Infections, Infertility, and Assisted Reproduction

Assisted reproductive technology (ART) treatment is vulnerable to the hazard of potential infection from many different sources: patients, samples, staff, and the environment. Culture of gametes and embryos in vitro provides multiple targets for transmission of potential infection, including the developing embryo, neighbouring gametes and embryos, the couple undergoing treatment, and other couples being treated during the same period. This unique situation, with multifaceted opportunities for microbial growth and transmission, makes infection and contamination control absolutely crucial in the practice of assisted reproduction, and in the laboratory in particular.

This unique and practical book provides a basic overview of microbiology in the context of ART, providing an up-to-date guide to infections in reproductive medicine. The relevant facets of the complex and vast field of microbiology are condensed and focused, highlighting information that is crucial for safe practice in both clinical and laboratory aspects of ART. This is an essential publication for all ART clinics and laboratories.

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To: our families,

Robbie and Bethany
John and Justin
Paul and Maxwell James

With love and thanks for their patience, tolerance, and support.

Contents

Foreword

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Wolbachia are gram-negative, intracellular bacteria that shelter in the gonads of invertebrates, and have profound effects on the fertility of their hosts. In some species, infected hosts can only reproduce parthenogenetically, in others cytoplasmic incompatibility prevents infected males from breeding with uninfected females, and in some cases genetically determined male embryos are transformed into females. *Wolbachia* engineers effects, as do all parasites, for selfish ends. Although this bizarre pathology is unknown in medical science, the relationships between microbes and human fertility are nonetheless complex, fascinating and important for the practice of reproductive medicine.

Unfortunately, and usually without advance warning, microbes occasionally enter the clinical laboratory through infected semen or vaginal tissue. When this occurs, a patient's treatment outcome may be seriously compromised because microbes can quickly deplete nutrients in culture media and alter the pH, and it would be irresponsible to knowingly transfer an infected embryo or semen to a patient. Bacterial and fungal growth are often obvious and easily tested, but how often do infectious agents go unrecognized and contribute to the problems of infertility, treatment failure and even possibly affect the child-to-be?

This is the first book on medical microbiology that has been written by experts in reproduction for clinical scientists and physicians in their own field. They are to be congratulated on filling a gap in the

literature between microbiology and assisted reproduction, which they achieve in three sections. The first serves as a primer of medical microbiology for readers who are unfamiliar or rusty on the subject. The second focuses on microbes that have implications for human reproduction, whether by causing infertility (a familiar example being *Chlamydia*) or by jeopardizing reproductive safety (such as HIV). In the final section, the practical implications of this knowledge are addressed in the context of infertility, and especially the setting of the clinical embryology laboratory. Every embryologist is trained in sterile techniques, filtration of media and prudent use of antibiotics to keep out the bugs, but a deeper knowledge of the foundations of safe and effective practice is an undervalued safeguard for patient care.

Preface

The world of microbes is intrinsically fascinating. Microbes are abundant in every place on earth where larger living creatures exist, and they can also thrive in habitat extremes where no other kind of organism can survive for long: from deep under the sea to the stratosphere – up to 32 km in the atmosphere, in oil formations and in hot telluric water. It is estimated that the total biomass of microbes probably exceeds that of all the plants and animals in the biosphere. This biomass is predominantly composed of bacteria, and these microorganisms play a crucial role in recycling much of the organic material in the biosphere. Despite their minute size, microorganisms carry out all the fundamental processes of biochemistry and molecular biology that are essential to the survival of all living species. Although their size may give them the illusion of being 'primitive', their range of biochemical and biophysical capabilities is far wider than that of higher organisms. One of their most important properties is adaptability and versatility, a key feature in their long history of evolution. Fossil records suggest that at least some members of the microbial world, oxygen-producing cyanobacter-like organisms, had evolved 3.46 billion years ago (Schopf, 1993); a viable fungus, *Absidia corymbifera*, was recovered from the right boot that accompanied the frozen, well-preserved prehistoric corpse, 'Ice Man', aged approximately 5300 years (Haselwandter & Ebner, 1994).

Records of microbial disease that probably influenced the course of history can be found in archaeological sites of early civilizations, as well as in later

periods of history. A hieroglyph from the capital of ancient Egypt dated approximately 3700 BC illustrates a priest (Ruma) with typical clinical signs of a viral infection, paralytic poliomyelitis. The mummified body of the Pharaoh Siptah, who died in 1193 BC, also shows signs of classic paralytic poliomyelitis, and the preserved mummy of Rameses V has facial pustular lesions suggesting that his death in 1143 BC was probably due to smallpox. This virulent disease was endemic in China by 1000 BC, and had reached Europe by 710 AD. Hernando Cortez transferred the disease to the Americas in 1520, and it appears that around 3 500 000 Aztecs died of smallpox within the next two years – arguably precipitating the end of the Aztec empire.

In the early 1330s an outbreak of deadly Bubonic plague occurred in China, one of the busiest of the world's trading nations, and rapidly spread to Western Asia and Europe. Between 1347 and 1352 this plague, 'The Black Death', killed 25 million people – one-third of the population of Europe – with far-reaching social, cultural and economic repercussions.

The world of assisted reproduction is equally fascinating, and is one that also has a long history of evolution. The concept of assisted procreation by human artificial insemination was a topic of discussion between Jewish philosophers as early as the third century AD, and tales exist of fourteenth-century Arab horse breeders obtaining sperm from mated mares belonging to rival groups, using the sperm to inseminate their own mares. Assisted reproduction explores the fundamental principles behind the creation of a new life, the intricate biological mechanisms that are involved when mature gametes come into contact, combine genetically and set in motion a cascade of events leading to the correct expression of genes that form a new individual.

Microbiology and assisted reproduction both deal with a miniature world, magnified for observation with the help of microscopy. Culture of microorganisms and of preimplantation embryos in vitro requires special media and growth conditions to promote cell division, and both are visualized and assessed at various stages following cell division. A knowledge of microbiology is fundamental to the safety and success of assisted reproductive techniques – but the field of microbiology is vast, and continues to increase in complexity with the discovery of new organisms and implementation of new medical treatments. The field of assisted reproductive technology also continues to expand and develop, particularly in areas of science and biotechnology. Members of an assisted reproduction team are not usually also experts in infectious diseases, and may find it difficult to identify and follow significant areas of microbiology that can impact upon their practice.

The purpose of this book is to select areas and topics in microbiology that are specifically relevant to assisted reproductive technology (ART), in order to provide a very basic background of facts and fundamental principles. A background of understanding can help prevent contamination and transmission of disease in ART, and also limit the opportunities for microbial survival in embryo culture and cryopreservation systems. The book is divided into three Parts:

Part I provides an outline of microorganism classification and identification, as a foundation for understanding the relationships and the differences between the types of organisms that may be encountered in routine ART practice. The microorganisms that are human pathogens or resident flora, and those that are routinely found in the environment are introduced. Each chapter includes an Appendix of antimicrobial drugs and their modes of action.

Part II details organisms that cause disease of the reproductive tract and those that are blood-borne pathogens, describing their

etiology, pathogenesis, diagnosis, pathology and treatment.

Part III describes the practical application of microbiology principles within an assisted reproduction laboratory.

REFERENCES

Haselwandter, K. & Ebner, M. R. (1994). Microorganisms surviving for 5300 years. *FEMS Microbiology Letters*, **116**(2), 189–93.

Schopf, J. W. (1993). Microfossils of the early Archean apex chart: new evidence of the antiquity of life. *Science*, **260**, 640–6.

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