experiments where RF exposure was real.	108
Table 4.12 Mean slopes for the control period (Period A) and the sham exposure period (Period B) in all experiments where there was no RF exposure.	109
Table 4.13 Mean change in regression slopes between the control period (Period A) and the RF/sham exposure period (Period B) for all conditions.	110

Table 4.14 showing <i>change</i> in average frequency of calcium ion fluctuations between Period A (control) and Period B (Sham/RF) in Jurkat T lymphocytes under various exposure conditions.	115
Table 4.15 Comparisons of the change in average frequencies (Period B –Period A) between all activated cells and unactivated cells (1.5W/kg); and 900MHz continuous wave exposed cells and 900MHz pulse modulated 214Hz exposed cells (1.5W/kg).	118
Table 4.16 A comparison between cells judged to be oscillating from a base-line, and those oscillating on a plateau level for sham exposed experiments and RF exposed experiments. There was no significant difference in the number of cells oscillating between exposed and sham exposed cells as determined by a χ^2 -test.	119
Table F.1 SAR calculations at 50 μ m distances from centre of viewing hole for 44mW forward power.	184
Table F.2 Average SARs as a proportion of viewing areas.	184

List of Abbreviations used

μm	micro-meters
μM	micro-molar
[Ca²⁺]_e	Extracellular calcium ion concentration
[Ca²⁺]_i	Intracellular calcium ion concentration
AM	Acetoxymethyl (when referring to fluorophores)
AM	Amplitude Modulation (when referring to EM energy)
APC	Antigen Presenting Cell
BAPTA	1,2-bis(2-aminophenoxy) ethane-n,n,n',n'-tetraacetic acid
Ca²⁺	Calcium ions
CaN	Calcineurin (Ca ²⁺ /Calmodulin protein phosphatase)
CD3	Cluster of differentiation No. 3
CD4	Cluster of differentiation No. 4
CD8	Cluster of differentiation No. 8
CIF	Calcium influx factor
Con-A	Concanavalin-A
CRAC-channels	Calcium release-activated calcium channels
dB	Decibels
dBm	Decibels relative to one milliwatt
DAG	Diacylglycerol
DC	Direct current
DMSO	Dimethyl sulphoxide
EGTA	ethylene glycol-bis(β-aminoethyl ether)-n,n,n',n'-tetraacetic acid
ELF	Extremely Low Frequency
EM	Electro-magnetic
EMF	Electromagnetic field
F	Fluorescence
GHz	giga-hertz
IκB	Inhibitory protein kappa B

ICNIRP	International Commission on Non-Ionising Radiation Protection
IL-1	Interleukin-1
IL-2	Interleukin-2 (Formerly <i>T Cell Growth Factor</i>)
IP₃	Inositol 1,4,5-trisphosphate
K_{Ca}	Calcium-activated potassium channels
K_d	Dissociation constant
LDH	Lactate dehydrogenase
MEKK	Mitogen activated protein kinase kinase- kinases (also called MAPKKK)
MHz	mega-hertz
min	minute
mW	milliwatts
MMP	Method of multipoles
NA	Numerical Aperture
NFκB	Nuclear Factor kappa B
NF-AT	Nuclear Factor of Activated T cells
NK	Natural Killer cell
nm	nano-meters
nM	nano-molar
P	Power (dBm)
PHA	Phytohemagglutinin
PIP₂	Phosphatidylinositol 4,5-bisphosphate
PKC	Protein kinase C
PM	Pulse Modulation
PMT	Photo multiplier tube
RF	Radiofrequencies
ROI	Region of interest
s	seconds
S₁₂	Transmission loss between points 1 and 2 (dB)
S₁₃	Transmission loss between points 1 and 3 (dB)
SAR	Specific Absorption Rate (W/kg)

T	Tesla
TCR	T Cell Receptor
TNF	Tumour Necrosis Factor
UHF	Ultra high frequency
UV	Ultra violet
W	watts

Abstract

The work presented here outlines experiments done using a novel RF exposure chamber. This device allows biological cells to be exposed to microwave radiation similar to those emitted by mobile telephones, whilst imaging them using a laser scanning confocal microscope.

Jurkat E6.1 T lymphocytes in the exposure chamber were kept within $\pm 0.5^{\circ}\text{C}$ of 37°C , allowing for the investigation of possible athermal effects of microwave energies. These cells were loaded with the fluorescent probe Fluo-3 AM, which is specific for calcium ions, and were monitored over two 10minute periods. The first period being a control period, the second being a period where the cells were either exposed to RF energy or sham exposed. Another 5min imaging period was for the positive control, where maximal fluorescence can be achieved by the addition of the ionophore *A23187*.

5 different conditions for cell exposure were investigated. Both continuous wave 900MHz and continuous wave 900MHz pulse modulated at 217Hz exposures were carried out on cells that were either unactivated, or those that were activated by the mitogen *phytohaemagglutinin* (PHA). For these 4 conditions the average Specific Absorption Rate (SAR) was calculated to be 1.5W/kg. A group of unactivated cells were also exposed to continuous wave 900MHz energy with an average SAR calculation of 7.5W/kg.

Results showed that no significant changes in calcium ion levels occurred when averaged fluorescence slopes were compared between RF exposed cells and the control period. The mean change in slopes (exposed/sham period – control period) between cells that were exposed and those sham exposed also showed no significant difference.

Following an inference made in the work of Galvanovskis *et al.* (1999)¹ who showed there is a change in the calcium ion oscillation spectrum as a result of 50Hz magnetic fields, a measure of the mean frequencies of all cells was determined using a Fast Fourier Transform (FFT) analysis. The change in the average mean frequencies in cells was then measured for all conditions. Of statistical significance was the change in average mean frequency between the control period and the sham/exposed period between cells that were exposed and those sham exposed, when cells were activated by PHA. The results also showed that there was an overall drop in average mean frequency as a result of RF exposure. Assuming there is a biological significance to the findings of this thesis, careful analysis of the calcium dynamics of tissue samples and cell types associated with RF exposure from mobile phones would need to be carried out to determine what they are. This was unfortunately beyond the scope of the present study.

¹ Galvanovskis et al (1999), *Bioelectromagnetics* 20: 269-276.