Index

accreditation of quality management systems 20 - 6accreditation process 22-6 audit visits 23-6 definition of accreditation 22 non-compliances 23-6 time limit on validity 22 adjustment, definition 99 air conditioning systems 124 - 7air quality air conditioning systems 124 - 7biological safety cabinets (BSC) 127-9 laminar air flow (LAF) hoods 127-9, 145 alarm systems for laboratory equipment 11-14 albumin used in culture media 34 - 5altitude, influence on pH 66-7 ambient air quality see air quality amino acids (organic osmolytes) in culture media 46-9, 62 role in intracellular pH regulation 62 ammonia (NH₃) production in media 33 antibiotic resistance 141, 145 antibiotics, use in culture media 142, 147 ART (Assisted Reproductive Technology) and Murphy's Law 1-7, 189 approach to problem-solving 2 - 7history of development 2, 8 importance of apparently trivial phenomena 2-7 recognizing that it is difficult 2 regulation and standards 8-9 use of troubleshooting flowcharts 6 Aspergillus infection 144-5 204

β-alanine, organic osmolyte in culture media 47-9 B2 medium (Ménézo) 28 Bavister model for regulation of intracellular pH 62 betaine, organic osmolyte in culture media 47-9 bioassays culture of surplus embryos 17 key points in the use of 16 procedures 15-16 quality control issues 15-16 single or multipronucleate embryo culture 17 sperm viability test 17 biological safety cabinets (BSC) 127-9 bisphenol A (BPA) harmful effects of 32 in plasticware 32 blood-borne infections 139, 148 - 51Bourn Hall, Cambridge, UK 8 bovine embryos, effects of light exposure 107 Brown, Louise 2, 5, 8, 37 BSE (bovine spongiform encephalopathy) 139 buffers 68-70 Burns, Robert 1 calcium precipitation in culture media 28, 33 calibration, definition 99 calibration certificates 10-11 cell cytoplasm, effects of culture media osmolality 45

ATP production in the embryo

audit of quality management

systems 23-6

59-60

cell DNA repair capacity 111 cell volume regulation and culture media osmolality 45–9 certification of quality management systems 20–6

definition of certification 21 - 2time limit on validity 22 chemical fingerprinting of mineral oils 37 Chlamydia trachomatis infection 143 CO₂ measurement methods 118 - 20College of American Pathologists (CAP) 8-9 competency assessments 192 - 4computer-assisted sperm assessment (CASA) 80 conduction, definition 80 conflict escalation model (Glasl) 198 conflict resolution 195-8 consumable products and supplies, quality control 14-15 continuing professional development (CPD) 193 convection, definition 80-1 Crabtree effect 60 crisis management 198 cryopreservation of gametes, embryos, and tissue 153 changes associated with 153 factors affecting cellular response to freezing 153 - 4stresses on cells 153 troubleshooting flowchart 167 troubleshooting process map for vitrification 166 cryopreservation process, media osmolality 55 cryopreservation protocols controlled-rate freezers 160 cryoprotectant agents 153-5 seeding 156 slow freezing (equilibrium freezing) 154-6, 158-60 vitrification (nonequilibrium/ultra-rapid freezing) 156-8



cryoprotectant agents 153-5 cryostorage troubleshooting flowchart 168 cryostore audit 165-9 cryostore management 160-2 contact with patients who have frozen material 165-9 cryostore audit 165-9 dewars 161-2 donor cryobanks 168 final disposition of embryos 165-9 liquid nitrogen safety precautions 162-4 removing cryopreserved material from storage 165-9 cryostore specifications 162-3 culture dish preparation effects on culture osmolality 49-55 practical tips for keeping osmolality constant 52-5 culture media 29-34 amino acids (organic osmolytes) 46-9, 62 antibiotics in 142, 147 calcium precipitation problem 28, 33 effects of ammonia (NH₃) production 33 effects of glucose 49 effects of glutamine decomposition 33 formulations 45-7 generation of hydrogen peroxide (H₂O₂) 29-30 generation of reactive oxygen species (ROS) 29-30 110-13 glutathione (GSH) depletion 29 - 30handling and storage guidelines 30-4 HEPES in 111-12 human serum albumin (HSA) 34-5 inorganic ion content 45-6 long-term effects of culture conditions 29-30 organic osmolytes (amino acids) 46–9, 62 osmolality see osmolality of culture media pH see pH of culture media

photo-oxidation 105-6 pre-gassing 32-4 pyruvate in 111-12 quality control issues 15-16 record-keeping 30 riboflavin in 105-6, 111-12 selection considerations 30 testing before use 15-16 troubleshooting approach to optimization 28-9 vulnerability of embryos 29 - 30culture media conditioning 34 - 7mineral oil overlay 35-7 protein supplement 34-5 culture systems for IVF long-term monitoring of babies born 38 new directions 38 optimizing media 28-9 troubleshooting flowchart 39 troubleshooting process map 39 cumulative sum analysis (CUSUM plot) 20 CZB (Chatot-Ziomek-Bavister) medium 46 dewars 161-2 liquid nitrogen sterilization 149 - 51diffusion, definition 45 DIPSO buffer 68, 70 DNA damage caused by ROS 111 effects of light exposure 105 repair capacity of embryos and gametes 111 donor cryobanks 168 Dr Kershaw's Cottage Hospital, Oldham, UK 2 Drama Triangle (Karpman) 195 - 8Earle's Balanced Salt Solution (EBSS) 28, 45-6 earthquakes, risk evaluation and management 199–201 EDTA (ethylenediaminetetraacetic acid) 30 Edwards, Robert 2, 8, 45-6, 107 Einstein, Albert 192 electrical connections, safety issues 198-9

embryo fragmentation link with exposure to ROS 111 response to external hypotonic solution 47, 49 embryology team see managing an IVF team embryos culture of surplus embryos 17 DNA repair capacity 111 final disposition of 165-9 heat loss from 82-5 Emden-Meyerhof-Parnas (EMP) pathway 59 emergencies, on-call staff 194 enzyme function, influence of pH 59-60 equipment alarm systems 11-14 calibration certificates 10-11 documentation 10-14 monitoring laboratory equipment 10-14 servicing and maintenance 10 - 14specifications 10 ethical considerations in final disposition of embryos 169 European Committee for Standardization (CEN) 8 - 9European Union Tissues and Cells Directive 2004/23/ EC 8-9, 32 flowcharts symbols 5 troubleshooting tool 5-7 follicular fluid microorganisms in 143 temperature 79-80 Fourier, Joseph 82 fragmentation see embryo fragmentation freezers, controlled-rate freezers 160 fungal contamination in IVF culture 144-5 gamete intrafallopian transfer (GIFT) procedure 107 gas cylinders

regulators and equipment for incubator gases 124

206

Index

gas cylinders (cont.) safety precautions 122-3 storage 122 use of a manifold 124 gases CO₂ measurement methods 118 - 20monitoring gas mixtures 118-22 oxygen measurement methods 118, 121-2 quality and consistency of gas supplies 117-118 troubleshooting flowchart for a gas supply problem 136 troubleshooting process map for a gas supply problem 135 Genba Kaizen quality improvement policy 20 Glasl, Friedrich, conflict escalation model 198 glucose in culture media, effects on embryos 49 glutamine effects of decomposition in media 33 organic osmolyte in culture media 47–9 glutathione (GSH) depletion in culture media 29-30 glutathione peroxidase 29-30 glutathione-5-transferase 29-30 glycine organic osmolyte in culture media 46-9 role in intracellular pH regulation 62 glycine transporter GLYT1 47 - 9Good's Buffers 68-70 Ham's F-10 medium 45-6 hamster embryos, effects of light exposure 106 HBV (hepatitis B virus) handling of infectious material 148-51 infection from body fluids 139 HCV, handling of infectious material 148-51 heated stage 175-6 see also stage warmers Henderson-Hasselbach (H-H) equation 64-7

HEPES buffer 29, 68-70 111-12, 180 photo-oxidation 105-6 HIV handling of infectious material 148-51 infection from body fluids 139 Human Fertilization and Embryology Authority (HFEA) 8-9 cumulative sum analysis (CUSUM plot) 20 Risk Tool 189-90 use of surplus embryos 17 human serum albumin (HSA) 34 - 5humidity in incubators 86 Hurricane Sandy (October 2012) 200 hyaluronan medium 180 hyaluronidase medium 180-2 Hyase[™] medium 180-2 hydrogen peroxide (H₂O₂) generation in culture media 29 - 30production induced by light 105 - 6hypo-osmotic swelling test for sperm viability 47 ICSI (intracytoplasmic sperm injection) media 180 HEPES 180 hyaluronan 180 hyaluronidase 180-2 **MOPS 180** polyvinyl pyrrolidine (PVP) 180 ICSI procedures components of a micromanipulation station 171 dish and pipette preparation 182-3 dish type 181-2 focusing the microscope 179 ICSI procedure troubleshooting tips 184-5 location of the micromanipulation station 171 microdroplet arrangement 181-2 micromanipulation equipment 171-9

micromanipulation preparation 183-4 patient management 185-7 training for 179 troubleshooting flowchart for micromanipulation 187 troubleshooting process map for sperm capture 186 troubleshooting tips 182-7 incubators 85-100 ambient temperature effects 95-6 humidity 86 managing temperature control 86 measuring temperature 96-9 monitoring equipment performance 95-6 practical tips for settings and maintenance 99-100 temperature mapping 87-95 infections in IVF culture antibiotic resistance 141, 145 antibiotics used in culture media 142 Aspergillus infection 144-5 blood-borne diseases 139, 148-51 Chlamydia trachomatis infection 143 cleaning protocols 147-8 fungal contamination 144-5 general principles of infection control 140-5 handling of infectious material 148-51 laboratory hygiene protocols 141-2 liquid nitrogen dewar sterilization 149-51 microbes commonly found on human skin 140-1 microorganisms in follicular fluid 143 microorganisms in semen samples 141-3 mycoplasma infection 143 normal human flora 141-2 potential sources of contamination 139 Schistosoma haematobium infection 145-6 semen as source of infection 141 strategies to minimize infection 145-8



Trichomonas vaginalis infection 146 troubleshooting flowchart 150 troubleshooting process map 149 tuberculosis (TB) 145 vaginal flora 143 viruses 139, 148-51 yeast infection (Candida spp.) 144-5 inorganic ion content of culture media 45-6 International Committee Monitoring Assisted Reproductive Technologies (ICMART) 19 International Electrotechnical Commission (IEC) 8-9 International Organization for Standardization (ISO) accreditation of quality management systems 20 - 6certification of quality management systems 20-6 quality management standards 8-9 ISO 9000:2000 8-9, 22-6 ISO 9001:2008 8-9, 22-6 ISO 14644-1 124 ISO 14971 199 ISO 15189:2012 8-9, 22-6, 32, 70 ISO/IEC 17025:2005 8-9, 22-6 IVF, regulation and standards 8-9 IVF team see managing an IVF team Joint Commission International (JCI) 8-9 Key Performance Indicators (KPIs), monitoring and assessment 18-20 Krebs-Ringer bicarbonate (KRB) solution 45 KSOM (Potassium Simplex Optimized Medium) 28 laminar air flow (LAF) hoods

laser used in micromanipulation 177-9 Law of Cooling (Newton) 82 - 3Law of Three Ts in ART 101 Levy-Jennings Quality Control Charts 18–20 light exposure cell repair mechanisms 105 effects in sea urchin embryos 105 embryo and oocyte repair capacity 111 measurement 105 mitigating damage from light-induced ROS 110-13 photo-oxidation damage 105-6 potential damage to gametes and embryos during ART 104 potentially damaging wavelength range 109-10 production of reactive oxygen species (ROS) 105 - 6sources in the human IVF laboratory 107-10 toxicity in non-primate embryos and gametes 106 - 7toxicity in tissue cultures 106 troubleshooting flowchart 113 troubleshooting process map 112 types of damage caused to cells 105-7 use of filters in the IVF laboratory 107-10 liquid nitrogen dewar sterilization 149-51 liquid nitrogen safety precautions 162-4 cryostore specifications 162-3 safety procedures 163 M16 culture medium 45-6 managing an IVF team 190 - 8communication 195-8 competency assessments 192 - 4conflict resolution 195-8 continuing professional

development (CPD) 193

Drama Triangle (Karpman) 195-8 factors in good teamwork 195 multidisciplinary teamwork 190 on-call staff arrangements 194 out-of-hours access to the IVF lab 194 safe workload 190-2 scheduling of workload 190 - 2staff training 192-4 supervision during training 192 - 4team dynamics 195 troubleshooting process map 201 visitors to the IVF lab 195 manipulators used in micromanipulation 176-7 Matthews, Robert 1 micromanipulation applications in ART 171 focusing the microscope 179 troubleshooting flowchart 187 troubleshooting process map for sperm capture 186 see also ICSI micromanipulation equipment 171-9 heated stage 175-6 see also stage warmers laser 177-9 manipulators 176-7 micropipettes 177 microscope 173-4 optics 174 pipette holders 176 syringes 177 micromanipulation station components 171 location 171 microorganisms commonly found on human skin 140-1 normal human flora 141-2 range found in semen 142-3 vaginal flora 143 see also infections in IVF culture micropipettes 177 microscope for micromanipulation 173-4 focusing 179 optics 174

127-9, 145

large offspring syndrome (LOS)

in cattle and sheep 29

208

Index

mitochondrial function,

influence of pH 59-60 MOPS buffer 68, 70, 180 mouse embryo assays (MEA) 15-16, 30 mouse embryos, effects of light exposure 107 Murphy's Law in ART 1-7, 189 mycoplasma infection 143 natural disasters, crisis management planning 198-201 Newton, Isaac, Law of Cooling 82-3 nuclear organizing region (NOR) 47 nucleocytoplasmic ratio 47, 49 oils used as culture media overlays 35-7 effects of light exposure 112 see also culture dish preparation on-call staff arrangements 194 oocytes DNA repair capacity 111 removal of cumulus cells (denudation) 180-2 organic osmolytes (amino acids) in culture media 46-9, 62 role in intracellular pH regulation 62 osmolality and culture dish preparation 49-55 definition 44-5 distinction from osmolarity 44 - 5measurement 45 of tubal fluid 46-7 of uterine secretions 46-7 osmolality of culture media cell volume regulation 45-9 cryopreservation process 55 culture media formulations 45 - 7effects of glucose in culture media 49 effects of organic osmolytes (amino acids) 46-9 effects on cell cytoplasm 45 embryo fragmentation in low osmolality 47, 49 measurement 55-6

osmolality range of commercial media 49 sensitivity of gametes and embryos 45, 57 troubleshooting flowchart 56 troubleshooting process map 55 osmolarity, definition 44-5 osmometers 45, 55-6 osmosis, definition 45 osmotic pressure definition 45 methods of measurement 44-5 out-of-hours access to the IVF lab 194 oxygen measurement methods 118, 121-2 Pasteur effect 60 Payne, James 1 Peck, M. Scott 2 pН influence on enzyme function 59-60 influence on mitochondrial function 59-60 origin of the term 64 pH measurement 70-5 devices used to monitor pH in culture media 73-5 pH of culture media buffers 68-70 culture media pH equilibration 68 Henderson-Hasselbach (H-H) equation 64-7 influence of altitude 66-7 influence on embryo development 62 optimal pH for in vitro gametes and embryos 63-4 physico-chemical regulation 64-70 quality control indicator 75 role of amino acids (organic osmolytes) 62 troubleshooting flowchart 74 troubleshooting process map 73 zwitterions 62, 68-70 pH regulation and ATP production in the embryo 59-60 in gametes and embryos 59-64

influence on embryo development 59-60 intracellular pH homeostasis 59-64 troubleshooting flowchart 74 troubleshooting process map 73 phosphofructokinase (PFK) 59 photo-oxidation damage 105-6 pipette holders used in micromanipulation 176 plasticware bisphenol A (BPA) in 32 quality control 15 VOCs in 32 polyvinyl pyrrolidine (PVP) medium 180 power supply contingencies for power loss 198 safety of electrical connections 198-9 troubleshooting during power loss 199 uninterruptable power supply (UPS) units 198 pre-gassing of media 32-4 prion diseases 139 problem-solving approach in ART 2-7 proline, organic osmolyte in culture media 47-9 Purdy, Jean 2, 8, 45-6 pyruvate, protective effects in culture media 111-12 Quality Control (QC) importance in ART 8-9 role of pH measurement 75 Quality Management System (QMS) 8-9 accreditation 20-6 audit visits 23-6 bioassays 15–16 certification 20-6 consumable products and supplies 14-15 corrective action 23-4 culture media testing before use 15-16 elements of 9-20 equipment monitoring 10 - 14equipment servicing and maintenance 10-14 equipment specifications 10

		Index	209
flowchart 25	standard anarsting	aumlus ambruos, cultur	ro of 17
key performance indicators	standard operating procedures (SOPs) 189–90	surplus embryos, cultur syringes used in	

(KPIs) monitoring and assessment 18-20 non-compliances 23-6 preventive action 23-4 process map 24 record keeping 9-10 risk management 189-90 standard operating procedures (SOPs) 10 study protocols 20-1 rabbit embryos, effects of light exposure 106 radiation, definition 81 reactive oxygen species (ROS) embryo response to excess glucose 49 generation in media and buffers 29-30 mitigating damage from 110-13 production induced by light 105 - 6reducing exposure of gametes and embryos 110-13 record-keeping in ART 5 embryo culture records 30 importance of 9-10 religious considerations in final disposition of embryos 169 riboflavin in culture media, photo-oxidation 105-6, 111 - 12risk management contingencies for loss of utilities 198 crisis management 198 definition of "safety" 199 earthquake risk 199–201 emergency planning 199-201 evaluation of risk 199-201 HFEA Risk Tool 189-90 managing an IVF team 190 - 8Murphy's Law 1-7, 189 natural disasters 198-201 Quality Management System 189-90 safety of electrical

connections 198–9 sources of risk in ART 189

procedures (SOPs) 189–90 troubleshooting during power loss 199 troubleshooting flowchart 202 troubleshooting process map 201 risk matrix 201 Saccharomyces cerevisiae (yeast), metabolic pathways 59-60 safety, definition in terms of risk 199 Schistosoma haematobium infection 145-6 sea urchin embryos, effects of light exposure 105 semen as source of infection 141 - 3semen assessment, temperature for 80 semen samples, range of microorganisms found in 142 - 3Serum Substitute Supplement (SSS) 35 simplex optimization method 28 single or multipronucleate embryo culture 17 Sørensen, Søren Peder Lauritz 64 sperm, lack of DNA repair capacity 111 sperm assessment, computerassisted (CASA) 80 Sperm Viability Index (SVI) 17 sperm viability test 17 hypo-osmotic swelling test 47 staff see managing an IVF team staff training 192-4 stage warmers, temperature control 87, 89-99 see also heated stage standard operating procedures (SOPs) 10, 189-90 study protocols 20-1 standards and quality control in ART 8-9 statistical process control (SPC) 20 Steptoe, Patrick 2, 8 study protocols 20-1

micromanipulation 177 taurine 49 organic osmolyte in culture media 46-9 role in intracellular pH regulation 62 teamwork see managing an IVF team temperature control in IVF ambient temperature effects 95 - 6equipment settings and maintenance 99-100 measuring temperature 96-9 monitoring equipment performance 95-6 temperature sensors 96-9 thermometers 96-9 temperature control in IVF culture detrimental effects of fluctuation 79-80 follicular fluid temperature 79-80 heat loss from an embryo 82 - 5incubators 85-100 Law of Three Ts in ART 101 Newton's Law of Cooling 82 - 3optimal temperature range 79 - 80principles of thermal energy 80 - 5stage warmers 87, 89-99 temperature mapping of equipment 87-95 thermal coins 83-5, 90 troubleshooting flowchart 101 troubleshooting process map 100 temperature for semen assessment 80 temperature gradient, and heat transfer 82 temperature sensors 96-9 thermal coins 83-5, 90 thermal energy, principles of 80-5 thermometers 96-9

210	
210	

Index

tricarboxylic acid (TCA) cycle 59-60 Trichomonas vaginalis infection 146 troubleshooting approach in ART 2-7 definition 4-5 early development of ART 2 general flowchart for ART 6 guide 4 process map 4 use of flowcharts 5-7 tryptophan in media, photo-oxidation 106 tubal fluid osmolality 46-7 tuberculosis (TB) 145 Tyrode's solution 45-6 tyrosine in media, photo-oxidation 106

uninterruptable power supply (UPS) units 198 uterine secretions, osmolality 46 - 7vaginal flora 143 viruses blood-borne infections 148-51 infection from body fluids 139 visitors to the IVF lab 195 vitrification (non-equilibrium/ ultra-rapid freezing) 156-8 troubleshooting process map 166 VOCs (volatile organic compounds) adverse effects in an ART setting 129 awareness of potential toxicity in ART 137-8

detection and measurement 131–4 elimination 134–6 from cleaning products 147–8 in plasticware 15, 32 in the IVF laboratory 129–31 sources in the environment 129–31 troubleshooting flowchart for a potential VOC problem 137

Westgard Rules 19 Whitten's medium 45 workload, safety issues 190-2

yeasts

Candida spp. infection 144–5 Saccharomyces cerevisiae metabolic pathways 59–60

zwitterions 62, 68-70