In EU legislation, a new directive on the protection of animals used for scientific purposes was issued in 2010 (EU, 2010), which updates and replaces the 1986 Directive 86/609/EEC (EEC, 1986). This new directive took full effect on January 1, 2013, and aims to anchor the principle of the "Three Rs" to Replace, Reduce, and Refine the use of animals.

In the EU, the use of animals for scientific or educational purposes should only be considered when a nonanimal alternative is unavailable (preamble 12). Member states need to ensure that, wherever possible, a scientifically satisfactory method or testing strategy, not entailing the use of live animals, is used instead (Article 4.1). Moreover, according to the directive, nonhuman primates (NHPs) are exempted from use in animal studies whenever possible. This is reflected in Article 8.1(b) as there should be scientific justification that the purpose of the procedure (animal study) cannot be achieved by the use of species other than NHPs.

6.5 THE EU BIOSIMILAR mAbs GUIDELINE

At the same time that the EU legislation on the protection of animals used for scientific purposes was being finalized, the EMA Working Party on Similar Biological Medicinal Products (BMWP) was drafting a guideline on the nonclinical and clinical issues of the development of biosimilar mAbs (EMA, 2012). The EMA has organized several workshops to discuss the feasibility of the development of biosimilar mAbs with the scientific community and pharmaceutical companies. With respect to nonclinical development, the debate was focusing on the need of animal studies in the comparability exercise and more specifically, on the justification of using nonhuman primates as laboratory animals.

As specified in ICH guideline S6 (R1), safety evaluation programs should include the use of relevant species (ICH, 2011). In the case of monoclonal antibodies, a relevant species is one in which the test material is pharmacologically active due to the expression of an epitope. Monoclonal antibodies are biologicals showing high species and target specificity, and mostly only animal species closely related to humans are pharmacologically responsive to the investigational medicinal product. For many mAbs, the nonhuman primate is often the only relevant species, and therefore, pharmacology and toxicity studies are often performed in the cynomolgus macaque (*Macaca fascicularis*) (Chapman et al., 2012). When comparative nonclinical studies have to be powered to detect small differences in efficacy and safety, these studies must be of considerable size to reach this goal. Studies in NHPs (and nonrodents in general) have small group sizes, and interindividual variability further reduces the sensitivity of these studies to detect differences in pharmacological response.

The usually small group size in animal studies (especially when nonrodents are being used) limits the sensitivity of these studies to detect relevant differences in safety and efficacy.

Another approach to provide safety data for some mAbs may be the use of animal models of disease, in which the pharmacological activity of a pharmaceutical can be shown and which can be used to demonstrate pharmacodynamics activity. Examples are the use of SCID mice with xenotransplants of tumors for oncology products, or transgenic mouse models, such as Tg197 carrying a modified human