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# Surface characterization of gold nanoparticles for nanomaterial specific toxicity screening: FT-IR method





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# Surface characterization of gold nanoparticles for nanomaterial specific toxicity screening: FT-IR method

*Caractérisation de surface des nanoparticules d'or pour criblage de toxicité spécifique de nanomatériaux — Méthode FT-IR*



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# Foreword

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Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO/TS 14101 was prepared by Technical Committee ISO/TC 229, *Nanotechnologies*.

# Introduction

Gold nanoparticles (AuNPs) can be controlled with regard to size, shape and surface ligands, making them ideal for the study of relationships between their physicochemical properties and cytotoxicity on living bodies[1][2][3]. Among the various properties of AuNPs, surface ligand characteristics, such as the chemical composition, molecular structure and quantity of bound molecules, were found to play an important role in determining the behaviour of AuNPs, e.g. the degree of aggregation or agglomeration in solution, binding with biomolecules in cell culture media and cytotoxicity to living cells[4][5][6][7][8][9][10][11][12]. On the other hand, surface ligand modification is not always successful in the synthesis step, and the degree of ligand exchange should be identified prior to the property specific cytotoxicity test of AuNPs in order to obtain reliable and consistent results.

FT-IR (Fourier transform infrared) absorption spectroscopy is one of the most useful tools for NP surface ligand identification and quantification. By using the FT-IR method, the structures and relative quantities of ligand molecules bound to NP surfaces can be analysed[13][14][15][16][17][18][19][20]. However, the low concentrations and aqueous environment of synthesized AuNPs will complicate the interpretation of measurement results. Low concentrations of AuNPs result in small absorbance values, which can easily be influenced by background noise or the absorbance of trace impurities. Since cytotoxicity tests are performed in aqueous environments, we should analyse what is on the surface of AuNP in aqueous solutions if we want to study the effect of the surface characteristics on cytotoxicity of AuNPs. However, water molecules strongly absorb IR light over a wide frequency range, disabling IR absorption analysis on the solutes in very low concentrations. It is necessary to develop measurement guidelines by which the above issues can be minimized. In this project, we seek to develop a Technical Specification (TS) for the observation of chemical moieties bound to the synthetic AuNPs in the form of dehydrated films, which can deliver the information about the molecular species bound to AuNPs when they were in aqueous solutions. Although the standardization of FT-IR measurement procedures will be the basis for this Technical Specification, a great deal of weight will also be given to the sample preparation procedure for correct FT-IR analysis.

# Surface characterization of gold nanoparticles for nanomaterial specific toxicity screening: FT-IR method

## 1 Scope

This Technical Specification provides guidelines for the identification of the surface bound molecules using FT-IR of dehydrated gold nanoparticle (AuNPs) films both before and after nanomaterial (NM) cytotoxicity testing.

NOTE 1 AuNPs may have surface bound ligands prior to testing and may be additionally covered (or coated) with organic- or bio-molecules during the cytotoxicity test.

NOTE 2 Nucleic acids, amino acids, lipids or membrane components binding to AuNPs can be observed by FT-IR spectroscopy by detection of absorption bands corresponding to phosphodiester, amine or lipid, respectively, although the type of nucleic acids, proteins or lipid cannot be identified in detail based on IR spectra.

## 2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendment) applies.

ISO/TS 27687, *Nanotechnologies — Terminology and definitions for nano-objects — Nanoparticle, nanofibre and nanoplate*

## 3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO/TS 27687 and the following apply.

**3.1**
**attenuated total reflection mode**
**ATR Mode**
instrumental mode of operation in which the incident angle of IR light on the crystal is adjusted to be higher than the critical angle

NOTE The light is completely reflected by the upper surface of the crystal, and the intensity of the light is attenuated through absorption by materials covering the upper surface of the crystal. The frequency of IR light absorbed is used to identify the absorbed chemical moiety, and the fraction of light that is absorbed is used to quantitate the amount of that moiety present.

**3.2**
**dialysis**
process by which small molecules or ions diffuse through a membrane, thus causing their separation from larger molecules in solution and from suspended matter

[ISO 6107-2:2006, definition 38]

**3.3**
**Fourier transform infrared spectroscopy**
**FT-IR**
analytical chemical technique based on absorption of infrared radiation by chemical moieties in the specimen, used to identify and quantitate the absorbing chemical moieties



 