Mimicking the biological activity in the clinical situation is not always necessary. A correlation between the expected clinical response and the activity in the biological assay should be established in pharmacodynamic or clinical studies.

The results of biological assays should be expressed in units of activity calibrated against an international or national reference standard, when available and appropriate for the assay utilized. Where no such reference standard exists, a characterized in-house reference material should be established, and assay results of production lots should be reported as in-house units.

Often, for complex molecules, the physicochemical information may be extensive but is unable to confirm the HOS, which, however, can be inferred from the biological activity. In such cases, a biological assay, with wider confidence limits, may be acceptable when combined with a specific quantitative measure. Importantly, a biological assay to measure the biological activity of the product may be replaced by physicochemical tests only in those instances where:

- Sufficient physicochemical information about the drug, including HOS, can be thoroughly established by such physicochemical methods, and relevant correlation to biologic activity can be demonstrated
- There exists a well-established manufacturing history
- Physicochemical tests alone are used to quantify the biological activity (based on appropriate correlation) (results should be expressed in mass)

For the purpose of lot release, the choice of relevant quantitative assay (biological and/or physicochemical) should be justified by the manufacturer.

9.9.8 Immunochemical Properties

When an antibody is the desired product, its immunological properties should be fully characterized. Binding assays of the antibody to purified antigens and defined regions of antigens should be performed, as feasible, to determine affinity, avidity, and immunoreactivity (including cross-reactivity). In addition, the target molecule bearing the relevant epitope should be biochemically defined and the epitope itself defined, when feasible.

For some DS or DP, the protein molecule may need to be examined using immunochemical procedures (e.g., ELISA and Western blot) and utilizing antibodies that recognize different epitopes of the protein molecule. Immunochemical properties of a protein may serve to establish its identity, homogeneity, or purity or may serve to quantify it.

If immunochemical properties constitute lot release criteria, all relevant information pertaining to the antibody should be made available.

9.9.9 Purity, Impurities, and Contaminants

9.9.9.1 Purity

The determination of absolute, as well as relative, purity presents considerable analytical challenges, and the results are highly method-dependent. Historically, the relative purity of a biological product has been expressed in terms of specific activity (units of