Preface

Large-scale protein production has come a long way with the onset of recombinant DNA technology in the 1980s. Initially microbes, such as bacteria and yeast, were the choice of host used to produce commercially important proteins; their short generation time and growth to high densities in bio-fermenters were valuable traits. As technology became more sophisticated, other hosts such as cell lines, animals, and plants were explored. Plants lagged behind most other systems primarily because initial biotechnical work focused on agricultural improvement to crops rather than their use for the expression of novel products.

Attention has since turned to using plants as hosts to produce commercially important proteins. Many reviews have been written about the theoretical aspects of this topic but the present volume is focused on commercial successes: case studies of projects that have commercial potential or products that have already been commercialized, illustrating the advantages that plants can have over bacterial, fungal, or animal cell culture hosts. These case studies demonstrate the hurdles that must be overcome and the benefits of using plants to produce industrial and pharmaceutical proteins as well as vaccine antigens. It is predicted that plant protein production is the beginning of a new paradigm for the commercial production of proteins that over the next decade will expand dramatically.

The commercialization of plant-produced proteins has progressed slowly over the past 15 years since the first introduction of a commercial product demonstrating feasibility. Many factors have contributed to this slow progress, but, in brief, the technology was not robust and predictable in the early stages to compete on a strictly cost basis with other existing platforms, and there was little motivation to fund technology improvements to a system that was considered a threat to existing platforms. In the last several years, however, the advantages of plant production systems beyond the unit costs are enabling the acceptance of the technology. The clear front-runner is the move into an animal-free source of proteins for cell cultures. This may soon be followed by an animal-free source of therapeutics, a rapid system for the production of parenteral vaccines, orally delivered vaccines, and industrial enzymes that can only be produced on the scale that a plant system can provide. The advantages of plant-produced proteins beyond the unit cost are the

key to the initial commercialization. In the longer term as the technology becomes more engrained into the industry, this approach can be used for a variety of other proteins where plants can compete on unit cost as well.

In this volume, the focus is on products from plants that have either been commercialized or that are near commercialization. We have chosen protein products that illustrate the promise of the system, for example, highly purified proteins free of concerns over animal pathogen contaminants, directly delivered proteins such as orally delivered vaccines, or minimally processed industrial products.

This book is divided into four parts. The first part on *highly purified proteins* describes trailblazing technologies that are effective for the production of proteins at commercial production levels, at pharmacological and research-grade purities. Some of these proteins are toxic to cells when expressed at even moderately high levels, so they represent a major advance in strategies for the production of proteins that may interfere with normal cellular pathways. These strategies may be modified for use in non-plant systems.

The second part on *vaccines* examines strategies for administration of plant-produced antigens through oral and parenteral routes and for human and veterinary applications. The failure of straightforward approaches to vaccine production for pathogens that show antigenic drift has been addressed by the use of novel strategies such as transmission blocking vaccines, and these strategies may be extrapolated to other vector-transmitted diseases. Antigens that are presented in a structural form that resembles the pathogen are also examined. For veterinary application, vaccines effective for use in domestic herds and wild animals are examined. Some of the outcomes pursued are effectiveness, rapid production, cost-effectiveness, and ease of administration.

The third part on *industrial proteins* evaluates the production of proteins that have applications in the paper and food industries. A unique feature of these proteins is that they can perform their purpose without purification to homogeneity. Cellulase enzymes are effective for conversion of cellulose to biofuels but also for making wood amenable for conversion to paper pulp without the use of environmentally unsafe chemicals. Thus, the indirect effects of the use of these enzymes are also beneficial.

The final part on *future directions* examines the benefit of plants as hosts and reviews some of the possible applications and the regulatory and public perspectives with regard to their use.

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