Preface

Cytochromes P450 (P450) comprise a large superfamily of proteins that are of central importance in the detoxification or activation of a tremendous number of foreign hydrophobic compounds, including many therapeutic drugs, chemical carcinogens, and environmental pollutants. Many of these enzymes are induced by the compounds they metabolize. In addition, genetic polymorphisms of P450 genes can lead to adverse drug reactions. Consequently, P450s are one of the most extensively studied groups of proteins, being investigated by researchers in fields as diverse as toxicology, pharmacology, genetics, environmental biology, biochemistry, and molecular biology. The wide range of techniques that have been applied to the P450s reflects the diverse backgrounds of the many researchers active in this field.

The second edition of *Cytochrome P450 Protocols* contains a collection of key "core" techniques for the investigation of P450s. Although the emphasis is on P450s of mammalian origin, many of the methods described are suitable for the investigation of P450s from any source. Also included in this edition are chapters on the flavin-containing monooxygenases (FMOs), another family of proteins that are important in the metabolism of xenobiotics, and that share several substrates in common with the cytochromes P450.

Each chapter is written by researchers who have been involved in the development and application of the particular technique to P450s or FMOs. Protocols are presented in a step-by-step manner, with extensive cross-references to notes that highlight critical steps, potential problems, and alternative methods. We hope that this format will enable researchers who have no previous knowledge of the technique to understand the basis of the method and to perform it successfully.

Cytochrome P450 Protocols begins with a chapter on P450 nomenclature and classification, which will serve both as an introduction to those new to the field and as a guide for more experienced workers wishing to name their pet P450. Although not formally divided into sections, the remaining chapters are grouped according to topic. These include methods for spectral analysis and purification of P450s; enzymatic assays of P450s and FMOs; expression of P450s and FMOs in heterologous systems; production and use of anti-peptide antibodies; transfection of hepatocytes for gene regulation studies; P450 reporter gene assays; in situ hybridization; analysis of genetic polymorphisms;

vi Preface

and P450 allele nomenclature, including a description of the P450 allele website. Because of the increasing importance of in vitro systems for pharmacotoxicology research, we have included several chapters on the preparation and culture of rodent and human hepatocytes and the production of bone marrow stem cells. The final chapters describe more specialized techniques for the generation of mice with targeted gene disruptions.

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