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Titolo	Subcellular fractionation [[electronic resource]] : a practical approach / / edited by J.M. Graham and D. Rickwood
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Descrizione fisica	1 online resource (360 p.)
Collana	The practical approach series
Altri autori (Persone)	Graham J. M <1943-> (John M.) Rickwood D (David)
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Nota di contenuto	Cover; Contents; List of contributors; Abbreviations; 1. Homogenization of tissues and cells; 1. Introduction; 2. Aims of the homogenization procedure; 3. Influence of sample type; 4. Homogenization media; 5. Methods of homogenization; Type 1 homogenizers; Type 2 homogenizers; 6. Homogenization of tissues and cells; Mammalian liver; Brain; Muscle; Mammalian tissue culture cells; Plant organelles; Yeast; Other fungi and algae; Trypanosomes; Bacteria; References; 2. Isolation of subcellular fractions; 1. Introduction; 2. Composition of a tissue homogenate; 3. Properties of cell organelles Factors affecting organelle density and size 4. Centrifugal methods for the separation of organelles; Separation by size; Separation by density; Density perturbation; 5. Non-centrifugal procedures; Immunoisolation; Separation by electrophoresis; 6. Identification of separated material; Marker enzymes; Introduced markers for endocytic and exocytic pathways; Characteristic non-enzymatic proteins; 7. Assessment of the purity of fractions; Purity and purification; Problems from cell heterogeneity within tissues; Problems arising from organelle fragmentation Missorting in the exocytic and endocytic pathways 8. Fractionation problems; No separation; Aggregation following resuspension of a fraction; Poor recovery of markers; Damage to cell structures; 9. A

systematic approach to cell fractionation; Preliminary studies; Determination of the properties of components of the homogenate; Method development; Simplification of the separation; References; 3. Isolation and characterization of nuclei and nuclear subfractions; 1. Introduction; 2. Methods of preparing purified nuclei; Types of cells and tissue samples; Homogenization media Homogenization methodsCentrifugation conditions; Assays of nuclear purity; 3. Methods for purifying metaphase chromosomes; 4. Isolation of nuclear subfractions; Preparation of nucleoli; Preparation of nuclear membranes; Isolation of nuclear matrix; Preparation of nucleoids; 5. Isolation of nucleoprotein complexes; Isolation of polynucleosomes of chromatin; Ribonucleoproteins; 6. Isolation of nuclear macromolecules; Isolation of nuclear proteins; Isolation of nuclear RNA; Isolation of DNA; 7. Functional assays of nuclei; Analysis of DNA-binding proteins; Transcription assays; References

4. Subcellular fractionation of mitochondria1. Introduction; 2. Purification of mitochondria from various eukaryotic sources; Introduction; Protocols for purification of mitochondria from several eukaryotic sources; Further purification of mitochondrial fractions; 3. Determination of mitochondrial purity; Introduction; Use of the oxygen electrode to determine mitochondrial integrity; Determination of the integrity of the mitochondrial outer membrane; Glucose hexokinase trap method for estimation of P:O ratios; 4. Subfractionation of mitochondria
Preparation of submitochondrial particles by sonication

Sommario/riassunto

Many investigations into the structure and function of cells and tissues require the isolation of a particular membrane or subcellular component (organelle). This book covers all the necessary aspects, from breaking up the cells (homogenization), via a variety of separation techniques (the isolation and fractionation chapters), to characterization of the separated organelles.
