6th Advanced course in embryology
6 July 2013 - London, UK
Venue
Park Plaza River Bank London
18 Albert Embankment
London SE1 7TJ, UK

Language
The official language of the workshop will be English.

Scientific secretariat
Serono Symposia International Foundation
Salita San Nicola da Tolentino, 1/B
00187 - Rome, Italy

Associate Project Manager: Simona Pantaleoni
Specialist Medical Advisor: Angelo Marino
Phone: +39 06 420 413 569
Fax: +39 06 420 413 677
info@seronosymposia.org

Serono Symposia International Foundation is a Swiss Foundation
with headquarters in 14, rue du Rhône, 1204 Geneva, Switzerland

Organising secretariat
Meridiano Congress International
Via Sapri, 6 - 00185 Rome (Italy)
Senior Project Manager: Federica Russetti
Phone: +39.06.88.595.209 - Fax: +39.06.88.595.234
E-mail: f.russetti@meridiano.it

We value your opinion!
We are continually trying to develop and improve our educational initiative to provide you with cutting-edge learning activities.
Before and after this educational course you will be asked to answer an online survey to help us better tailor our future educational initiatives.
We thank you for participating!

Register to Serono Symposia International Foundation website:
www.seronosymposia.org

follow us on twitter SSIF_RM
http://twitter.com/SSIF_RM
6th Advanced course in embryology

Serono Symposia International Foundation live educational course on:

6th Advanced course in embryology
6 July 2013 - London, UK

Aim of the live educational course
The course offers an interactive programme with many of the lectures followed by video sessions and debate on hot topics. The aim is to give learners both the latest knowledge and the opportunity to share their experiences with renowned biologists and embryologists. The 2013 course will be dedicated exclusively to the most important aspects of ART laboratory procedures - providing a comprehensive view of advanced techniques such as cryopreservation, risk management in the laboratory and embryo viability.

Learning objectives
After attending this course learners will be able to:
• Analyse the clinical effectiveness of new diagnostic tools for male infertility and apply these new acquisitions in clinical practice
• Identify new objective criteria for selecting the embryos with the highest implantation potential
• Recognise the actual bias in the quality management in IVF laboratory
• Modify the routine clinical activity necessary to deliver quality procedures in IVF laboratory

Target audience
Expert biologists and embryologists working in assisted reproductive medicine seeking the latest information about advanced techniques and scientific innovation.

Accreditation
Serono Symposia International Foundation (www.seronosymposia.org) is accredited by the European Accreditation Council for Continuing Medical Education (EACCME) to provide the following CME activity for medical specialists. The EACCME is an institution of the European Union of Medical Specialists (UEMS), www.uems.net. The course on “6th Advanced course in embryology” (London, UK – July 6, 2013) is designated for a maximum of 6 (six) hours of European CME credits (ECMEC). Each medical specialist should claim only those credits that he/she actually spent in the educational activity. EACCME credits are recognized by the American Medical Association (AMA) towards the Physician’s Recognition Award (PRA). To convert EACCME credit to AMA PRA category 1 credit, please contact the AMA.

SSIF adheres to the principles of the Good CME Practice group'

All Serono Symposia International Foundation programmes are organized solely to promote the exchange and dissemination of scientific and medical information. No forms of promotional activities are permitted. All Serono Symposia International Foundation programmes are made possible thanks to the unrestricted Educational grants received from: Arseus Medical, Besins Healthcare, Bristol-Myers Squab, Celgene, Centre d’Escorossi Múltiple de Catalunya (Vall d’Hebron University Hospital), Centre -Hépato-Biliaire (Hôpital Paul Brousse), Croissance Conseil, Cryo-Save, Datanalysis, Dos33, Esaote, Ferring, Fondazione Humanitas, Fundación IVI, GE Healthcare, GlaxoSmithKline Pharmaceuticals, IPSEN, International Society for Fertility Preservation, Johnson & Johnson Medical, K.I.T.E., Karl Storz, Lumenis, Merck Serono Group, PregLem, Richard Wolf Endoscopie, Sanofi-Aventis, Stallergenes, Stoler, Teva Pharma, Toshiba Medical Systems, Université Catholique de Louvain (UCL), University of Catania.
Scientific organiser

Robert Fischer
Fertility Centre Hamburg
Hamburg, Germany

Scientific co-organisers

Alan Thornhill
Department of Biosciences, University of Kent, UK
Chair, ALPHA, Scientists in Reproductive Medicine

Zsolt Peter Nagy
Reproductive Biology Associates, Atlanta, Georgia, USA
Secretary, ALPHA, Scientists in Reproductive Medicine

Serono Symposia International Foundation developed this programme in collaboration with: ALPHA, Scientists in Reproductive Medicine
List of faculty members

Edson Borges
Fertility - Centro de Fertilização Assistida
São Paulo, Brazil

Mona Bungum
Laboratory Director, Reproductive Medicine Centre (RMC)
Skåne University Hospital
Malmö, Sweden

Robert Fischer
Fertility Centre Hamburg
Hamburg, Germany

Gedis Grudzinskas
Editor (clinical) RBM Online
London, UK

Howard Jacobs
London Medicine Clinic
London, UK

Henry J. Leese
Professor of Biology Hull York Medical School
Hull York, UK

Nicholas Macklon
Department of Obstetrics and Gynecology
University of Southampton
Southampton, UK

Peter Zsolt Nagy
Reproductive Biology Associates
Atlanta, Georgia, USA

Laura Rienzi
G.EN.E.R.A Centre for Reproductive Medicine
Rome, Italy

Karen Turner
Oxford Fertility Unit
Oxford Business Park North
Oxford, UK

Jonathan Van Blerkom
University of Colorado
Boulder, Colorado, USA

Dagan Wells
Institute of Reproductive Sciences
Oxford, UK
### Session I: New technologies in reproductive medicine

<table>
<thead>
<tr>
<th>Time</th>
<th>Activity</th>
<th>Chair(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>09.00</td>
<td>Serono Symposia International Foundation (SSIF) opening and scientific organiser introduction to the course</td>
<td>R. Fischer (Germany)</td>
</tr>
<tr>
<td>09.10</td>
<td>High-magnification sperm morphology examination and ICSI outcomes</td>
<td>E. Borges (Brazil)</td>
</tr>
<tr>
<td>09.35</td>
<td>The effect of sperm DNA fragmentation on miscarriage rates</td>
<td>M. Bungum (Sweden)</td>
</tr>
<tr>
<td>10.00</td>
<td>Mitochondrial activity during oocyte and embryo development</td>
<td>J. Van Blerkom (USA)</td>
</tr>
<tr>
<td>10.25</td>
<td>Question time</td>
<td></td>
</tr>
<tr>
<td>10.50</td>
<td>Coffee break</td>
<td></td>
</tr>
<tr>
<td>11.20</td>
<td>Nutritional requirements: from the oocyte to the blastocyst, implication for embryo culture</td>
<td>H. Leese (UK)</td>
</tr>
<tr>
<td>11.45</td>
<td>Proteomics of the human endometrium and uterine fluid: a pathway to biomarker discovery</td>
<td>N. Macklon (UK)</td>
</tr>
<tr>
<td>12.10</td>
<td>Question time</td>
<td></td>
</tr>
<tr>
<td>12.30</td>
<td>Lunch break</td>
<td></td>
</tr>
</tbody>
</table>

### Session II: From subjective to objective criteria in embryo selection: where are we?

<table>
<thead>
<tr>
<th>Time</th>
<th>Activity</th>
<th>Chair(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>13.30</td>
<td>How to identify embryos with the highest implantation potential</td>
<td>P. Z. Nagy (USA)</td>
</tr>
<tr>
<td>13.55</td>
<td>Video Session on laboratory techniques</td>
<td></td>
</tr>
<tr>
<td>14.20</td>
<td>PGS: state of the ART, why, when, how?</td>
<td>D. Wells (UK)</td>
</tr>
<tr>
<td>14.45</td>
<td>How to implement QMS in ART laboratory</td>
<td></td>
</tr>
<tr>
<td>15.10</td>
<td>Question time</td>
<td></td>
</tr>
</tbody>
</table>

### Session III: Success in IVF: Debate on who is the most important?

<table>
<thead>
<tr>
<th>Time</th>
<th>Activity</th>
<th>Chair(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>16.10</td>
<td>Debate on who is the most important?</td>
<td></td>
</tr>
<tr>
<td>17.10</td>
<td>Question time and take home messages</td>
<td></td>
</tr>
<tr>
<td>17.50</td>
<td>Conclusion remarks</td>
<td></td>
</tr>
<tr>
<td>18.00</td>
<td>End of the live educational course</td>
<td></td>
</tr>
</tbody>
</table>
Serono Symposia International Foundation adheres to guidelines of the European Accreditation Council for Continuing Medical Education (EACCME) and all other professional organizations, as applicable, which state that programs awarding continuing education credits must be balanced, independent, objective, and scientifically rigorous. Investigative and other uses for pharmaceutical agents, medical devices, and other products (other than those uses indicated in approved product labeling/package insert for the product) may be presented in the program (which may reflect clinical experience, the professional literature or other clinical sources known to the presenter). We ask all presenters to provide participants with information about relationships with pharmaceutical or medical equipment companies that may have relevance to their lectures. This policy is not intended to exclude faculty who have relationships with such companies; it is only intended to inform participants of any potential conflicts so participants may form their own judgments, based on full disclosure of the facts. Further, all opinions and recommendations presented during the program and all program-related materials neither imply an endorsement, nor a recommendation, on the part of Serono Symposia International Foundation. All presentations solely represent the independent views of the presenters/authors.

The following faculty provided information regarding significant commercial relationships and/or discussions of investigational or non-EMEA/FDA approved (off-label) uses of drugs:

<table>
<thead>
<tr>
<th>Faculty Name</th>
<th>Information Provided</th>
</tr>
</thead>
<tbody>
<tr>
<td>Edson Borges</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Mona Bungum</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Robert Fischer</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Gedis Grudzinskas</td>
<td>Declared the participation in Merck Serono and IBSA sponsored speaker’s bureau</td>
</tr>
<tr>
<td>Howard Jacobs</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Henry J. Leese</td>
<td>Declared honoraria or consultation fees from Irvine Scientific, and being a stakeholder and adviser to Novocellus Ltd</td>
</tr>
<tr>
<td>Peter Zsolt Nagy</td>
<td>Declared honoraria or consultation fees from EMD Serono, MERCK, Origio; being a member of Origio, Unisense, MEB and being a stakeholder of MEB.</td>
</tr>
<tr>
<td>Laura Rienzi</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Jonathan Van Blerkom</td>
<td>Declared no potential conflict of interest</td>
</tr>
<tr>
<td>Dagan Wells</td>
<td>Declared receipt of grants and contracts: Merck-Serono (grant) and being stakeholder in Reprogenetics (stock)</td>
</tr>
</tbody>
</table>

The following faculty have provided no information regarding significant relationship with commercial supporters and/or discussion of investigational or non-EMEA/FDA approved (off-label) uses of drugs as of June 21, 2013.

Karen Turner
Nicholas Macklon
Biosketch
Dr. Edson Borges Jr. obtained his MD degree in 1984 at the University of Campinas, Brazil. He then prepared his first PhD, in Urology, in 2005, at the Federal University of São Paulo and his second PhD in Gynaecology, in 2007, at the Botucatu Medical School in São Paulo State University.

Since 1992, he is the founder, partner and managing director of Fertility – Centro de Fertilização Assistida in São Paulo, Brazil and Scientific Director and Head of 1 Sapientiae Institute in São Paulo, Brazil.

Dr. Borges has published over 100 papers, review articles, books and chapters and is a member of the editorial boards of several scientific Journals.

Mona Bungum is embryologist, PhD and laboratory director of Reproductive Medicine Centre, Skanes University Hospital. Currently her main research interest is sperm DNA integrity testing in fertility. She has been author of about 40 per-reviewed papers and book-chapters.

Robert Fischer is Founder and Medical Director of the IVF unit at the Hamburg Fertility Center, a leading German IVF centre, and one of the largest in the country. In July 1998 the Fertility Center of Hamburg was one of the first centres in Germany and worldwide to introduce certified quality management according to ISO 9001. In 2002 the IVF laboratory was ISO 17025 certified. Prior to this, Robert Fischer was Medical Director of the first outpatient IVF unit in Hamburg. Author of numerous publications in national and international scientific journals and books, as well as a lecturer at conferences worldwide, Dr Fischer is an active member of the American Society of Reproductive Medicine, founding member of the European Society of Human Reproduction and member of its advisory committee. He is also a founding member of the AG Gynäkologische Endokrinologie und Fortpflanzungsmedizin and Berufsverband Reproduktionsmedizinischer Zentren, both in Germany.
Biosketch

Gedis Grudzinskas
Editor (clinical) RBM Online
London, UK

Currently in independent practice as a consultant in Gynaecology and Infertility, formerly Professor of Obstetrics & Gynaecology (1983-2003) and until 2008 Emeritus Professor, at the Royal London Hospital and St Bartholomew’s Hospital, and until 2008 Medical Director of the London Bridge Fertility, Gynaecology and Genetics Centre. Present interests and activities include Reproductive Medicine, Infertility, Gamete donation, Fertility Preservation and education in Scientific Writing.

Howard Jacobs
London Medicine Clinic
London, UK


His clinical and research interests focussed on disorders of human reproduction and their correction. He retired from the University and the National Health Service in 1998 and from all clinical practice in 2008.

Henry J. Leese
Professor of Biology Hull York Medical School
Hull York, UK

Henry Leese’s research has been on early mammalian embryos, particularly, periconceptual nutrition. His BSc (Reading) and PhD (Imperial College) were in Physiology and Biochemistry. He has worked in Zurich and Harvard Medical School, is a Fellow of the Royal College of Obstetricians and Gynaecologists [ad eundem], Honorary Fellow of the Association of Clinical Embryologists and British Fertility Society and received the Marshall Medal of the Society for Reproduction and Fertility in 2010. He was a member of the HFEA from 1998-2002. He is Editor-in-Chief of Human Fertility and teaches human nutrition and metabolism to students in biology and medicine.
Nick Macklon is Professor of Obstetrics and Gynaecology at the University of Southampton, and co-founder and Director of the Complete Fertility Centre Southampton. After training in Edinburgh and Glasgow, he was appointed Senior Lecturer at Erasmus University Rotterdam, working with Bart Fauser. He subsequently moved to Utrecht as Professor of Infertility and Periconceptional Medicine and Departmental Head of Reproductive Medicine and Gynaecology, and returned to the UK in 2009. He has published extensively in the fields of ovarian stimulation and endometrial receptivity, and has edited several textbooks. He is a past Chairman of the ESHRE Special Interest Group in Reproductive Medicine and editorial appointents include Associate Editorship of Human Reproduction Update, Human Reproduction, Reproduction and RBM Online. He currently holds visiting professorships at Adelaide University and the Copenhagen University Hospital.

Dr. Nagy obtained his MD (1986) and his Ob&Gyn degrees (1996) at the Semmelweis Medical University in Budapest. His PhD was granted at the Free University of Brussels (VUB) in 1997, on the topic of “ICSI: technical and biological aspects to increase efficiency”. Dr. Nagy has acquired a distinctive knowledge and experience on embryo science including novel assessment methods. More recently, he has focused on the area of cryopreservation, especially oocyte vitrification, that has contributed to the development of a donor cryo-bank. He is author or co-author of more than 150 publications and book chapters. Dr. Nagy serves on the board of several professional societies and is Associate Editor for Human Reproduction and RBM Online. Dr. Nagy is the Scientific and Laboratory Director at Reproductive Biology Associates, Atlanta, Georgia.

Dr. Laura Rienzi is Senior Clinical Embryologist, Adjunct professor at the University of Rome (Tor Vergata) and Laboratory Director at G.EN.E.R.A Centres for Reproductive Medicine in Rome, Urbertide and Marostica, Italy. With academic degrees in Biology and Reproductive Medicine she has an intensive activity including educational, editorial and practitioner and author of almost 100 articles, reviews and book chapters. Her primary areas of expertise are within cell preservation, embryo morphology and optimization of laboratory procedures for IVF.
Biosketch

Karen Turner
Oxford Fertility Unit
Oxford Business Park North
Oxford, UK

As an experienced embryologist, Karen joined Oxford Fertility Unit to lead the laboratory team in 2002. The Unit now performs over 2000 cycles per annum. Previously, Karen has lead laboratory teams at both Sheffield Fertility Centre and Burton Centre for Reproductive Medicine. Karen is a State Registered Clinical Scientist. She was Chair of the Association of Clinical Embryologists (ACE), the UK professional body for Embryologists, from 2000 to 2003 and was an External assessor on the Training Committee for a number of years. Karen was the first embryologist to sit on the British Fertility Society committee and has previously been an external advisor for the HFEA. She has recently become the first President of the ACE.

Jonathan Van Blerkom
University of Colorado
Boulder, Colorado, USA

Jonathan Van Blerkom is a Professor in the Department of Molecular, Cellular and Developmental Biology at the University of Colorado in Boulder and Laboratory Director at Colorado Reproductive Endocrinology at the Rose Medical Center in Denver [USA]. He has been engaged in studies of molecular and cellular aspects of mammalian development since 1970 and beginning in 1982, clinician IVF and studies of human follicles, oocytes and embryos. He has published numerous original research articles, reviews, and coauthored or edited books dealing with early mammalian development, including the human. He has been an invited speaker at international conferences and symposia and served as associated editor for several journals in the field of reproductive medicine. He is currently a Section Editor for Reproductive BioMedicine Online and the North American Editor for Zygote. He is actively engaged in research focused on the role(s) of mitochondria in early development and the molecular organization of the oolemma with respect to how they determine developmental competence for the human oocyte and early embryo.
Dagan Wells has been actively involved in preimplantation genetic diagnosis (PGD) and the study of human gametes and embryos for over two decades, conducting his first PGD cases in 1992. He spent several years developing novel PGD tests at University College London, accomplishing the first comprehensive chromosome analysis of cells from human embryos in 1998, using a combination of whole genome amplification and comparative genomic hybridisation (CGH). In 1999 Dagan moved to the United States and joined Reprogenetics, one of the world’s largest providers of PGD services. In 2003 he initiated Reprogenetics’ highly successful single gene PGD program, testing embryos for numerous serious inherited conditions. Dagan later joined the faculty of Yale University Medical School, where he spent four years running a research laboratory, before returning to the UK in late 2007. His growing research group is now located in the Nuffield Department of Obstetrics and Gynaecology at the University of Oxford. Dagan’s work has led to the publication of more than 120 peer-review publications and book chapters and has been recognized with the award of numerous prizes and grants. He is a Fellow of the Royal College of Pathologists and currently serves on the Editorial Boards of several international journals. Dagan also directs Reprogenetics-UK, an independent laboratory offering state-of-the-art PGD services to IVF clinics throughout the UK and internationally.
Abstracts
In the last decade a new approach involving real-time, high-magnification observation of unstained spermatozoa, named 'motile sperm organelle morphology examination' (MSOME), has been introduced. This system reaches a magnification of x6,600 to x13,000. The presence of vacuoles in the sperm nucleus seems to be the most important defect detected by MSOME. The origin of sperm vacuoles is disappointingly unknown and even after several investigations the question remains: Are sperm vacuoles degenerative structures or common physiologic features of the sperm head? The majority of the studies suggest that there is a link between the presence of vacuoles and sperm function, either with the acrosome reaction, chromatin condensation or DNA integrity. The MSOME is the first advent after ICSI for the evaluation of sperm nucleus. Since it enables the identification of minor morphological abnormalities in the sperm cell that will be referred to ICSI it results in a promising technique that has come to stay.

The incorporation of MSOME, together with a micromanipulation system, has allowed the introduction of a modified ICSI procedure called intracytoplasmic morphologically selected sperm injection (IMSI). Positive associations between IMSI and the percentage of high quality embryos, pregnancy, implantation, miscarriage and delivery and live birth rates have been previously demonstrated. The improvements with IMSI technique were observed mainly in cases of previous implantation failures, high sperm DNA fragmentation rate, male factor, advanced maternal age and unexplained infertility.

This presentation will discuss the efficacy of MSOME and IMSI techniques, focusing attention on the potential clinical application of the selection of strictly morphologically normal spermatozoa in patients undergoing intracytoplasmic sperm injection treatments.
During the last decade, the assessment of sperm DNA integrity has emerged as a new and promising biomarker of semen quality that may help in the discrimination between infertile and fertile men and in predicting pregnancy outcome in assisted reproduction (ART). Moreover, sperm DNA fragmentation has been linked to the risk of miscarriage, and therefore suggested to be used as a diagnostic tool in couples with recurrent pregnancy loss. However, although male partners in couples experiencing recurrent pregnancy loss are shown to have increased amounts of sperm DNA fragmentation compared to controls, the evidence for a direct association between sperm DNA fragmentation and pregnancy loss is controversial. One reason for the conflicting data is likely the different sperm DNA fragmentation tests used in the studies as well as in the metaanalysis published. Several tests to assess sperm DNA damage are available, however, they all are based on different methodology, they measure different aspects of DNA damage and the clinical value of the tests vary. The exact knowledge about which type of DNA damage the different tests measure is unclear, and few of the tests have the ability to distinguish between single stranded and double stranded DNA breaks. Such information could be relevant as the type of DNA damage could have different effect on the outcome of pregnancy.

In order to further clarify to which degree sperm DNA fragmentation tests may be used as a diagnostic tool, in counselling patients with subfertility, miscarriage problems or other we need to refine available tests or develop new able to specify the type of DNA damage. Moreover the need for more optimal sperm selection methods able to deselect sperm with DNA damage for ART is obvious.
All organelles are essential for normal cell function, and defects that occur because of disease, toxic exposure, including reactive oxygen species (ROS), or mutation effecting nuclear or mitochondrial DNA (mtDNA), can have adverse consequences ranging from cytopathologies associated with reduced function (e.g., myopathies) to cellular lethality. While a similar statement can be made for organelle function in the mature oocyte and preimplantation stage embryo, recent studies of how developmental competence is established and maintained during these stages in general, and for the human oocyte in particular, have focused attention on one organelle, the mitochondrion. Most are familiar with their bioenergetic role in producing ATP, yet how this activity translates into normal nuclear, cytoplasmic and plasma membrane function during preovulatory oocyte maturation, fertilization and the preimplantation embryogenesis has only recently emerged.

This presentation has four aims or learning objectives: (i) to acquaint clinical embryologists with the different functions and activities mitochondria have at these stages and how these organelles can be involved in the regulation of developmental competence; (ii) to discuss the notion that differential mitochondrial activity at these stages may be spatially compartmentalized and as such, influence redox-, ROS- and calcium-dependent signaling pathways, which may have corresponding spatially localized regulatory roles in normal information flow associated with competence; (iii) to distinguish between mtDNA copy number and mitochondrial mass as determinants of competence in human IVF such that disproportionate segregation of mitochondria in cleaving human embryos with an otherwise normal mitochondrial mass and mitochondrial DNA complement can lead to early developmental arrest and embryo demise; and (iv) to address the question of whether mitochondria can be considered the primary nexus of most developmental ills that are observed in human IVF. The last issue will be discussed in terms of whether proposed strategies of mitochondrial rescue or rejuvenation suggested for women of advanced maternal age, or where IVF has previously failed, are valid in a physiological context for clinical IVF where no known pathogenic mtDNA mutation exists at high load.
Oxidative metabolism provides the major source of energy for oocytes and preimplantation embryos. Oocyte metabolism is augmented through interaction with the granulosa/cumulus cells. In terms of nutrient and oxygen consumption, the early stages of preimplantation development are relatively quiescent metabolically, before a sharp rise in activity at the blastocyst stage due to the high energy demands of the Na+, K+, ATPase required for blastocoel cavity formation and of protein synthesis for the first net growth of the embryo which begins at this time. Pyruvate is obligatory for the first cleavage division of the zygote after which a range of substrates including amino acids and endogenous triglyceride may be utilised. Prior to morula formation, glucose consumption is low although there is a small glucose requirement for intracellular signaling purposes. Glucose uptake increases at the blastocyst stage with a proportion being converted to lactate by ‘aerobic glycolysis’, at least in vitro, and a further quantity being channeled through the pentose phosphate pathway to provide biosynthetic precursors. These findings, [summarised in Leese, 2012] have been valuable in deciding what constituents should be included in embryo culture media but less useful in defining the concentrations themselves. Data on the nutrient and ionic content of oviduct and uterine fluids have been more useful in establishing quantitative requirements. The zygotes of large animals such as the cow, pig and sheep, and most likely those of the human, can survive reasonably well in culture for at least 72 hours in the absence of exogenous nutrients while mouse zygotes die within 10 hours. Such information has led to a re-awakening of interest in endogenous energy reserves, notably triglyceride, during preimplantation development, and in the adaptable, semi-autonomous nature of the early embryo. Non-invasive assays indicate that the uptake of pyruvate and glucose, and the turnover (depletion and appearance) of amino acids, are associated with subsequent embryo viability and have the potential to provide biomarkers with which to optimize the composition of embryo culture media and increase the evidence base in this field. The approaches used in developing human embryo culture media will be illustrated by examining the case for supplementation with the nutrient creatine, which after conversion to creatine phosphate via the enzyme creatine (phospho) kinase provides a buffer against changes in ATP concentration. In somatic cells, creatine kinase associates with the mitotic spindle to provide a local source of ATP; an association which is present in the early mouse embryo [Forsey et al, 2013]. Arguably, the addition of creatine to embryo culture media might help ensure that cytokinesis, a critical process in cleavage stage embryos proceeds physiologically, but extensive preliminary research, which will be discussed, would be required before this was attempted. In summary, a variety of approaches are required in the formulation of human embryo culture media, coupled with a range of endpoints of embryo health; non-invasive and invasive, including the long-term health of the offspring, to ensure efficacy and safety in clinical IVF.

References
Current understanding of the molecular process which renders the human endometrium receptive has as yet failed to be translated into effective therapeutic modulators of receptivity. Assisting progress in this area is the emergence of new analytic technologies which hold promise to accelerate understanding of the regulation of endometrial receptivity. The ability to simultaneously analyze a large number of molecules within or secreted by the endometrium is opening a new “window” on this complex process. ‘Secretomics’ describes the application to the analysis of protein constituents of endometrial secretions. The particular value of secretomics in the study of the endometrium derives from the simple and minimally invasive means by which material for study can be obtained (endometrial fluid aspiration). This offers both researchers and clinicians a window on the intra-uterine environment.

The endometrial secretome is known to contain a number of mediators which modulate endometrial receptivity, and which may be involved in the maintenance and nurturing of ascending spermatozoa and the preimplantation embryo. The primary components are proteins, amino acids, electrolytes, glucose, urea, cytokines, growth factors, metalloproteinases and their inhibitors, immunoglobulins, alpha-1 antitrypsin precursor, haptoglobin and transferrin. Endometrial fluid aspiration may offer a non-invasive window on the receptive state as it may be performed immediately prior to embryo transfer in IVF cycles without negatively affecting implantation rates. In a study of women undergoing IVF, a profile of cytokines, growth and signaling factor concentrations measured in endometrial secretions aspirated immediately prior to undergoing embryo transfer were found to be significantly correlated with the chance of successful implantation.

Developments in mass spectrometry using protein profiling and peptide sequencing to elucidate underlying cellular biological processes have also opened up the field of proteomics to the study of endometrial receptivity. A recent study compared the proteomes of pre-receptive versus receptive human endometrium. Two proteins, annexin A2 and stathmin 1, were suggested as important in predicting the receptivity status.

The application of proteomics technology to the endometrium may be utilised in the future to search for novel biomarkers of endometrial receptivity and infertility, and to increase understanding of the molecular basis of diseases such as endometriosis.
Embryo viability assessment is one of the most critical procedures in the embryology laboratory. The clear goal of assisted reproduction is to provide the maximum chance of pregnancy, but at the same time to minimize the risk of multiple implantation – can be achieved if a single embryo with the highest implantation potential is selected for transfer. Additionally, if supernumerary embryos are available, those with viability should be identified for cryopreservation for future use.

Traditional way of assessing embryo viability in the last decades has been by observing morphological characteristics by light microscope. Basic information, such as cell number, fragmentation, cytoplasmic features, and other attributes have been correlated with implantation potential, however, this association have not been always consistently strong, providing a relatively low predictive value. Recently, a number of novel approaches to assess embryo viability have been proposed. Different molecules/components of the spent embryo culture media have been targeted, including pyruvate, glucose, oxygen, HLA-G, and Leptin. High technicality procedures, such as Ultra-microfluorescence, Microspectrophotometry, Enzyme-linked immunoabsorbent assay have been employed showing correlation with embryo developmental potential and in some cases with implantation potential. It has also been demonstrated that protein complement of the culture environment of embryo is also altered pending developmental potential and viability of the embryo. Surface-enhanced laser desorption ionization time-of-flight mass spectrometry and protein microarray techniques have been used to measure specific changes of the proteome. Amino-acid turn-over has also been reported to be a useful marker of embryo development in recent years. Gene expression patterns tested from the cumulus cells of the freshly retrieved oocyte also showed good correlation with derived embryo developmental potential, yet adding one more option for additional tool for gaining information on embryo viability. Metabolomic profile of spent culture medium, employing vibrational spectroscopy (Near infrared or Raman spectroscopy) also demonstrated good predictive value, correlating with embryo viability. Recent scientific studies using time-lapse video monitoring and evaluation of specific cleavage pattern of embryo development demonstrated clear correlation with blastocyst developmental potential and viability of the embryo.

All these novel tools and approaches are greatly promising, however, it is required that they will be tested thoroughly in prospective clinical trials before routine clinical implementation.
This year sees the twentieth anniversary of the first attempts to improve IVF success rates by assessing embryos for chromosome abnormalities (aneuploidy), a strategy sometimes referred to as Preimplantation Genetic Screening (PGS). The underlying theory of PGS is based upon three simple facts: 1) human oocytes and embryos are frequently chromosomally abnormal; 2) aneuploidy is almost always lethal; 3) chromosome abnormalities have very little effect on embryo morphology. This means that in a typical IVF cycle many of the embryos produced will carry chromosome abnormalities and yet these embryos will often be chosen for transfer because they look just as ‘good’ as their chromosomally normal siblings. Inevitably, the transfer of non-viable embryos, at the expense of healthy embryos from the same cohort, will harm pregnancy rates.

The theory underlying PGS seems extremely attractive and yet the clinical utilization of PGS has been controversial. For the most part, the controversy stems from data produced during a series of randomized controlled trials (RCTs) that aimed to quantify the improvements to IVF outcomes (if any) delivered by PGS. None of the trials were able to demonstrate any benefits attributable to PGS (See Fragouli and Wells, 2012 for review). So where are we now? The entire concept of PGS has been revitalized in the last few years by the introduction of new technologies for chromosome analysis. Data now beginning to emerge has indicated that the failure of the earlier randomized clinical trials was primarily a consequence of deficiencies in the embryological and/or genetic techniques used. In particular, the strategy of using fluorescence in situ hybridisation (FISH) to assess a relatively small number of chromosomes in cleavage stage embryos seems to have been problematic. Information from studies using the new generation of genetic technologies, which permit a comprehensive chromosomal analysis, strongly supports the concept of PGS.

There is now excellent data confirming that, even at the blastocyst stage, many embryos are aneuploid and that this problem rapidly worsens with advancing female age (Fragouli et al., 2012). For women aged 30-35 years, ~40% of blastocysts are chromosomally abnormal, while for those aged 40-42 aneuploidy affects ~70%. Additionally, it has recently been proven that transfer of chromosomally abnormal embryos leads to implantation failure or miscarriage in at least 95% of cases (Scott et al., 2012). Furthermore, data from thousands of embryos clearly demonstrates that the impact of aneuploidy on morphology is too subtle for this problem to be avoided by assessing the morphological appearance of embryos (Alfarawati et al., 2011). These facts all suggest that PGS should work, but what evidence is there that it can really make a difference in the clinic? After the disappointments of the first generation of PGS technologies, doctors and embryologists in many centers are understandably cautious about the utilization of the new chromosome screening methods.

Evidence that second-generation PGS does indeed improve outcomes is now accumulating at a rapid rate. At the time of writing four randomized controlled trials had reported significantly elevated implantation and/or pregnancy rates per cycle and dramatically reduced rates of miscarriage (Schoolcraft et al., 2012; Yang et al., 2012; Forman et al., 2013; Scott et al., 2013). Interestingly, despite these studies using different genetic techniques for chromosome assessment, occurring in different labs and focusing on different populations of patient, all showed a similar level of benefit, with implantation rates increased by about one-third in the cycles that had PGS.

Arguments that PGS does not improve IVF outcomes appear to be fading and even some of the most fervent critics of chromosome testing have reassessed their position and begun to offer PGS to their patients. However, publication of further randomized trials in peer-review journals will be needed before the case for PGS can be considered irrefutable. At present, several important questions concerning the use of PGS still need to be answered: There are several methods available for chromosome analysis, does it matter which is used? There is data to support the use of PGS in good prognosis patients and poor prognosis patients, so should it be offered to all couples or are their specific patient groups that benefit more than others? Most of the randomized trials supporting the use of PGS have employed biopsy at the blastocyst stage. Does PGS also work when applied to polar bodies or blastomeres biopsied at the cleavage stage? This lecture will consider the latest PGS data in an attempt to answer some of these questions.

References
In 2006 the European Tissue Directive made it mandatory for a Quality Management System to be in place in the IVF laboratory. Quality Management is a broad term that describes a program of evaluating the quality of care using a variety of methodologies and techniques and it covers issues like quality assurance, risk management and quality improvement.

Within the IVF laboratory, quality management involves examining every process in terms of what goes into it e.g. gametes, medication; what happens to it whilst in there e.g. technical procedures, equipment; and what comes out e.g. embryos, pregnancies; thereby constantly striving for best practice. ISO 9000 is one set of quality standards that laboratories often choose to measure themselves against if implementing a QMS. These standards look at different areas which encompass anything from management responsibility to measurement, analysis and improvement within the laboratory.

In implementing a QMS within the laboratory, a unit is encouraged to consider many things like who is responsible for the various duties within and who deals with the responsibilities in that individual’s absence. Communication within and between teams and feedback from customers (patients) is a necessary part of QMS so that the laboratory can learn and improve. It is important to ensure that Standard Operating Procedures (SOP) are in place in the laboratory and that every process and procedure is covered by an SOP, that staff are following the SOP and that they are performing them to the required standard.

Monitoring and measuring of processes within the laboratory, through audit where appropriate, is key to ensuring that those areas identified as critical to the laboratory perform as expected and that any downward trends are detected early. Such key performance indicators may include things like fertilisation rates, blastocyst rates or implantation rates. With regards to staff performance, internal and external quality control schemes can be adopted to ensure that all individuals perform at the same level e.g. in performing semen analyses or grading embryos.

The overall impact of implementing a QMS within the laboratory is to streamline processes, enable better monitoring, measurement and response to any issue and thus overall improve outcome both in terms of patient experience and success rates.