

Index

A

Accidental cell death, trauma, 1–2
Acridine orange
 Caenorhabditis elegans apoptosis studies, 414–415, 423
 Drosophila apoptosis assay
 applications, 346
 embryo staining, 345
 larva tissue staining, 345–346
 materials, 344–345
 overview, 344
 recipes, 347
 troubleshooting, 346
Actinomycin D, apoptosis induction assay
 in hematopoietic cells
 incubation conditions, 31
 materials, 30–32
 substitute agents for apoptosis induction, 31–32
Annexin V and propidium iodide uptake assay
 applications, 78, 260, 263
 controls, 260
 distinguishing apoptosis from necrosis, 75, 258
 flow chart, 76
 flow cytometry, 76–77
 fluorescence-activated cell sorting, 260–262
 materials, 75, 258–259
 recipes, 79
 staining, 76, 259
 time-lapse imaging
 applications, 270–271
 cell preparation, 265, 267–268
 data analysis, 269
 materials, 264–265
 microscopy, 266–269
 overview, 264
 troubleshooting, 269–270
 troubleshooting, 77–78
APAF-1. *See* Apoptotic protease activating factor-1
APO-1/Fas. *See* Death-inducing signaling complex
Apoptosis
 actinomycin D induction. *See* Actinomycin D
 Bcl-2 proteins. *See* Bak; Bax
 Caenorhabditis studies. *See* *Caenorhabditis elegans* apoptosis
 caspase-activating complexes. *See* Apoptosome; Death-inducing signaling complex
 caspase cleavage. *See* Caspases
 cytochrome *c* release assay. *See* Cytochrome *c* release assay
 distinguishing from other cell death modalities. *See also* Annexin V and propidium iodide uptake assay
 cell lines for study, 252
 crystal violet viability assay
 cell preparation, 255
 materials, 254
 overview, 254
 recipes, 256
 staining and measurement, 256

troubleshooting, 256
overview, 23–27
riptosome analysis by caspase-8 immunoprecipitation
 applications, 278–279
 immunoprecipitation, 275–276
 materials, 272–273
 overview, 272
 protein sample preparation, 273–274
 recipes, 279
 troubleshooting, 278
 western blot, 276
 work flow, 274
strategies, 250–252
DNA content assay by cell cycle stage.
 See Propidium iodide
Drosophila studies. *See* *Drosophila* apoptosis
extrinsic, 2–3
Hoechst 33342 nuclear staining. *See* Hoechst 33342
inducers, 21–22
intrinsic, 2
mammalian in vivo measurements
 immunohistochemistry
 B-cell lymphoma transplantation in mice and apoptosis induction, 325–326
 fragmented DNA detection, 327–328
 materials, 324–325
 overview, 324
 recipes, 328–329
 troubleshooting, 328
 tumor tissue collection and fixation, 326–327
 overview, 313–315
 positron emission tomography of hematological tumor response to drugs
 applications, 318
 B-cell lymphoma transplantation in mice, 317
 data analysis, 318
 fluorodeoxyglucose administration and uptake measurement, 317–318
 materials, 316–317
 overview, 316
 recipes, 319
 positron emission tomography of solid tumor response to drugs
 applications, 323
 data analysis, 323
 fluorodeoxyglucose administration and uptake measurement, 322
 materials, 320–321
 overview, 320
 tumor cell transplantation in mice, 321–322
 propidium iodide staining
 B-cell lymphoma transplantation in mice and apoptosis induction, 331–332

cell lysis and staining, 332
flow cytometry, 333
materials, 330–331
overview, 330
recipes, 333–334
tumor tissue collection and cell suspension preparation, 332
mitochondrial transmembrane potential assay. *See* Mitochondrial transmembrane potential assay
overview, 2
TUNEL assay. *See* TUNEL assay
Apoptosome
 activation in vitro, 183
 assembly and analysis in vitro
 applications, 104–105
 cell lysate preparation, 100
 dATP activation of lysates, 100–102
 fluorimetric analysis of caspase activity, 102–103
 gel filtration, 102
 materials, 98–99
 overview, 98, 101
 procaspase preparation for bioassay, 103–104
 recipes, 105–106
 SDS-PAGE and western blot, 103
 troubleshooting, 104
 overview, 95–96
Apoptotic protease activating factor-1 (APAF-1), 142, 181
Autophagy-dependent cell death
 assays. *See* *Caenorhabditis elegans* autophagy; *Drosophila* autophagy; Mammalian cell autophagy
 distinguishing from other cell death modalities, 25
 inhibitors and inducers, 281
 LC3. *See* MAP1LC3B
 overview, 5, 279–280
 types, 279
B
Bak
 activation assay overview, 116
 blue native PAGE and antibody gel shift assay of conformational change and oligomerization
 applications, 138
 cell permeabilization, 136
 gel electrophoresis, 137
 materials, 135–136
 overview, 135
 recipes, 138–139
 troubleshooting, 138
 conformational change in activation
 flow cytometry
 applications, 129
 cell harvesting, 128
 conformation-specific antibodies, 128
 flow cytometry, 128
 materials, 127–128

Index

- Bak (*Continued*)
overview, 127
recipes, 129
troubleshooting, 128–129
immunoprecipitation analysis
cell harvesting, 123
cell lysis, 124
detergents, 125
materials, 122–123
overview, 122
protein G Sepharose bead incubation,
123–124
recipes, 125
troubleshooting, 124
western blot, 124
oligomerization assay during apoptosis
chemical cross-linking, 132
disulfide linking, 132
materials, 131–132
overview, 131
recipes, 133–134
western blot, 133
overview, 115–116
Bax
activation assay overview, 116
blue native PAGE and antibody gel shift
assay of conformational change
and oligomerization
applications, 138
cell permeabilization, 136
gel electrophoresis, 137
materials, 135–136
overview, 135
recipes, 138–139
troubleshooting, 138
conformational change in activation
flow cytometry
applications, 129
cell harvesting, 128
conformation-specific antibodies, 128
flow cytometry, 128
materials, 127–128
overview, 127
recipes, 129
troubleshooting, 128–129
immunoprecipitation analysis
cell harvesting, 123
cell lysis, 124
detergents, 125
materials, 122–123
overview, 122
protein G Sepharose bead incubation,
123–124
recipes, 125
troubleshooting, 124
western blot, 124
oligomerization assay during apoptosis
chemical cross-linking, 132
disulfide linking, 132
materials, 131–132
overview, 131
recipes, 133–134
western blot, 133
overview, 115–116
subcellular localization and membrane
integration analysis
apoptosis induction, 118
applications, 120
carbonate extraction, 119–120
materials, 117–118
overview, 117
permeabilization of cells, 118–119
recipes, 120–121
troubleshooting, 120
western blot, 120
Bcl-2 proteins. *See* Bak; Bax
 β -N-Acetyl glucosaminidase (NAG)
cytosolic cathepsin and β -N-acetyl
glucosaminidase activity assays
applications, 214
data analysis, 213
digitonin extraction, 210–211
flow chart, 212
materials, 209–210
overview, 209
plate assays, 211–213
recipes, 214–215
troubleshooting, 213–214
lysosomal membrane permeabilization assay
using cytosolic activity, 206–207
Bimolecular fluorescence complementation.
See Caspases
Biological safety. *See* Safety
Blots, cell death research publications
interpretation of images, 19
preparation of images, 15–16
Bovine serum albumin–gold. *See* Lysosomal
membrane permeabilization assay
C
Caenorhabditis elegans apoptosis
core pathway, 394
genetic analysis, 396
germline apoptosis
physiological apoptosis, 394–395
stress-induced apoptosis, 395
induction
genotoxic agent induction, 401, 404
irradiation induction, 402
materials, 400–401
microscopy, 402
overview, 400
recipes, 405
RNA interference, 401, 403
troubleshooting, 402–403
fluorescence microscopy
acridine orange staining, 414–415
applications, 418–419
live time-course studies using
fluorescent proteins, 415
materials, 413–414
overview, 413
quantitative analysis, 419
recipes, 419–420
RNA interference low-throughput
screening, 416–417
troubleshooting, 417–418
RNA interference high-throughput
screening
applications, 424–425
data analysis, 425–426
materials, 421–422
microscopy, 423–424
overview, 421
plate preparation, 422–423
recipes, 426
staining, 423
troubleshooting, 424
immunostaining for markers
acetone blocking powder
preparation, 431–432
applications, 432–433
gonad dissection and staining,
430–431
materials, 428–429
overview, 428
recipes, 54
troubleshooting, 432
light microscopy visualization
applications, 411–412
data analysis, 412
embryos, 408
germ cell apoptosis, 408–409, 411
image analysis, 409
materials, 407–408
overview, 407
recipes, 412
slide preparation, 408
troubleshooting, 410–411
overview of techniques for study, 396–398
somatic apoptosis, 393–394, 404
Caenorhabditis elegans autophagy
apoptosis, necrosis, and cell clearance, 441
dauer development, 440
fluorescent reporters, 442
genes, 437–439
green fluorescent protein–LGG-1 for
detection
alternative markers, 451
applications, 451
data acquisition, 450–451
fluorescence microscopy, 449
larva preparation, 448–449
materials, 447–448
overview, 447
recipes, 452
reporter development, 449–450
troubleshooting, 449
L1 arrest after starvation, 440
longevity pathways, 440–441
overview
apoptosis, 437–438
assays, 441–443
P granule degradation marker detection
of embryo autophagy
applications, 457
data analysis, 456–457
embryo mounting and microscopy, 455
markers
PGL-1, 456
SEPA-1, 456
SQST-1, 456
materials, 454–455
overview, 454
recipes, 457–458
troubleshooting, 455
paternal mitochondria degradation, 441
RNA interference-mediated inactivation
of autophagy genes
applications, 473–474
data analysis, 472–473
flow chart, 469
materials, 468–469
microscopy, 470–471
overview, 468, 471–472
plate preparation and incubation, 469–470
recipes, 474
troubleshooting, 470
western blot of LGG-1
applications, 464–465
flow chart, 460
gel electrophoresis, 461–462
immunoblotting, 462
marker establishment and characteristics,
463–464
materials, 459–460

- Caenorhabditis elegans* autophagy (*Continued*)
 - membrane transfer, 462
 - overview, 459
 - recipes, 465–466
 - sample preparation, 461
- Caspases
 - activating complexes. *See* Apoptosome; Death-inducing signaling complex
 - activation pathways, 142–143, 173–174
 - activity assays
 - overview, 178–179
 - commercial kits, 179
 - cytochrome *c* activation assay
 - apoptosome activation, 183
 - applications, 184
 - cytosolic extract preparation, 182–183
 - materials, 181–182
 - recipes, 184–185
 - troubleshooting, 183
 - fluorescence well plate assay
 - applications, 188–189
 - incubation conditions, 187
 - materials, 186–187
 - overview, 186
 - recipes, 189
 - troubleshooting, 188
 - antibody-based techniques
 - applications, 193
 - cell lysis, 192
 - cell treatment and harvesting, 191–192
 - flow chart, 191
 - gel electrophoresis, 192
 - materials, 190–191
 - overview, 190
 - recipes, 194
 - troubleshooting, 192–193
 - inhibitor binding to active caspases
 - applications, 198–199
 - cell incubation and lysis, 197
 - flow chart, 197
 - materials, 196–197
 - overview, 196
 - recipes, 199
 - troubleshooting, 198
 - western blot, 198
 - substrate verification
 - applications, 203
 - flow chart, 202
 - gel electrophoresis, 202
 - materials, 201–202
 - overview, 201
 - recipes, 203
 - troubleshooting, 202–203
- caspase-3 cleavage detection in apoptotic cells
 - antibody labeling, 81
 - applications, 82
 - cell preparation, 81
 - flow cytometry, 82
 - materials, 80–81
 - recipes, 83
- caspase-8 assay. *See* Death-inducing signaling complex
- caspase-Glo 3/7 assay for high-throughput screening, 493, 495–496
- cell death pathway overlap, 7–8
- Drosophila*
 - activity assays
 - fluorimetric assays, 358
 - lysate preparation, 358
 - materials, 357
 - overview, 357
 - recipes, 359
 - substrates, 359
- caspase-3 immunostaining
 - fixation and staining, 349
 - fluorescence microscopy, 349–350
 - materials, 348–349
 - overview, 348
 - recipes, 351
 - specificity, 350
 - troubleshooting, 350
- caspase types, 337
- functions outside of cell death, 3–4
- inhibitors, 177
- overview of assays, 26–27
- riptide analysis by caspase-8 immunoprecipitation
 - applications, 278–279
 - immunoprecipitation, 275–276
 - materials, 272–273
 - overview, 272
 - protein sample preparation, 273–274
 - recipes, 279
 - troubleshooting, 278
 - western blot, 276
 - work flow, 274
- single-cell imaging
 - advantages, 142, 144
 - bimolecular fluorescence
 - complementation
 - applications, 157–158
 - materials, 149–151
 - overview, 144–145, 149, 156–157
 - recipes, 158
 - single time point analysis of caspase activation, 152–154
 - time-lapse microscopy, 154–155
 - transfection with BiFC plasmid pairs, 151–152
 - troubleshooting, 156
 - fluorescence resonance energy transfer
 - applications, 168–170
 - cell number, 168
 - cell preparation, 161–162
 - controls, 162–163, 168
 - flow chart, 161
 - image processing and data analysis, 164–167
 - materials, 159–160
 - overview, 145–146, 159
 - recipes, 170
 - routine measurements, 163
 - sensors, 160
 - troubleshooting, 167–168
 - fluorescently labeled inhibitor of caspases, 147
 - fluorogenic substrates, 146–147
 - overview, 141
 - rationale, 141–142
 - substrates and cleavage motifs, 174–177
- Cathepsins
 - cell death role, 178
 - lysosomal membrane permeabilization assay
 - cysteine cathepsin assay with fluorogenic substrate
 - advantages, 240
 - cell preparation, 237
 - controls, 239–240
 - data analysis, 238–239
 - incubation conditions, 237, 239
 - lactate dehydrogenase assay, 239
 - materials, 236–237
 - overview, 226–227, 236
 - recipes, 241
- cytosolic cathepsin and β -*N*-acetylglucosaminidase activity assays
 - applications, 214
 - data analysis, 213
 - digitonin extraction, 210–211
 - flow chart, 212
 - materials, 209–210
 - overview, 209
 - plate assays, 211–213
 - recipes, 214–215
 - troubleshooting, 213–214
- cytosolic enzyme activity assay overview, 206–207
- CD95. *See* Death-inducing signaling complex
- Colony-forming assay, cell survival quantification
 - adherent cells
 - applications, 50
 - cell plating, 48–49
 - fixation and staining, 49
 - materials, 47–48
 - overview, 47
 - recipes, 50
 - hematopoietic cells on soft agar
 - applications, 53
 - cell plating, 52–53
 - colony counting, 53
 - materials, 51–52
 - overview, 51
 - recipes, 53–54
 - overview, 23
- CRISPR, cell death applications, 8
- Crystal violet viability assay
 - cell preparation, 255
 - materials, 254
 - overview, 254
 - recipes, 256
 - staining and measurement, 256
 - troubleshooting, 256
- Cytochrome *c*
 - caspase activation assay
 - apoptosome activation, 183
 - applications, 184
 - cytosolic extract preparation, 182–183
 - materials, 181–182
 - recipes, 184–185
 - troubleshooting, 183
 - release assay
 - applications, 88
 - flow chart, 86
 - fluorescence microscopy, 87–88
 - incubation conditions, 86–87
 - materials, 85
 - overview, 27, 85
 - recipes, 88
- Cytospinning with rapid staining, morphological analysis of cell death
 - applications, 58
 - cell harvesting, 56–57
 - cytospinning, 56
 - hematoxylin and eosin staining, 57
 - materials, 55–56
 - overview, 55
 - recipes, 58
- D**
- Data normalization, guidelines for publication, 16–17
- Death, defining for a cell, 6, 11–12

Index

- Death-inducing signaling complex (DISC)
activation, isolation, and analysis
 applications, 112
 caspase-8 assay, 111–112
 cell lysis and treatment, 110–111
 flow chart for affinity purification, 109
 immunoprecipitation, 111
 materials, 107–108
 overview, 107
 recipes, 112–113
 SDS-PAGE and western blot, 112
 troubleshooting, 112
overview, 95–96
- DIAP1, 336–337
- DISC. *See* Death-inducing signaling complex
- DNA fragmentation, overview of assays, 25–26
- Drosophila* apoptosis
acridine orange assay of dying cells
 applications, 346
 embryo staining, 345
 larva tissue staining, 345–346
 materials, 344–345
 overview, 344
 recipes, 347
 troubleshooting, 346
- caspase activity assays
 fluorimetric assays, 358
 lysate preparation, 358
 materials, 357
 overview, 357
 recipes, 359
 substrates, 359
- caspase-3 immunostaining
 fixation and staining, 349
 fluorescence microscopy, 349–350
 materials, 348–349
 overview, 348
 recipes, 351
 specificity, 350
 troubleshooting, 350
- caspase types, 337
- development and metamorphosis, 335–336
- machinery, 336–338
- overview of assays, 338
- RNA interference studies of death stimuli
 cell culture, 354
 cell lines, 355–356
 double-stranded RNA
 cell treatment, 354–355
 synthesis, 353–354
 materials, 352–353
 overview, 352
 quantitative polymerase chain reaction,
 355
 recipes, 356
 total RNA preparation, 355
 viability assays, 354
- TUNEL assay
 applications, 342
 fluorescence microscopy, 341–342
 incubation conditions, 341
 materials, 340–341
 overview, 340
 recipes, 343
 troubleshooting, 342
- Drosophila* autophagy
 functions, 361
 LysoTracker staining
 applications, 369
 autophagy induction by amino acid
 starvation, 365
 cultured cells, 365–366
- flow chart, 366
 flow cytometry, 366–367
 fluorescence microscopy, 368
 materials, 364–365
 ovaries, 367–368
 overview, 368
 recipes, 369–370
 staining, 366
 troubleshooting, 368–369
- ovary, 362
- Ref(2)P assays in ovaries
 applications, 377–378
 flow chart, 375
 immunofluorescence microscopy,
 374
 materials, 372–373
 ovary preparation, 374
 overview, 372
 recipes, 378–379
 troubleshooting, 376–377
 western blot, 376
- E**
- Error bars, interpretation in cell death research
 publications, 19
- F**
- Fas ligand, apoptosis induction, 21
- Figures, cell death research publications
 blots and gels
 interpretation of images, 19
 preparation of images, 15–16
 graphs
 interpretation, 10–11
 preparation, 14–15
 image quality, 13
 microscopy images, 16
 statistics, 12–13
- Flow cytometry. *See* Annexin V and propidium
 iodide uptake assay; Bak; Bax;
 Caspases; *Drosophila* autophagy;
 Lysosomal membrane permeabi-
 lization; Mammalian cell
 autophagy assays; Mitochondrial
 transmembrane potential assay;
 Propidium iodide
- Fluorescence resonance energy transfer.
 See Caspases
- Fluorodeoxyglucose. *See* Positron emission
 tomography
- G**
- Gel filtration, apoptosome, 102
- Gels, cell death research publications
 interpretation of images, 19
 preparation of images, 15–16
- Granzyme B, cell death role, 3, 178
- Graphs, cell death research publications
 interpretation, 10–11
 preparation, 14–15
- H**
- Hazardous chemicals. *See* Safety
- Hematoxylin and eosin staining. *See*
 Cytospinning with rapid staining
- High-throughput screening (HTS)
 bioinformatics, 495–496
 cell death assay applications, 477–478
 design of assay
- cell density, 483
 controls, 483–484
 data density maximization, 482
 drug action, 482
 dynamic range, 498
 initial considerations, 478–479
 limitations of assay, 484–485
 live versus fixed analysis, 482–483
 model system, 481–482
 reproducibility, 485
 timing of effect, 484
 validation, 485
 work flow, 484
- drug development, 8–9
- modes
 cell suspensions versus adherent cells,
 481
 high-content imaging mode, 479–480
 plate reader mode, 480–481
- RNA interference screening. *See also*
 Caenorhabditis elegans apoptosis
 caspase-Glo 3/7 assay, 492, 495–496
 cell counting with nuclear staining,
 493–494
 CellTiter-Fluor assay, 492, 495–496
 data analysis, 494–496
 high-content microscopy, 498–499
 materials, 488–490
 multiplex assay validation, 499–500
 overview, 488
 reverse transfection and treatment,
 490–492
 troubleshooting, 496–498
- Hoechst 33342, nuclear staining
 applications, 62
 fluorescence microscopy, 62
 materials, 60–61
 overview, 60
 recipes, 63
 staining, 61–62
 troubleshooting, 62
- HTS. *See* High-throughput screening
- I**
- Images, publication guidelines, 13, 15–16
- L**
- Laboratory safety. *See* Safety
- LGG-1. *See* *Caenorhabditis elegans* autophagy
- Life, defining for a cell, 7, 11–12
- LMP. *See* Lysosomal membrane
 permeabilization
- Lysosomal membrane permeabilization (LMP)
 assays
 bovine serum albumin–gold release assay
 advantages, 244
 data analysis, 244
 fixation and microscopy, 243–244
 materials, 242–243
 overview, 226–227, 242
 recipes, 246
 tracer loading, 243
 cysteine cathepsin assay with fluorogenic
 substrate
 advantages, 240
 cell preparation, 237
 controls, 239–240
 data analysis, 238–239
 incubation conditions, 237, 239
 lactate dehydrogenase assay, 239

Index

- p62 (*Continued*)
genetic manipulation in ovary
 applications, 390–391
 autophagy induction and analysis, 389–390
germline clone generation, 387–388
germline expression of interfering RNA, 388–389
materials, 386–387
overview, 386
recipes, 391–392
somatic clone generation, 388
troubleshooting, 390
immunohistochemistry on paraffin sections
 for mammalian cell autophagy detection
 antibody and peptide preparation, 304–305
 antigen retrieval, 305
 applications, 306–307
 deparaffinization and rehydration, 305
 hematoxylin counterstaining, 306
 materials, 303–304
 mounting and imaging, 306
 overview, 303
 staining, 305–306
 troubleshooting, 306
Ref(2)P assays in *Drosophila* ovaries
 applications, 377–378
 flow chart, 375
 immunofluorescence microscopy, 374
 materials, 372–373
 ovary preparation, 374
 overview, 372
 recipes, 378–379
 troubleshooting, 376–377
 western blot, 376
Perforin, cell death induction, 3
PET. *See* Positron emission tomography
Phosphatidylserine (PS)
 annexin V binding. *See* Annexin V and propidium iodide uptake assay
 overview of assays, 26
Phospho-MLKL antibody, necroptosis marker, 8
PI. *See* Propidium iodide
PIDosome, 142, 174
Positron emission tomography (PET)
 hematological tumor apoptosis response to drugs
 applications, 318
 B-cell lymphoma transplantation in mice, 317
 data analysis, 318
 fluorodeoxyglucose administration and uptake measurement, 317–318
 materials, 316–317
 overview, 316
 recipes, 319
 solid tumor apoptosis response to drugs
 applications, 323
 data analysis, 323
 fluorodeoxyglucose administration and uptake measurement, 322
 materials, 320–321
 overview, 320
 tumor cell transplantation in mice, 321–322
Propidium iodide (PI)
 annexin V and propidium iodide uptake assay. *See* Annexin V and propidium iodide uptake assay
apoptotic cell DNA content assay by cell cycle stage
 applications, 73
 cell preparation, 70
 flow chart, 71
 flow cytometry, 71–73
 materials, 69–70
 overview, 69
 recipes, 73
 staining, 70–71
cell death quantification
 advantages and limitations, 45
 cell harvesting, 43
 data analysis, 45
 flow cytometry, 44–45
 materials, 42–43
 propidium iodide treatment, 43
 recipes, 46
marker use for living and dead cells, 22, 42
tumor apoptosis assay in mice
 B-cell lymphoma transplantation in mice and apoptosis induction, 331–332
 cell lysis and staining, 332
 flow cytometry, 333
 materials, 330–331
 overview, 330
 recipes, 333–334
 tumor tissue collection and cell suspension preparation, 332
PS. *See* Phosphatidylserine
Pyroptosis
 marker development, 8
 overview, 4
R
Radiation safety. *See* Safety
RAIDD, 142, 145, 157, 174
Rapi-Diff staining. *See* Cytospinning with rapid staining
Reactive oxygen species. *See* Mammalian cell autophagy
Receptor-interacting protein kinase (RIPK), 177, 250–252, 272, 278–279
Ref(2)P. *See* p62
Replicates, guidelines for publication, 17
RIPK. *See* Receptor-interacting protein kinase
Riptosome
 analysis by caspase-8 immunoprecipitation
 applications, 278–279
 immunoprecipitation, 275–276
 materials, 272–273
 overview, 272
 protein sample preparation, 273–274
 recipes, 279
 troubleshooting, 278
 western blot, 276
 work flow, 274
 necrosis role, 250–252
RNA interference. *See* *Caenorhabditis elegans* apoptosis; *Drosophila* apoptosis; *Drosophila* autophagy; High-throughput screening
S
Safety
 biological safety, 506–507
 general cautions, 503–505
 hazardous chemical general properties, 507–508
radiation safety, 506
resources, 503
waste disposal, 505
Sequestosome-1. *See* p62
Single-cell imaging. *See* Caspases
Statistical analysis, cell death research, 12–13
T
Tetramethylrhodamine ethyl ester. *See* Mitochondrial transmembrane potential assay
TRAIL, apoptosis induction, 21–22, 95–96
Trypan blue
 assay for cell death quantification
 calculations, 40
 cell harvesting, 39
 flow chart, 39
 hemocytometer counting, 39–40
 materials, 38
 recipes, 41
 trypan blue treatment, 39
Drosophila cell viability assay, 354
uptake in dead cells, 22, 38
TUNEL assay
 acridine orange assay of dying cells
 applications, 346
 embryo staining, 345
 larva tissue staining, 345–346
 materials, 344–345
 overview, 344
 recipes, 347
 troubleshooting, 346
 applications, 66
 B-cell lymphoma apoptosis detection after transplantation in mice, 327–328
Drosophila apoptosis
 applications, 342
 fluorescence microscopy, 341–342
 incubation conditions, 341
 materials, 340–341
 overview, 340
 recipes, 343
 troubleshooting, 342
 flow chart, 65
 fluorescence microscopy, 66
 incubation conditions, 65–66
 materials, 64–65
 overview, 64
 recipes, 67
U
UAS-GAL4. *See* *Drosophila* autophagy
Ultraviolet radiation-induced cell death
 adherent cell culture, 35
 Caenorhabditis elegans apoptosis, 402
 materials, 34–35
 overview, 34, 36
 radiation exposure, 35–36
 recipes, 37
W
Western blot. *See* Apoptosome; Bak; Bax; Blots; *Caenorhabditis elegans* autophagy; Caspases; Death-inducing signaling complex; *Drosophila* autophagy; Mammalian cell autophagy; Riptosome