**Checklist for Protocol of Analysis**

| TEST | | INFORMATION REQUIRED | REMARKS |
|---|---|---|---|
| **Physical Tests** | **Statement according to pharmacopoeias or photocopies from pharmacopoeias shall not be accepted. Details of test methods shall include the following items:** | | |
| • Appearance<br>• Colour, Clarity and Opalescence<br>• Visible particles<br>• Subvisible particles<br>• pH<br>• Osmolarity<br>• Moisture content<br>• Extractable volume<br>• Dissolution time<br>• Homogeneity test<br>• Others | 1 | List of equipment and apparatus | |
| | 2 | List of chemical, reagents and media | |
| | 3 | Preparation of solutions such as sample, reference standard (if applicable), medium, buffer, etc | |
| | 4 | Volume and temperature of sample solution (if applicable) | |
| | 5 | Setting up of analytical instrumentation (if applicable) | |
| | 6 | Testing condition/ parameter (if applicable) | |
| | 7 | Testing procedure | |
| | 8 | System suitability tests (if applicable) | |
| **Identification / Characterisation Tests** | **Details of test methods shall include the following items:** | | |
| • Peptide Mapping<br>• Identification of preservative and active substance<br>• Precipitate reaction<br>• Microscopic examination<br>• Colony morphology<br>• Virus identification<br>• Others | 1 | List of equipment and apparatus | |
| | 2 | List of chemical, reagents and media | |
| | 3 | Preparation of solutions such as sample, standard, system suitability solution, mobile phase, medium, buffer, etc (the amount of chemical/sample/ standard and volume of diluents used in the preparation must be stated) | |
| | 4 | Setting up of analytical instrumentation | |
| | 5 | Testing condition/ parameter such as HPLC parameter, etc | |
| | 6 | Testing procedure | |
| | 7 | System suitability tests and acceptance criteria of system suitability test. | |
| | 8 | Complete formula for calculation (if applicable) and interpretation of results | |
| | 9 | Image of SDS PAGE/ IEF/ electropherogram/ TLC/ UV spectrum/ IR spectrum/HPLC chromatogram etc for blank, sample, standard and system suitability solution | |

| **Assay/ Potency/ Content/ Dissolution test** | **Details of test methods shall include the following items:** | | |
|---|---|---|---|
| • Protein concentration<br>• Content of active ingredient and preservative<br>• Bioassay/ Potency (animal- based, cell culture- based and biochemical- based)<br>• Dissolution test<br>• Others | 1 | List of equipment and apparatus | |
| | 2 | List of chemical, reagents and media | |
| | 3 | Preparation of solutions such as sample, standard, system suitability solution, mobile phase, medium, buffer, etc (the amount of chemical/sample/ standard and volume of diluents used in the preparation must be stated).<br>Stability and storage condition of sample and standard solutions | |
| | 4 | Setting up of analytical instrumentation | |
| | 5 | Testing condition/ parameter such as HPLC parameter, animal criteria, etc | |
| | 6 | Testing procedure | |
| | 7 | System suitability tests and acceptance criteria of system suitability test. | |
| | 8 | Data analysis, complete formula for calculation (the formula must provide in the unit stated in COA) and interpretation of results | |
| | 9 | HPLC chromatogram/ UV spectrum (if applicable) for blank, sample, standard and system suitability solution | |
| **Purity/ Impurities Tests** | **Details of test methods shall include the following items:** | | |
| • Known impurities<br>• Unknown Impurities<br>• High Molecular Weight Protein<br>• Monomer<br>• Dimer<br>• Aggregates<br>• Residual solvent | 1 | List of equipment and apparatus | |
| | 2 | List of chemical, reagents and media | |
| | 3 | Preparation of solutions such as sample, standard, system suitability solution, mobile phase, medium, buffer, etc (the amount of chemical/sample/ standard and volume of diluents used in the preparation must be stated)<br>Stability and storage condition of sample and standard solutions | |
| | 4 | Setting up of analytical instrumentation | |
| | 5 | Testing condition/ parameter such as HPLC parameter, etc | |
| | 6 | Testing procedure | |
| | 7 | System suitability tests and acceptance criteria of system suitability test. | |
| | 8 | Data analysis, complete formula for calculation (the formula must provide in the unit stated in COA) and interpretation of results | |
| | 9 | Image of SDS PAGE/ IEF/ electropherogram/ TLC/ HPLC chromatogram etc for blank, sample, standard and system suitability solution. | |

| **Other Safety Test** | **Details of test methods shall include the following items:** | | |
|---|---|---|---|
| • Pyrogen Test<br>• Bacterial Endotoxins Test<br>• Sterility Test | 1 | Refer to DRUG REGISTRATION GUIDANCE DOCUMENT (DRGD) under GUIDELINE FOR THE SUBMISSION OF PROTOCOL OF ANALYSIS (POA) | |
| **Others** | **Details of test methods shall include the following items:** | | |
| • Test for absence of virulent mycobacteria<br>• Test for excessive dermal reactivity<br>• Specific toxicity test<br>• Abnormal toxicity test (innocuity)<br>• Others | 1 | List of equipment and apparatus | |
| | 2 | List of chemical, reagents and media | |
| | 3 | Preparation of solutions such as sample, standard, medium, buffer, etc | |
| | 4 | Testing condition /animal criteria | |
| | 5 | Testing procedure | |
| | 6 | Calculation of the result (if applicable) or calculation method used | |
| | 7 | animal test: - specific requirement for the animal used such as weight, age, sex (if applicable) etc<br>- dose used and injection technique | |