Furthermore, as more biologically based therapeutic agents and delivery methods are invented, conventional pharmacology and toxicity tests may not always be appropriate to determine biological activity and to assess the safety of these agents. Transgenic technology facilitates the development of suitable animal models for testing conventional drug candidates and therapeutic agents that are derived from animal tissues (1). This chapter will provide an overview of different gene transfer methods to manipulate animal genomes and discuss their application in preclinical evaluation of new drugs, including assessment of the biological activities, metabolism, and toxicological effects of the drugs under development. Finally, a prospective view about transgenic applications in drug industry will be provided.

2. PRODUCTION OF TRANSGENIC ANIMALS

2.1. Principles

Two principal methods of changing animal genomes in the laboratory are random addition and targeted alteration of specific DNA sequences under investigation (Fig. 1). Random addition of genetic material into an animal genome is accomplished by introducing recombinant DNA directly into the pronuclei of oocytes that are then allowed to develop into mature animals (2). In technical terms, these animals are generally referred to as transgenic animals.

The second method is to introduce targeted alteration of genetic material at a specific genomic locus. This is accomplished by first changing a specific DNA segment in embryonic stem (ES) cells by a "gene-targeting" procedure (3,4), followed by the incorporation of the genetically altered ES cells into embryos which are then allowed to develop into mature animals. Animals derived from these embryos are generally called chimeric animals because they are composed of two genetically distinct cell populations (5). One cell population is derived from the recipient embryos and the second is derived from the genetically altered ES cells. By breeding the chimeric animals to a strain that is genetically distinct from the source of ES cells, animals with one set of chromosomes inherited from the ES can be selected among the offspring according to ES-specific genetic markers. Technically, these animals are referred to as gene-targeted animals because they inherit the targeted gene on one set of chromosomes that are derived from the ES cells. These gene-targeted animals, usually at a heterozygotic state, are subsequently bred within the same line to generate homozygote animals carrying the same targeted gene in both sets of chromosome. These animals are homozygous, gene-targeted animals.

To achieve a higher level of specificity in terms of where and when a specific gene-targeting event should occur, a more sophisticated approach combines the use of transgenic and gene-targeting procedures.