BIOPHARMACEUTICAL LABORATORY SERVICES

WHEN YOU NEED TO BE SURE

SGS
SGS helps pharmaceutical, biopharmaceutical, and medical device companies by providing services along the entire drug development pathway. With over 35 years of experience as a global, life science, contract research organization, SGS provides integrated solutions from preclinical activities to Phase I-IV clinical trials, bioanalytical, pharmaceutical development, biologics characterization, biosafety, and quality control testing of small and large molecules, raw materials, containers and finished products. SGS has established several outsourcing models to adapt to its different client profiles. SGS can assist clients by identifying the best customized outsourcing model within our wholly-owned, global analytical laboratory network: from dedicated suites with-in one of our facilities to integrating and managing staff at a client’s site.
BIOPHARMACEUTICAL TESTING SERVICES

To support the rapidly developing biopharmaceuticals market, SGS offers a wide range of services in order to provide a comprehensive testing portfolio.

**BIOPHARMACEUTICAL TESTING SERVICES**

**BIOMARKERS TESTING SERVICES**

SGS provides a comprehensive range of biomarker services for biologics, including: virology, cell and molecular biology, as well as microbiology and electron microscopy. Health Authorities, including the US FDA and the EMA, require biologic products to undergo safety testing to demonstrate that all cell banks, viral banks, raw materials of animal origin, bulk harvests, and batches of clinical drug are free of bacteria, fungi, mycoplasma, viruses and other potential contaminants such as endotoxins.

**BIOSAFETY TESTING SERVICES**

As a leading bioanalytical service provider, with GLP/GMP compliant laboratories, SGS serves pharmaceutical and biopharmaceutical companies of all sizes with PK/PD testing, immunoassays, and cell-based assays at the preclinical and clinical stage of drug development. At SGS, bioanalysis testing is underpinned by a large list of validated methods and biomarkers – over 700 assays to date. SGS maintains its reputation as an industry pioneer by actively pursuing the assay development and validation of the more innovative biomarkers.
Choosing SGS as a partner provides access to the specialized expertise within the laboratory network for biopharmaceutical characterization, biosafety and bioanalytical testing. Additionally, services such as analytical chemistry and microbiological testing are available globally, in close proximity to where your facilities are located.
SGS’ range of services dedicated to biopharmaceutical product characterization bring the most recent developments in testing technology to companies across the globe. In 2010, SGS acquired the M-Scan Group, the world leaders in the application of advanced mass spectrometry techniques for protein and carbohydrate structure analysis. As a result, SGS broadened its service offering for biologics characterization with GLP/GMP contract analytical services, expert consultancy, and training.

SGS offers a full package of peptide, protein and glycoprotein analyses to GLP/cGMP standards and is the perfect partner for clients looking to outsource biologic product characterization. Additionally, SGS can provide analysis of post-translational modifications (glycosylation, disulfide bridges, etc), as well as higher order structure. All analyses are designed to help clients comply with ICH Q6B guidelines.
PROTEIN CHARACTERIZATION

PEPTIDE MAPPING

Peptide mapping can be used in drug discovery and throughout the manufacturing process for quality control between batches to produce a unique ‘fingerprint’ of an individual protein and to compare this with the theoretical gene-derived amino acid sequence. Protein mass mapping involves:

- Fragmenting the protein molecule using specific enzymatic or chemical methods
- Analyzing the resulting peptide mixture using direct MS or online LC-MS

Additionally peptide mass mapping can be extended to:

- Disulfide bridge assignment
- N-terminal and C-terminal sequence confirmation (along with MS/MS sequencing)
- Screening and identification of sites of post-translational modification such as glycosylation or phosphorylation
- Guiding the choice of signals for subsequent MS/MS or gas phase sequence analysis

AMINO ACID ANALYSIS

Amino Acid Analysis is performed throughout the manufacturing process, where it is used to ensure consistency between drug batches.

Amino Acid Analysis can be used for:

- Protein concentration calculations and characterization
- Determination of extinction coefficient by coupling this process with UV spectrophotometry for optical density measurement
- Total amino acids, including cysteine, tryptophan, and hydroxyproline
- Free amino acids, including asparagine and glutamine

Meets ICH Q6B Guidelines
MOLECULAR WEIGHT ANALYSIS

The intact molecular weight is a key characteristic of a molecule’s identity. SGS’ molecular weight analysis techniques include mass spectrometry, offering accuracy of measurement over a wide molecular weight range, with minimal sample consumption. Furthermore, any difference between theoretical and measured mass may be indicative of potential modifications.

With a wide range of mass spectrometry instruments, SGS is able to select the most appropriate ionization technique for your compound, including:

- Matrix Assisted Laser Desorption Ionization Mass Spectrometry (MALDI-MS)
- Electrospray Ionization Mass Spectrometry (ES-MS)
- Liquid Chromatography with Mass Spectrometry Detection (LC-MS) for complex mixtures.

GEL ELECTROPHORESIS

SGS’ protein gel electrophoresis service offers you a range of techniques for various applications in biopharmaceutical product characterization, such as measuring molecular weight and characterizing impurities. SGS can help you:

- Assess the molecular weight and purity of a protein, including the identification of the presence of truncated forms
- Determine the isoelectric point of a protein and establish the presence of charge heterogeneity due to post-translational modifications, such as glycosylation or deamidation, with isoelectric focusing (IEF)
- Assess product-related impurities, such as aggregation with native PAGE
- Study cellular protein expression, resolving tens to hundreds of proteins through a combination of IEF and SDS-PAGE analysis (2D)

LIQUID CHROMATOGRAPHIC PATTERNS

SGS offers a range of techniques, including Size Exclusion Chromatography (SEC), Ion Exchange Chromatography (IEX) and Reverse-Phase Liquid Chromatography (RP-HPLC) Analysis. SGS can help:

- Acquire data on the identity, homogeneity and purity of proteins and glycoproteins
- Identify the unique fingerprint of an individual protein with RP-HPLC analysis
- Purify and separate proteins for further characterization and analysis with IEX chromatography
- Demonstrate comparability between different batches of protein sample lots
- Assess molecular weight or size with Size Exclusion Chromatography (SEC)

CAPILLARY ELECTROPHORESIS ANALYSIS

When superior isoelectric focusing resolution and reproducibility is required, capillary isoelectric focusing analysis (cIEF) is the answer.

SGS’ analytical services enable you to:

- Determine the isoelectric point of a protein
- Establish the presence of charge heterogeneity due to post-translational modifications, such as glycosylation or deamidation
PROTEIN SEQUENCING

C-TERMINAL SEQUENCE ANALYSIS

The determination of C-terminal microheterogeneity is crucial to guarantee a high-quality biopharmaceutical product. SGS C-terminal sequencing experts can identify the intact C-terminus of your product and where post-translational modification has produced truncated versions or “ragged ends”.

Because there is no standard approach to confirming the C-terminal sequence, our clients rely heavily on our expertise in selecting a cutting-edge approach to C-terminal protein sequencing. We base our choice of method on our past experience and factors such as:

- Protein or peptide size
- Glycosylation state
- Theoretical C-terminal amino acid sequence

Confirmation of C-terminal sequence can usually be achieved in combination with the following:

- Peptide mapping
- ES-MS/MS sequencing
- Enzymatic digestions
- Molecular weight analysis

N-TERMINAL SEQUENCE ANALYSIS

Using automated Edman Degradation Sequencing, also known as Gas Phase Sequencing (GPS), our experts work on samples in solution or from proteins that have been separated using SDS-PAGE and blotted onto PVDF membranes. GPS will:

- Sequentially cleave the constituent amino acids from the N-terminus of the sample
- Derivatize and separate the produced amino acids by reverse-phase high pressure liquid chromatography
- Visualize the amino acids by UV detection for each cycle of Edman chemistry
- Quantify the amino acids by comparison to a standard mixture

In addition to N-Terminal Sequencing SGS is able to assess possible truncations of the amino-terminus or levels of chemically blocked termini that usually prevent full sequencing. Additionally, we may also be able to remove the blocking group before sequencing.

DE NOVO PROTEIN SEQUENCING

De novo protein sequencing is the process by which the amino acid sequence is deduced without prior knowledge of the DNA or protein sequence. This differs from sequence confirmation, where the protein/DNA sequence is already known and the sequence data obtained is used to confirm that it is correct.

Performed by MS/MS, SGS’ de novo sequencing service reveals structural information by fragmenting intact peptides within a mass spectrometer. In addition, or as an alternative, sequencing may be performed by N-terminal sequencing, which sequentially separates amino acids from the N-terminus of a peptide using Edman Chemistry.

De novo sequencing is used in the identification of unknown proteins in proteomic type analyses. Another important application is in the characterization of antibody variable regions.

Meets ICH Q6B Guidelines
Proteomics is the investigation of gene expression at the protein level. Gene expression and hence cellular protein content can vary both quantitatively and qualitatively according to treatment regimes and/or stress conditions. SGS provides an effective proteomic analysis to investigate gene expression at the protein level.

SGS’ proteomics services help you to:

- Identify proteins and protein modifications such as phosphorylation, mutation or glycosylation
- Detect low levels of a peptide present in a mixture
- Conduct impurity profile analysis for recombinant proteins, detecting the presence of minor levels of impurity proteins such as host cell proteins

Our proteomics service strategy includes the following:

- Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) to separate and visualize protein content
- Reverse-Phase Liquid Chromatography (RP-HPLC) to separate and visualize protein content
- Electrospray Ionization with Tandem Mass Spectrometry (ES-MS/MS) to sequence peptides produced from the digestion of proteins
- Nano Liquid Chromatography with Tandem Mass Spectrometry Detection (LC-MS/MS) to sequence peptides produced from the digestion of proteins
- Protein sequence database interrogation to identify proteins in the sample
ANALYSIS OF STRUCTURAL MEASUREMENTS

Meets ICH Q6B Guidelines

High Performance Anion Exchange Gas Chromatography-Mass Spectrometry (GC-MS)

■ Determine the presence of carbohydrate impurities, for example glucose contamination of a sample from purification column matrices or dialysis systems
■ Conduct an initial investigation prior to more detailed analysis of the glycosylation

Monosaccharide composition analysis is most commonly performed using:
■ Gas Chromatography-Mass Spectrometry (GC-MS)
■ High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD)

■ Glycosylation plays an important role in the safety and efficacy of drug products.

At SGS, we have shaped our glycosylation analysis service to satisfy the requirements of ICH 6QB, so you can be sure you are meeting regulatory expectations. Our experts are always on hand to give advice on what may be most appropriate for you, whether you are using glycosylation analysis for drug discovery or at any stage of the drug manufacturing process.

Highlighting where factors such as cell-line and bioreactor conditions have affected the structure, linkage or composition of glycans within a protein, it is also a key tool in helping to protect your brand’s reputation for high-quality products.

MONOSACCHARIDE COMPOSITION ANALYSIS

SGS offers monosaccharide analysis to help you identify the type of glycosylation (such as N-linked and/or O-linked) and the extent to which glycosylation has occurred:

■ Determine the presence of carbohydrate impurities, for example glucose contamination of a sample from purification column matrices or dialysis systems
■ Conduct an initial investigation prior to more detailed analysis of the glycosylation

Glycosylation site analysis strategies

■ Glycosylation Site Analysis
■ Glycan Population Analysis
■ Linkage Analysis

Glycosylation plays an important role in the extent possible.

■ Monosaccharide composition analysis to help you identify the type of glycosylation (such as N-linked and/or O-linked) and the extent to which glycosylation has occurred:

■ Determine the presence of carbohydrate impurities, for example glucose contamination of a sample from purification column matrices or dialysis systems
■ Conduct an initial investigation prior to more detailed analysis of the glycosylation

Monosaccharide composition analysis is most commonly performed using:
■ Gas Chromatography-Mass Spectrometry (GC-MS)
■ High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD)

Glycosylation sites are initially identified as part of a peptide mapping study using Liquid Chromatography–Mass Spectrometry (LC-MS). Glycosylation at each individual site is then characterized following release of the glycans from each purified glycopeptide using a range of effective techniques, including:

■ Molecular ion data from Matrix Assisted Laser Desorption Ionization Mass Spectrometry (MALDI-MS)
■ Fragment ion information from Electrospray Mass Spectrometry (ES-MS)
■ Chromatographic profiles from High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD)
■ Hydrophilic Interaction Liquid Chromatography with Fluorescence Detection (HILIC-FLD)
OLIGOSACCHARIDE POPULATION ANALYSIS

Oligosaccharide population analysis provides you with a powerful technique to determine the overall population profile of the N-glycans or O-glycans present in a glycoprotein.

SGS’ oligosaccharide analysis helps you to:

- Identify unusual glycosylation profiles
- Confirm the presence of N- or O-glycosylation
- Detect potentially immunogenic glycan structures

Oligosaccharide population analysis is performed following release of the glycans from the protein using the following techniques:

- Molecular ion data from Matrix Assisted Laser Desorption Ionization Mass Spectrometry (MALDI-MS)
- Fragment ion information from Electrospray Mass Spectrometry (ES-MS)
- Chromatographic profiles from High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD)
- Hydrophilic Interaction Liquid Chromatography with Fluorescence Detection (HILIC-FLD)

LINKAGE ANALYSIS

Linkage (methylilation) analysis of glycoproteins and carbohydrates can be used to determine the identities of the various linkages between the monosaccharides present. Plus, in conjunction with oligosaccharide population analysis, SGS can provide a detailed picture of the glycans and identify specific linkages of functional importance, such as 3- versus 6-linked sialic acid.

SGS’ techniques include Gas Chromatography-Mass Spectrometry (GC-MS) linkage (methylilation) analysis to help you detect potentially immunogenic glycans.

SIALIC ACID ANALYSIS

Sialic acid analysis helps you to determine the identities and quantities of the sialic acid species present, an important analysis since sialic acid content may influence serum half-life of a glycoprotein.

SGS offers techniques to help you:

- Determine and quantify sialic acid species, such as N-Acetylenuraminic acid (NeuAc) and N- N-Glycolyneuraminic acid (NeuGc)
- Screen glycoproteins manufactured using different cell lines

This analysis is performed using High Performance Anion Exchange Chromatography with Pulsed Amperometric Detection (HPAEC-PAD).

DISULFIDE BRIDGE ANALYSIS

The number and position of disulfide bridges (also called disulfide bonds) plays a crucial role in obtaining a correctly folded protein product essential for function.

SGS’ analytical services help you to:

- Identify the number and position of disulfide bridges present
- Detect low levels of potential mismatched or scrambled disulfide bridge forms using powerful mass spectrometric-tric techniques

SGS achieves disulfide bridge and free sulfhydryl group assignment using peptide mapping-based strategies. Analysis generally involves digestion using a suitable enzyme and/or chemical and examination of the resulting mixture using Liquid Chromatography with Mass Spectrometry Detection (LC-MS) to isolate and identify potential disulfide bridged peptides.

The potential bridges can then be confirmed by reduction and re-analysis using:

- Molecular ion data from Matrix Assisted Laser Desorption Ionization Mass Spectrometry (MALDI-MS)
- Fragment ion information from Electrospray Mass Spectrometry (ES-MS)
- Electrospray Ionization with Tandem Mass Spectrometry (ES-MS/MS)
The three dimensional structure of a protein plays an important role in conferring biological function. Protein structure may potentially be altered during manufacturing, storage and delivery, thereby affecting safety and efficacy of the protein therapeutic. Thus, characterization and detailed knowledge of the biophysical properties such as confirmation, aggregation, and particulates formation during the drug development cycle is crucial.

SGS has devised its higher order structure characterization package to satisfy the regulatory requirements. Our experts are always at hand to give advice on what may be the most appropriate biophysical method for the characterization of the quality attributes at different stages of product life cycle.

**CIRCULAR DICHROISM (CD) ANALYSIS**

All new biotechnological or biological products require spectroscopic profiling as proof of conformance. SGS provides GLP/cGMP-compliant Circular Dichroism (CD) analysis and class-leading spectroscopic profiling to help you meet your regulatory obligations.

SGS offers CD spectroscopic analysis for secondary structure determination (alpha helix/beta sheets) and tertiary structure assessment. As a result, we can help you:

- Gain data for protein folding and conformational studies (secondary structure)
- Establish DNA/RNA interactions and enzyme kinetics
- Determine the purity of optically active substances
- Undertake quantitative analysis of pharmaceuticals
- Perform rapid scanning (time resolved) experiments

**PROTEIN NUCLEAR MAGNETIC RESONANCE**

Obtaining high-resolution data on unstructured proteins can be challenging. However, protein NMR spectroscopy is one of the only techniques to make it possible. Protein NMR service enables you to analyze the chemical structure and composition of biological molecules.

SGS’ protein NMR spectroscopy can help you:

- Obtain high-resolution data on intrinsically unstructured proteins through 1D and 2D NMR analysis
- Analyze the chemical structure and composition of biological molecules, such as proteins, oligosaccharides and nucleic acids
- Gain insight into the three-dimensional structure of molecules
- Assess the conformational integrity of protein-based therapeutics
- Compare a sample with a known published product

Research-level analysis are performed on two biologically equipped 500 MHz and 800 MHz shielded NMR spectrometers, with cryoprobe technology, providing some of the most sensitive instrumentation available.

Meets ICH Q6B Guidelines
DIFFERENTIAL SCANNING CALORIMETRY

Differential Scanning Calorimetry (DSC) is a popular biophysical tool to measure thermodynamic parameters of proteins and other macromolecules. DSC is used for:

- Determination of the thermal stability of the materials under investigation by determination of important calorimetric measurables like the transition midpoint or melting point (Tm), calorimetric enthalpy (ΔH) and van’t Hoff enthalpy (ΔHvH)
- Determination of the reversibility of the unfolding reaction in order to accomplish a full thermodynamic characterization

DSC is especially suitable in characterization and comparability studies where buffer conditions like pH, ionic strength and excipient composition are changed in pursuit of a more stable formulation. Since the stability of a protein depends on environmental conditions, Tm is a useful thermodynamic parameter to screen the stability of proteins on different formulations.

PROTEIN AGGREGATION SERVICES

Protein aggregation is a common problem during the manufacture and storage of proteins. It can be the cause of immunogenicity (small aggregates) or potential problems with administration (large aggregates). SGS offers you a range of protein aggregation services for therapeutic products and biopharmaceuticals.

SGS’ protein aggregation services can help you:

- Analyze protein aggregations in your biopharmaceuticals, in line with regulatory expectations and requirements
- Determine the amounts of aggregate present in your biopharmaceutical products with Analytical Ultra Centrifugation (AUC)
- Analyze biological molecules and identify molecular weight profiles with Size Exclusion Chromatography with Multi-Angle Laser Light Scattering (SEC-MALS)
- Test product related impurities and exogenous particulate contaminants in the size range 0.5nm to 6µm with Dynamic Light Scattering (DLS) analysis

As the world-leading provider of protein aggregation services, SGS delivers the experience, technical and regulatory expertise, and unique global network you need.
ANTIBODY PRODUCT ANALYSIS

Antibody products should be characterized according to ICH, EMA and FDA guidelines. SGS provides a full GLP/cGMP analysis package for product characterization, pharmaceutical development (preformulation method development and validation, stability) and identification of post translational modifications, which include:

Structural characterization and confirmation:
- Amino acid sequence
- Amino acid composition
- N- and C-terminal sequence
  - N-terminal sequencing (automated Edman)
  - MS/MS peptide sequencing
- Peptide map
- Sulphydryl groups and disulfide bridges
- Carbohydrate structure
  - Monosaccharide composition analysis
  - Sialic acid analysis
  - Oligosaccharide profiling by HPAEC-PAD, HILIC-FLD and MS
  - Linkage analysis
  - Glycosylation site analysis
- Deamidation
- Oxidation
- Stability & preformulation

Physicochemical properties:
- Molecular weight or size
  - Gel electrophoresis
  - MALDI-TOF or ES-Q-TOF-MS
- Isoform pattern
- Extinction coefficient
- Electrophoretic pattern
  - Imaging cIEF
- Liquid Chromatographic patterns
  - Reverse Phase
  - Size Exclusion
  - Ion-Exchange profiling
- Spectroscopic profiles
  - UV/Vis
  - Circular Dichroism
  - NMR
- Aggregation
  - Analytical Ultracentrifugation (AUC)
  - SEC-MALS
- Differential Scanning Calorimetry
In 2012, SGS acquired Vitrology in order to provide a comprehensive range of biosafety services for biologics, including: virology, cell and molecular biology as well as microbiology and electron microscopy. Health Authorities, including the US FDA and the EMA, require biopharmaceuticals to undergo safety testing to demonstrate that all cell banks, viral banks, raw materials of animal origin, bulk harvests, and batches of clinical drug are free of bacteria, fungi, mycoplasma, viruses and other potential contaminants such as endo-toxins. SGS supports clients by ensuring product safety in satisfying these regulatory requirements through a large range of validated assays and develops new services in the following areas:

- Raw material, bulk harvest testing and finished products testing (sterility, endotoxin, mycoplasma, viruses and other potential biological contaminants)
- Final product testing for residual DNA and other process related impurities
- Formulation and stability studies
- Regulatory and safety consultancy services
- Custom development of assays

In order to facilitate access to these services, SGS operates a Sample Processing Center from its Life Sciences facility in Lincolnshire, IL, just outside of Chicago. This logistic platform, with Biosafety Level 2 capabilities, serves as a centralized receiving point for North American clients with biologics samples for virus testing and related biologics assays.
SGS provides validated in vitro adventitious virus assays, and retrovirus assays, designed to meet US and European regulatory guidelines. The assay systems available are intended for cell bank characterization, or characterization of products derived from cell lines, including human or rodent origin and for batch release, both for clinical trials or market release. In addition, SGS provides clients with validated assays for the detection of viruses derived from animal products (e.g., of bovine or porcine origin), and for screening viral vectors/vaccines for the presence of infectious contaminants.

**IN VITRO ADVENTITIOUS VIRUS ASSAYS**

Manufacturers of biopharmaceutical products derived from cell lines of human or animal origin are required to test their products throughout production to confirm the absence of contaminating infectious viruses. Such testing is required on master cell banks, working cell banks, cells at the limit of in vitro cell age, and unprocessed bulk harvest material. Similar testing is also required for virus vector and vaccine seed stocks, although there may be difficulties in detecting contaminating viruses in the presence of a live virus vector, and a customised approach to testing may be required for such products. (ICH Q5A; FDA PTC 1993; Ph.Eur. 2.6.16)

The following validated in vitro assays for the characterization of products derived from rodent or human cell lines are available:

- 14 day *in vitro* assays using a custom-ized combination of cell lines MRC-5, Vero, CHO-K1, HeLa, HEK293, Murine, etc.
- 28 day *in vitro* assays using a custom-ized combination of cell lines MRC-5, Vero, CHO-K1, HeLa, HEK293, Murine, etc.

Indicator lines suitable for use in the testing of other products, or to extend the host range of the indicator panel, are also available.
RETROVIRUS INFECTIVITY ASSAYS

The majority of cells contain endogenous retrovirus or retrovirus-like sequences, and some cells (e.g., murine cells) may produce infectious retrovirus. Regulatory guidelines typically require retrovirus testing by infectivity assay and electron microscopy, on master cell banks and cells cultured up to or beyond the limit of in vitro cell age, as well as unprocessed bulk material.

Infectivity assays may involve:

- XC cell line assays to detect infectious ecotropic MLV (Murine Leukaemia Virus)
- S+L- (sarcoma-positive, leukemia-negative) assays using feline or mink cell lines, to detect amphotropic or xenotropic MLV
- Mus dunni cells, with either immuno-fluorescence end-point or a PG4 cell end-point, for detection of ecotropic, amphotropic, xenotropic and mink cell focus-forming viruses

For non-murine retroviruses, infectivity testing on appropriate indicator cells (selected for their susceptibility to different retrovirus types) is recommended, and may be augmented with sensitive PERT assays.

Validated retrovirus infectivity assays for the detection of Murine ecotropic, amphotropic, xenotropic and mink-cell focus forming viruses and co-cultivation assays to amplify viruses with a tropism for human cells are available.

SGS is able to design complex and customized tests to meet specific product and client requirements.
The threat of contamination of cell cultures by adventitious agents such as mycoplasma, endotoxin and viruses, should not be underestimated. Our services therefore test for a range of contaminants at any stage of drug development and production.

SGS can test for:

- Mycoplasma, which can alter virtually every cellular process, including hybridoma selection rates, protein and nucleic acid synthesis, and immunogenicity and chromosomal damage
- Viral Contamination, which can distort product biomedical and biotechnological research and clinical evaluation, and bring about significant productivity losses in biological manufacturing
- Endotoxins, which when present in a final product can induce an inflammatory response in patients

**TESTS FOR MYCOPLASMA DETECTION**
- Cell culture method (21 CFR 610.30 and EP 2.6.7)
- Indicator cells and DNA
- Fluorochrome staining
- Validated PCR based method with pre-enrichment or direct detection
- Identification of mycoplasma strains by DNA sequencing

**ENDOTOXIN TEST METHODS**
- LAL gel clot assay
- Photometric assay

**VIRUS TESTING**
- EP 2.6.16 and 5.2.3 and ICH Q5A
- In vitro adventitious virus assays by CPE, haemadsorption, and haemagglutination assay
- Virus titration by plaque assay

**SAMPLE MATERIALS**
- Biopharmaceutical samples, including: raw materials, intermediate products, API, bulk drugs, and finished products
- Working and master cell banks
- Culture media

Conducted by our network of international experts, whose skills and knowledge are second to none, SGS’ biopharma safety testing methods are compliant with FDA, USP, EP and ICH guidelines, and also help to ensure that biopharmaceutical products meet other international and local regulatory requirements.

Contact our consultants to discuss how SGS could help with your biologics safety testing and produce data to ensure the best possible performance from your product during production and once it hits the market.
SGS provides Transmission Electron Microscopy (TEM) services, including negative staining and thin sectioning techniques for the detection of adventitious agents throughout the biopharmaceutical and vaccine manufacturing process, testing:

- Master Cell Banks
- Working Cell Banks
- Cell lines
- Culture supernatants
- Fermenter bulk harvests
- Vaccines

SGS’ cell biologists have exceptional theoretical and practical scientific experience within the CRO and academic environment. Our highly trained electron microscopy staff has applied these techniques successfully to the biological manufacturing industry over the past 10 years. A Philips CM-10 transmission electron microscope, equipped with both plate and digital camera systems, can process digital images, providing fast throughput of results and audited reports.

SGS’ microscopy techniques include:

- Transmission Electron Microscopy
  TEM allows visualization of virus particles, both in biological fluids and in vitro. It allows direct visualization of particle size and morphology, intra- and extra-cellularly. TEM is also a reliable technique to identify viruses in biological samples. SGS offers protocols for the observation of viruses, virus-like particles, adventitious and other extraneous agents by TEM of at least 200 cell profiles.

- Virus Particle Detection & Quantification
  Negative Stain Electron Microscopy (NSEM) has been used extensively for over 50 years to identify and characterize virus particles. Negative staining is also used for the monitoring of retroviral load quantities in fermenter bulk harvest material prior to downstream purification/processing.

Murine (NS-1) cell area showing endogenous retroviruses; intracisternal A-type particles in distensions of the rough-surfaced endoplasmic reticulum, and C-type particles in the extracellular space. Photo courtesy of SGS Vitrology.
MOLECULAR BIOLOGY SERVICES

Molecular biology services by SGS are used in the quality control of biological manufacturing and batch release testing of drug product. Testing by SGS ensures that final products derived from continuous mammalian cell lines contain acceptable levels of residual host cell DNA (HCD), and that biological starting materials and mammalian cell substrates are free of contaminating viruses, including retroviruses.

SGS’ molecular biology experts were first to publish the NAT validation and assay control system currently recommended in the European Pharmacopoeia 2.6.21. (Nucleic acid amplification techniques) and 2.6.7. (Mycoplasmas: Validation of NAT for the detection of mycoplasma: guidelines).

Services Include:

■ Residual Host Cell DNA Assays validated to ICH Q2 guidelines
■ cGMP compliant lot release testing, in which the drug product requires a detailed qualification study with the relevant HCD assay, ensuring that the HCD assay meets the required specifications
■ Retrovirus testing using PERT assays, including a cellular DNA polymerase suppressant termed calf thymus DNA for high sensitivity, ensuring specific detection of retroviral RT activity
  • Quantification of Reverse Transcriptase (RT) by Quantitative Real Time Fluorescent Product Enhanced Reverse Transcriptase (qPERT) assay
  • Detection of Reverse Transcriptase (RT) by Real Time Fluorescent Product Enhanced Reverse Transcriptase (FPERT) assay, for highly sensitive qualitative fluorescent results are required for vaccine seed or bulk produced in human cell substrates
■ Species-specific Virus Detection by qPCR, for example: porcine circovirus & vesivirus
■ Mycoplasma detection by qPCR
■ Identity testing by isoenzyme and RAPD
■ Genetic stability testing

Each SGS Scientist has at least 10 years experience in setting the current biopharmaceutical industry standards for the development and validation of Platinum-grade high-throughput real-time quantitative PCR (qPCR) assays. SGS provides EU, US and Japanese pharmacopeia-compliant assays for a wide variety of applications.

All qPCR assays are performed on ABI 7900HT using TaqMan technology, in accordance to criteria required for maintaining 21CFR part 11 compliance. Our interactive IT system coupled with our ability to condense our workflow onto high-density 384-well qPCR applications provides enhanced assay turnaround times for our clients.

SGS has developed and validated in excess of 100 species-specific qPCR assays, covering viral pathogens from Human, Murine, Simian, Porcine, Insect, Duck, Avian, Bovine species.
As a leading bioanalytical service provider, with GLP/GMP compliant laboratories, we are able to serve pharmaceutical and biopharmaceutical companies of all sizes with a range of tests for drug development at both pre-clinical and clinical stages – from early to late phase. We also develop assays (including immunassays) from scratch.

SGS bioanalysis testing is underpinned by a large list of validated methods and biomarkers, and to maintain our reputation as an industry pioneer we have actively pursued the assay development and validation of some of the more innovative of these biomarkers with our clinical teams. To date we have over 700 assays validated. Our services span:

- Cell-based bioassays
- Serology and immunogenicity testing
- Metabolite profiling
- ADME ¹⁴C
- Method transfer, development, optimization and validation
- PD bioanalysis (biomarkers)
- PK bioanalysis (small molecules & biopharmaceuticals)
In collaboration with our Clinical Research Team, SGS has actively pursued innovative biomarker assay development and validation. To complement our range of immunoassays, we offer LC-MS/MS technology that has enabled us to extend our portfolio to include more biomarkers such as steroidal and hormonal substances in the low picogram range. SGS has also invested in the field of multiplexing, using the bead-based suspension and electrochemiluminescence-based platforms to offer robust immunoassays for application in clinical trials.

SGS has over 20 years experience in biomarker assay development services and analysis operating out of GLP/GMP laboratories. A staff of 125 scientists makes our bioanalytical facility one of the largest in Europe. Pharma and biotech companies of all sizes choose SGS as their preferred bioanalytical partner for the expertise and experience to develop assays de novo – and support large-scale routine sample analysis.

Whether for testing efficacy or safety of drugs in development, our biomarker bioanalytical services cover a range of therapeutic areas to include:

- Cardiovascular
- Central nervous system (CNS)
- Metabolic disorders
- Inflammation
- Thyroid disease
- Oncology
- Sexual health
- Bone disease
- Allergy and respiratory
CELL-BASED ASSAYS

SGS’ experts are highly experienced in both implementing client assays and developing them de novo. Our laboratories in France and Belgium are both GLP/GMP certified and routine audits from the local agencies have proven that our laboratories consistently attain the highest standards in GLP compliance. Assay targets include both small and large molecule drug products, as well as pathogenic organisms (GMP). In these (GLP) facilities, SGS provides bioanalytical support for approximately 100 pre-clinical studies per year.

SGS’ In Vitro assay service and cell-based bioassay services include:

MODELS:
- Monolayers: Customized sourcing of primary fibroblast or keratinocyte cultures
- Human and animal cell lines (L929, Vero, IMR-90, MRC-5, CHO, HL60, etc.)
- 3D skin model (epidermis – full thickness)
- 3D human corneal epithelium
- 3D human lung epithelium model
- CACO-2 (human small intestine model)
- Isolated human blood cells (PBMC, lymphocytes, monocytes, etc.)

STUDY TYPES
- Barrier and metabolic functionalities as in vivo
- Percutaneous absorption and metabolism, skin irritation, sensitization, corrosivity tests, phototoxicity, wound healing
- Prediction of eye irritation potential of chemicals (alternative for in vivo Draize test)
- Intestinal absorption and metabolism
- Medical device cytotoxicity
- Permeability of drug substances
- Toxicity of API or excipients
- API receptor binding assays
- Wound healing
- Physiological effect on cell cultures
- Biomarkers synthesis and release
- Bioassays for neutralizing antibodies
- Cytokine production
- Phagocytosis/Opsonization
- Study of Intracellular Enzyme Activity (COX, MPO, etc.)
- Study of Signal transduction (STAT-1, p38 MAPK, P44, AKT/PKB, etc.)
- Immune cell activation and toxicity study types:
  - Biomarkers (cell receptor activation & signaling)
  - Phagocytosis of neutrophiles/macrophage
  - Opsonization
  - Phototoxicity
- Flow cytometry: BD FACS Verse (Immunophenotyping/Immunotoxicity)
- Immunogenicity of vaccines
- Cell-mediated immunity

DETECTION OF ANTIBODY TITERS AGAINST PATHOGENS (NON-EXHAUSTIVE LIST)

VIRAL
- Influenza viruses (H1N1, H3N2, H5N1, B, ...)
- Polio viruses (type 1, 2 and 3)
- Hepatitis A & B
- Measles
- Mumps
- Rubella
- Varicella
- Yellow Fever
- HIV, ....

BACTERIAL
- Pertussis
- Diphtheria
- Tetanus
- Hib (H. influenzae)
- Streptococcus Group B & S. pneumoniae
- N. meningitidis
SEROLOGY, IMMUNOGENICITY & NEUTRALIZING ANTIBODIES

SGS has been working in the immunoanalysis and immunogenicity testing field for many years. Similar to cell-based assays, targets include both small and large molecule drug products (GLP), as well as pathogenic organisms (GMP). Our multidisciplinary team of principal scientists and immunoanalysts works with our clients to address their immunoanalytical requirements on a case-by-case basis, offering:

- Expertise for both pre-clinical and clinical testing
- Various assay formats (bridging or direct ELISA and RIA)
- Method transfer or development
- Method validation following current guidelines and literature
- Immunogenicity screening assays (peptide/protein drugs and therapeutic monoclonal antibodies):
  - Detection of anti-drug antibody (ADA)
  - Appropriate cut points
- Immunogenicity confirmatory assays:
  - Demonstration of ADA specificity
  - Use of confirmatory cut points
- Antibody characterization:
  - Neutralizing ADA assays
  - Cell-based assays
  - Isotyping
- Titration testing
- Cytokine profiling

DETECTION OF ANTIBODY TITERS AGAINST PATHOGENS (NON-EXHAUSTIVE LIST)

- **VIRAL**
  - Influenza viruses (H1N1, H3N2, H5N1, B, ...)
  - Polio viruses (type 1, 2 and 3)
  - Hepatitis A & B
  - Measles
  - Mumps
  - Rubella
  - Varicella
  - Yellow Fever
  - HIV, ...

- **BACTERIAL**
  - Pertussis
  - Diphtheria
  - Tetanus
  - Hib (H. influenzae)
  - Streptococcus Group B & S. pneumoniae
  - N.meningitidis

FLOW CYTOMETRY ASSAY SOLUTIONS

SGS also offers the highly versatile technology of flow cytometry analysis to support clinical development. We currently operate a BD FACS Verse (three lasers, eight colors) equipped with a high throughput sampler.
PHARMACOKINETIC TESTING

In order to support the bioanalysis of various classes of compounds (including peptides), SGS offers a large range of techniques and methods providing rapid high volume bioanalysis. Our method development processes focus on high-throughput technological capabilities (e.g. TurboFlow extraction, multiprobe robots).

SGS offers more than 700 validated bioanalysis methods that are ready for use with a very short lead-time. Validation criteria follow guidelines from the FDA (May 2001) and the Crystal City Conference (May 2006). All validations are tailored to specific program requirements. SGS has the expertise to both develop assays de novo (including immunoassays) and to support large scale routine sample analyses, from pre-clinical to clinical studies.

DRIED BLOOD SPOT ANALYSIS

Dry Blood Spot (DBS) technology has several advantages for toxicokinetic (TK) and pharmacokinetic (PK) analysis, including: reduced blood sampling collection, absence of post-collection processing, low biohazard risk, fewer required facilities for sample shipment and storage. Furthermore, sample analysis can also be simplified.

EARLY TO LATE PHASE CLINICAL TRIALS AND POST-MARKETING

To support the bioanalysis of various classes of compounds, a large range of techniques and methods are available to you. Also, an exceptional sample processing and analytical capabilities, combined with scientific expertise, enables us to provide rapid, high-volume bioanalysis:

- Mass Spectrometry
  - 29 LC-MS/MS, 5 GC-MS
- Immunoanalysis
  - Singleplexed Custom Immunoassay Development
  - EIA's
  - Radioimmunoassay (RIA)
  - Multiplexed Custom Immunoassays
  - BD Beads array and quantification by BD FACS Verse + High Throughput Sampler for 96- and 384-well microtiter plate capacity driven by BD FACSuite™ software.
- Classical HPLC
  - 6 HPLC systems equipped with UV, fluorescence and electrochemical detectors
- Automated sample preparation using robotics (multiprobe) or turbulent flow technology (cohesive)
- Large sample storage capacity combined with excellent sample tracking (400,000 sample storage and equivalent backup)
- Current throughput of more than 200,000 incurred samples per year
- Sample handling and tracking by the Watson LIMS
METABOLIC PROFILING

With over 25 years of experience, SGS provides ADME clinical studies and complete analysis with radiolabeled compounds.

SGS is your ideal partner in metabolite profiling and mass balance studies because of the following integrated capabilities:

- Analysis and clinical trials executed in collaboration with SGS’ own Phase I unit in Belgium
- Mass balance established in six to eight studies in humans per year
- Human metabolism studies by advanced LC-MS/MS and other technologies (QTrap, QToF, TopCount)
- Liquid scintillation counting and sample combustion for radiocarbon determination (mass balance studies)
- Determination of protein binding
- HPLC with on-line radio detector and MS for metabolic profiling
- Analysis of unlabeled drug by LC-MS/MS

MICRODOSING

SGS has performed microdosing studies at its clinical unit, and collaborates for the analytical portion with a selected partner that provides the highly sensitive analytics (accelerator mass spectrometry) required to quantitate the radioactivity in the biological samples derived from these clinical studies.
In addition to biopharmaceutical analytical testing, SGS provides Clinical Research Services that include:

- Trial Monitoring and Management (Phase I to IV)
- Clinical Pharmacology
- Regulatory/Medical Affairs and Pharmacovigilance
- Data Analysis and Reporting
- PK/PD Modeling and Simulation Services

SGS’ Clinical Research Services makes for fast and smooth testing and allow you to make ‘go / no go’ decisions about your product that are based on the most reliable data.

SGS can also serve as a one-stop, comprehensive service partner to facilitate development of your biological product. Such products not only contain a large molecule active ingredient but also small molecule components such as formulation additives, stabilizers, and other excipients. To complement our biopharmaceutical analytical testing capability, SGS can leverage its extensive expertise and experience in services for:

- Quality Control
- Analytical Chemistry
- Microbiological Testing
- Container Testing
- Extractables & Leachables Testing*
- Stability Studies *
- Preformulation
- Method Development and Validation
- Utilities Qualification & Monitoring (Gas, Air, Water)
- Medical Device Testing

*Outlined in detail in the following pages.
The assessment of extractables and leachables in bio/pharmaceutical products is an important step in drug product development. Processing equipment, as well as, primary and secondary container closures are potential vectors for chemical contaminants, and potential product degradation.

Monomer and polymer additives such as antioxidants, plasticisers, stabilizers, dyes, metal catalysts and other harmful chemicals may potentially migrate into the product under storage conditions. SGS provides a complete service for testing extractables in container materials and leachables in final products. These tests are conducted in cGMP compliant laboratories using technologies that detect ultra trace levels.

**SERVICES**
- Test strategy planning and data evaluation based on the available information
- Development of a tailored study design for extractables and leachables
- Extractables profiling (inorganic and organic extractables)
- Sequential extractions and alternative extraction techniques for isolating extractables in container materials
- Characterization of extractables by chromatographic and spectroscopic investigations
- Determination of the Analytical Estimation Threshold (AET)
- Calculation of the Qualification Threshold based on Safety Concern Threshold (SCT)
- Method development and validation of potential leachables in pharmaceutical products
- Performing of leachables studies on pharmaceutical products
- Reporting and evaluation of results within the current guidelines

**TECHNOLOGIES**
- HPLC-MS/MS, HPLC Q-Tof, HPLC-UV, DAD
- HS-GC, HS-GC-MS
- GC (FID, ECD, FID-NP), GC-MS
- GC-TEA (nitrosamines)
- ICP-OES, ICP-MS, AAS, IR
- FTIR
- TGA, DCS
- X-ray fluorescence analysis
- ASE (accelerated solvent extraction)
- Soxhlet
STABILITY STUDIES

From study design to storage, monitoring, analytical testing and documentation, SGS offers services to your complete satisfaction. With more than 20 years of experience and currently more than 100,000 samples in storage, SGS has the skills and the capacity to handle your stability projects. Globally SGS can provide its customers with the complete bandwidth of storage conditions in numerous climatic walk-in chambers and climatic cabinets with a total storage capacity of over 1,600 m³. Various refrigerators and freezers are available for storage at lower temperatures. All storage chambers are fully controlled with 24h/7d monitoring and alert systems (21 CFR part 11 compliant). For your safety, SGS operates back-up chambers for complete sample retrieval.

SERVICES

- Support in designing studies for real-time, stress tests and photo stability studies
- Development and validation of “stability indicating methods”
- Examination of stability-relevant parameters
- Storage and management of stability samples
- Interim reports for every testing period
- Comprehensive final report

CLIMATIC ZONES (ACCORDING TO ICH & WHO)

I. 21°C / 45% r.h.
II. 25°C / 60% r.h.
III. 30°C / 35% r.h.
IVa. 30°C / 65% r.h.
IVb. 30°C/75% r.h.

LONG-TERM, INTERMEDIATE AND ACCELERATED STORAGE

- 25°C / 40% r.h. (semi-permeable container study)
- 25°C / 60% r.h.
- 30°C / 65% r.h.
- 30°C / 70% r.h.
- 30°C / 75% r.h.
- 40°C / 75% r.h.
- 40°C / not more than (NMT) 25% r.h.
- +5°C
- −20°C
- −80°C
- Photostability
- Transport stability (freeze and thaw, cycle test)
- In-use stability
- Customer-specific conditions
Just as LIFE derives its source from nature’s flora and fauna, the life science industry finds its inspiration in nature for past and future discoveries. In our industry’s constantly evolving world of science and regulation, finding creative solutions is mandatory for success. We find common ground with our clients being Inspired by Life from drug discovery to market to solve these daily challenges in order to save lives. With a strong focus on bio-logics, the SGS life science mission is to safeguard the quality of medicines by providing professional and independent services in clinical research, pharmaceutical development, biologics characterization and quality control testing of pharmaceuticals, biopharmaceuticals and medical devices – thereby creating value for our clients, employees, shareholders and patients worldwide.
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