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(54) Title: FOURIER DOMAIN LOW-COHERENCE INTERFEROMETRY FOR LIGHT SCATTERING SPECTROSCOPY APPARATUS AND METHOD

(57) Abstract: An apparatus and method for obtaining depth-resolved spectra for the purpose of determining the size of scatterers by measuring their elastic scattering properties. Depth resolution is achieved by using a white light source in a Michelson interferometer and dispersing a mixed signal and reference fields. The measured spectrum is Fourier transformed to obtain an axial spatial cross-correlation between the signal and reference fields with near 1 μm depth-resolution. The spectral dependence of scattering by the sample is determined by windowing the spectrum to measure the scattering amplitude as a function of wavenumber.



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FOURIER DOMAIN LOW-COHERENCE INTERFEROMETRY FOR LIGHT SCATTERING SPECTROSCOPY APPARATUS AND METHOD

BACKGROUND OF THE INVENTION

1. Field of the Invention

The present invention relates to an apparatus and method for obtaining depth-resolved spectra for the purpose of determining structure by measuring elastic scattering properties. More particularly, Fourier domain, low-coherence interferometry techniques are applied to light scattering spectroscopy. This approach permits the viewing and recovery of depth-resolved structures, as well as obtaining spectroscopic information about scattered light as a function of depth.

2. Background of the Related Art

Accurately measuring small objects or other physical phenomena is a goal that is pursued in many diverse fields of scientific endeavor. For example, in the study of cellular biology and cellular structures, light scattering spectroscopy (LSS) has received much attention recently as a means for probing cellular morphology and the diagnosing of dysplasia. The disclosures of the following references are incorporated by reference in their entirety:

Backman, V., V. Gopal, M. Kalashnikov, K. Badizadegan, R. Gurjar, A. Wax, I. Georgakoudi, M. Mueller, C.W. Boone, R.R. Dasari, and M.S. Feld, IEEE J. Sel. Top. Quantum Electron., 7(6): p. 887-893 (2001); Mourant, J.R., M. Canpolat, C. Brocker, O. Esponda-Ramos, T.M. Johnson, A. Matanock, K. Stetter, and J.P. Freyer, J. Biomed. Opt., 5(2): p. 131-137 (2000); Wax, A., C. Yang, V. Backman, K. Badizadegan, C.W. Boone, R.R. Dasari, and M.S. Feld, Biophysical Journal, 82: p. 2256-2264 (2002);

Georgakoudi, I., E.E. Sheets, M.G. Müller, V. Backman, C.P. Crum, K. Badizadegan, R.R. Dasari, and M.S. Feld, *Am J Obstet Gynecol*, **186**: p. 374-382 (2002); Backman, V., M.B. Wallace, L.T. Perelman, J.T. Arendt, R. Gurjar, M.G. Muller, Q. Zhang, G. Zonios, E. Kline, T. McGillican, S. Shapshay, T. Valdez, K. Badizadegan, J.M. Crawford, M. Fitzmaurice, S. Kabani, H.S. Levin, M. Seiler, R.R. Dasari, I. Itzkan, J. Van Dam, and M.S. Feld, *Nature*, **406**(6791): p. 35-36 (2000); Wax, A., C. Yang, M. Mueller, R. Nines, C.W. Boone, V.E. Steele, G.D. Stoner, R.R. Dasari, and M.S. Feld, *Cancer Res*, (accepted for publication).

The LSS technique examines variations in the elastic scattering properties of cell organelles to infer their sizes and other dimensional information. In order to measure cellular features in tissues and other cellular structures, it is necessary to distinguish the singly scattered light from diffuse light, which has been multiply scattered and no longer carries easily accessible information about the scattering objects. This distinction or differentiation can be accomplished in several ways, such as the application of a polarization grating, by restricting or limiting studies and analysis to weakly scattering samples, or by using modeling to remove the diffuse component (s).

As an alternative approach for selectively detecting singly scattered light from sub-surface sites, low-coherence interferometry (LCI) has also been explored as a method of LSS. Experimental results have shown that using a broadband light source and its second harmonic allows the recovery of information about elastic scattering using LCI [7].

More recently, angle-resolved LCI (a/LCI) has demonstrated the capability of obtaining structural information by examining the angular distribution of scattered light from the sample or object under examination. The a/LCI technique has been successfully applied to measuring

cellular morphology and to diagnosing intraepithelial neoplasia in an animal model of carcinogenesis.

The above references are incorporated by reference herein where appropriate for appropriate teachings of additional or alternative details, features and/or technical background.

5

SUMMARY OF THE INVENTION

The claimed exemplary embodiments of the present invention address some of the issues presented above.

An object of the invention is to solve at least the above problems and/or disadvantages and to provide at least the advantages described hereinafter.

In one exemplary embodiment of the present invention, an apparatus comprises a first receiver that receives a first reference light and outputs a second reference light. A second receiver that receives a first sample light and outputs a second sample light and wherein the second sample light contains light scattered from a sample when at least a portion of the first sample light is scattered from a sample. A cross-correlator that receives and cross-correlates the second reference light with the second sample light. The cross-correlator may be a spatial cross-correlator.

In another exemplary embodiment of the present invention, a reference arm receives a first reference light and outputs a second reference light. A sample receives a first sample light and outputs a second sample light and wherein the second sample light contains light scattered from the sample when at least a portion of said first sample light is scattered from the sample. A spatial cross-correlator receives and cross correlates the second reference light with the second sample light. The spatial cross-correlator comprises a detector and a processing unit. The

detector outputs an interference term to the processing unit. The processing unit processes the interference term to yield depth resolved cross-correlation reflection profiles of the sample. The processing unit first applies a Gaussian window and then a Fourier transform transforms the interference term to yield depth resolved cross-correlation reflection profiles of the sample. The Fourier transform obtains an axial spatial cross-correlation between a signal field(s) and a reference field(s). A light source outputs light, which contains the first sample light and the first reference light.

In another exemplary embodiment of the present invention, a method comprises receiving a first reference light and outputting a second reference light. A first sample light is received and a second sample light is output. The second sample light contains light scattered from a sample when at least a portion of the first sample light is scattered from a sample along with the reception and cross correlation of the second reference light with the second sample light.

In another exemplary embodiment, a method comprises receiving light and splitting at least a portion of the light into reference light and sample light. At least a portion of said reference light is reflected from a reference surface to yield reflected reference light. At least a portion of the sample light is scattered from a sample to yield scattered sample light, and the scattered sample and the reflected reference light are mixed. Information is recovered about the scattered sample light. The mixing comprises detecting an intensity of the scattered sample light and the reflected reference light. Recovering information comprises extracting an interference term from a total intensity. Recovering information can further comprise applying a mathematical operator to the interference term to recover the spectral information about the scattered sample light at a particular depth to yield depth resolved cross-correlation reflection points of the sample. The mathematical operator used is preferably a Gaussian window.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention will be described in detail with reference to the following drawings in which like reference numerals refer to like elements wherein:

5 Figure 1A is a diagram of an exemplary embodiment of an fLCI system;

Figure 1B is a diagram of another exemplary embodiment of an fLCI system using fiber optic coupling;

Figure 2 is a diagram illustrating exemplary properties of a white light source;

10 Figure 3 is a diagram of an exemplary axial spatial cross-correlation function for a coverslip sample;

Figure 4 is a diagram of exemplary spectra obtained for front and back surfaces of a coverglass sample when no microspheres are present;

Figure 5 is a diagram of exemplary spectra obtained for front and back surfaces of a coverglass sample when microspheres are present;

15 Figure 6 is a diagram of exemplary ratios of spectra in Figures 4 and 5 illustrating scattering efficiency of spheres for front and back surface reflections;

Figure 7 is a diagram of a generalized version of the system shown in Figure 1;

Figure 8 is a block diagram of an exemplary embodiment of a method in accordance with the present invention; and

20 Figure 9 is a block diagram of another exemplary embodiment of a method in accordance with the present invention.

DETAILED DESCRIPTION OF PREFERRED EMBODIMENTS

In the following detailed description of the various exemplary embodiments, reference is made to the accompanying drawings that show, by way of illustration, specific embodiments in which the invention may be practiced. In the drawings, like numerals describe substantially similar components throughout the several views. These embodiments are described in sufficient detail to enable those skilled in the art to practice the invention. Other embodiments may be utilized without departing from the scope of the present invention. Moreover, it is to be understood that various embodiments of the invention, although different, are not necessarily mutually exclusive. For example, a particular feature, structure, or characteristic described in one embodiment may be included within other embodiments. Therefore, the following detailed description is not to be taken in a limiting sense. The scope of the present invention is delineated by the claims, along with the full scope of equivalents to which such claims are entitled.

The contents of the following references are incorporated by reference in their entirety:

Wojtkowski, M., A. Kowalczyk, R. Leitgeb, and A.F. Fercher, Opt. Lett., **27**(16): p. 1415-1417 (2002); Wojtkowski, M., R. Leitgeb, A. Kowalczyk, T. Bajraszewski, and A.F. Fercher, J. Biomed. Opt., **7**(3): p. 457-463 (2002); Leitgeb, R., M. Wojtkowski, A. Kowalczyk, C.K. Hitzenberger, M. Sticker, and A.F. Fercher, Opt. Lett., **25**(11): p. 820-822 (2000).

In general, spectral radar makes use of techniques where depth-resolved structural information is recovered by applying a Fourier transform to the spectrum of two mixed fields. In fLCI, the aforementioned approach used in spectral radar applications is extended to recover not only depth-resolved structure, but also to obtain spectroscopic information about scattered light as a function of depth. The capabilities of fLCI enable extracting the size of polystyrene

beads in a sub-surface layer based on their light scattering spectrum. The apparatus and method according to exemplary embodiments of the invention can be applied to many different areas. One such area of application is to recover nuclear morphology of sub-surface cell layers.

One exemplary embodiment of the fLCI scheme is shown in Figure 1A. White light
5 from a Tungsten light source 100 (e.g. 6.5 W, Ocean Optics™) is coupled into a multimode fiber 101 (e.g. 200 μm core diameter). The output of the fiber 101 is collimated by an achromatic lens 102 to produce a beam 104 (e.g. a pencil beam 5 mm in diameter). The beam 104 is then forwarded to an fLCI system 10.

This illumination scheme achieves Kohler illumination in that the fiber acts as a field
10 stop, resulting in the proper alignment of incident or illuminating light and thereby achieving critical illumination of the sample. In the fLCI system 10, the white light beam is split by the beamsplitter 106 (BS) into a reference beam 105 and an input beam 107 to the sample 108. The light scattered by the sample 108 is recombined at the BS 106 with light reflected by the reference mirror 114 (M).

15 The reference beam 105 in conjunction with the reference mirror 114 forms a portion of a reference arm that receives a first reference light and outputs a second reference light. The input beam 107 and the sample 108 form a portion of a sample arm that receives a first sample light and outputs a second sample light.

Those skilled in the art will appreciate that the light beam can be split into a plurality of
20 reference beams and input beams (e.g. N reference beams and N input beams) without departing from the spirit and scope of the present invention. Further, the splitting of the beams may be accomplished with a beamsplitter or a fiber splitter in the case of an optical fiber implementation of and exemplary embodiment of the present invention.

In the exemplary embodiment of the present invention shown in Figure 1A, the combined beam is coupled into a multimode fiber 113 by an aspheric lens 110. Again, other coupling mechanisms or lens types and configurations may be used without departing from the spirit and scope of the present invention. The output of the fiber coincides with the input slit of
 5 a miniature spectrograph 112 (e.g. USB2000, Ocean Optics™), where the light is spectrally dispersed and detected.

The detected signal is linearly related to the intensity as a function of wavelength $I(\lambda)$, which can be related to the signal and reference fields (E_s , E_r) as:

$$\langle I(\lambda) \rangle = \langle |E_s(\lambda)|^2 \rangle + \langle |E_r(\lambda)|^2 \rangle + 2\text{Re} \langle E_s(\lambda) E_r^*(\lambda) \rangle \cos \phi \quad (1)$$

10 where ϕ is the phase difference between the two fields and $\langle \dots \rangle$ denotes an ensemble average. The interference term is extracted by measuring the intensity of the signal and reference beams independently and subtracting them from the total intensity.

The axial spatial cross-correlation function, $\Gamma_{SR}(z)$ between the sample and reference fields is obtained by rescaling the wavelength spectrum into a wavenumber ($k = 2\pi/\lambda$) spectrum
 15 then Fourier transforming:

$$\Gamma_{SR}(z) = \int dk e^{ikz} \langle E_s(k) E_r^*(k) \rangle \cos \phi. \quad (2)$$

This term is labeled as an axial spatial cross-correlation as it is related to the temporal or longitudinal coherence of the two fields.

Another exemplary embodiment of an fLCI scheme is shown in Figure 1B. In this
 20 exemplary embodiment, fiber optic cable is used to connect the various components. Those skilled in the art will appreciate that other optical coupling mechanisms, or combinations

thereof, may be used to connect the components without departing from the spirit and scope of the present invention.

In Figure 1B, white light from a Tungsten light source 120 is coupled into a multimode fiber 122 and the white light beam in the multimode fiber is split by the fiber splitter (FS) 124
5 into a reference fiber 125 and an sample fiber 127 to the sample 130. The fiber splitter 124 is used to split light from one optical fiber source into multiple sources.

The reference light in reference fiber 125, in conjunction with a lens 126 (preferably an aspheric lens) and the reference mirror 128, forms a portion of a reference arm that receives a first reference light and outputs a second reference light. Specifically, reference light in reference
10 fiber 125 is directed to the reference mirror 128 by lens 126, and the reference light reflected by the reference mirror 128 (second reference light) is coupled back into the reference fiber 125 with lens 126. The sample light in sample fiber 127 and the sample 130 form a portion of a sample arm that receives a first sample light and outputs a second sample light. Specifically, sample light in sample fiber 127 is directed to the sample 130 by lens 131 (preferably as aspheric
15 lens), and at least a portion of the sample light scattered by the sample 130 is coupled into the sample fiber 127 by lens 131. In the exemplary embodiment shown in Figure 1B, the sample 130 is preferably spaced from lens 131 by a distance approximately equal to the focal length of lens 131.

At least a portion of the reflected reference light in reference fiber 125 and at least a
20 portion of the scattered sample light on sample fiber 127 are coupled into a detector fiber 133 by the FS 124.

The output of detector fiber 133 coincides with the input of a miniature spectrograph 132, where the light is spectrally dispersed and detected.

Figure 2 illustrates some of the properties of a white light source. Figure 2(a) illustrates an autocorrelation function showing a coherence length ($l_c = 1.2 \mu\text{m}$). Figure 2(a) shows the cross-correlation between the signal and reference fields when the sample is a mirror, and this mirror is identical to the reference mirror (M). In this exemplary scenario, the fields are identical and the autocorrelation is given by the transform of the incident field spectrum, modeled as a Gaussian spectrum with center wavenumber $k_0 = 10.3 \mu\text{m}^{-1}$ and $1/e$ width $\Delta k_{1/e} = 2.04 \mu\text{m}^{-1}$ (Figure 2 (b)).

Figure 2(b) shows an exemplary spectrum of light source that can be used in accordance with the present invention.

From this autocorrelation, the coherence length of the field, $l_c = 1.21 \mu\text{m}$ is determined. This is slightly larger than the calculated width of $l_c = 2/\Delta k_{1/e} = 0.98 \mu\text{m}$, with any discrepancy most likely attributed to uncompensated dispersion effects. Note that rescaling the field into wavenumber space is a nonlinear process which can skew the spectrum if not properly executed [13].

In data processing, a fitting algorithm is applied (e.g. a cubic spline fit) to the rescaled wavenumber spectrum and then resampled (e.g. resample with even spacing). The resampled spectrum is then Fourier transformed to yield the spatial correlation of the sample. Those skilled in the art will appreciate that other frequency based algorithms or combinations of algorithms can be used in place of the Fourier transform to yield spatial correlation. One example of a software tool that can be used to accomplish this processing in real time or near real time is to use LabView™ software.

In one exemplary embodiment of the present invention, the sample consists of a glass coverslip (e.g., thickness, $d \sim 200 \mu\text{m}$) with polystyrene beads which have been dried from suspension onto the back surface ($1.55 \mu\text{m}$ mean diameter, 3% variance). Thus, the field scattered by the sample can be expressed as:

$$E_s(k) = E_{\text{front}}(k) e^{ik\delta z} + E_{\text{back}}(k) e^{ik(\delta z + nd)} \quad (3)$$

In equation 3, E_{front} and E_{back} denote the field scattered by the front and back surfaces of the coverslip, and δz is the difference between the path length of the reference beam and that of the light reflected from the front surface and n the index of refraction of the glass. The effect of the microspheres will appear in the E_{back} term as the beads are small and attached closely to the back surface. Upon substituting equation 3 into equation 2, a two peak distribution with the width of the peaks given by the coherence length of the source is obtained.

In order to obtain spectroscopic information, a Gaussian window is applied to the interference term before performing the Fourier transform operation. Those skilled in the art will appreciate that other probabilistic windowing methodologies may be applied without departing from the spirit and scope of the invention. This makes it possible to recover spectral information about light scattered at a particular depth.

The windowed interference term takes the form:

$$\langle E_s(k) E_r^*(k) \rangle \exp[-((k - k_w) / \Delta k_w)^2]. \quad (4)$$

The proper sizing of a windowed interference term can facilitate the processing operation. For example, by selecting a relatively narrow window (Δk_w small) compared to the features of E_s and E_k , we effectively obtain $\langle E_s(k_w) E_r^*(k_w) \rangle$. In processing the data below, we use $\Delta k_w = 0.12 \mu\text{m}^{-1}$ which degrades the coherence length by a factor of 16.7. This exemplary

window setting enables the scattering at 50 different wavenumbers over the $6 \mu\text{m}^{-1}$ span of usable spectrum.

In Figure 3, an axial spatial cross-correlation function for a coverslip sample is showed according to one embodiment of the invention. Figures 3(a) and (b) shows the depth resolved cross-correlation reflection profiles of the coverslip sample before and after the processing operations. In Figure 3(a), a high resolution scan with arrows indicating a peak corresponding to each glass surface is shown. In Figure 3(b), a low resolution scan is obtained from the scan in Figure 3(a) is shown by using a Gaussian window.

Note that the correlation function is symmetric about $z = 0$, resulting in a superposed mirror image of the scan. Since these are represented as cross-correlation functions, the plots are symmetric about $z = 0$. Thus the front surface reflection for $z > 0$ is paired with the back surface reflection for $z < 0$, and vice versa.

In Figure 3(a), the reflection from the coverslip introduces dispersion relative to the reflection from the reference arm, generating multiple peaks in the reflection profile. When the spectroscopic window is applied, only a single peak is seen for each surface, however several dropouts appear due to aliasing of the signal.

To obtain the spectrum of the scattered light, we repeatedly apply the Gaussian window and increase the center wavenumber by $0.12 \mu\text{m}^{-1}$ between successive applications. As mentioned above, $\Delta k_w = 0.12 \mu\text{m}^{-1}$ is used to degrade the coherence length by a factor of 16.7. This results in the generation of a spectroscopic depth-resolved reflection profile.

Figures 4 (a) and (b) show the spectrum obtained for light scattered from the front (a) and back (b) surfaces of a coverglass sample respectively, when no microspheres are present.

The reflection from the front surface appears as a slightly modulated version of the source spectrum. The spectrum of the reflection from the rear surface however has been significantly modified. Thus in equation 3, we now take $E_{front}(k) = E_s(k)$ and $E_{back}(k) = T(k) E_s(k)$, where $T(k)$ represents the transmission through the coverslip.

5 In Figure 5, the spectra for light scattering obtained for front (a) and back (b) surfaces of a coverglass sample when microspheres are present on the back surface of the coverslip are shown in Figures 5 (a) and (b). It can be seen that the reflected spectrum from the front surface has not changed significantly, as expected. However, the spectrum for the back surface is now modulated. We can examine the scattering properties $S(k)$ of the microspheres by writing the
10 scattered field as $E_{spheres}(k) = S(k) T(k) E_s(k)$ and taking the ratio $E_{spheres}(k) / E_{back}(k) = S(k)$, which is shown as a solid line in Figure 6(a). It can be seen from this ratio that the microspheres induce a periodic modulation of the spectrum.

In Figure 6 (a), a ratio of the spectra found in Figure 4 and Figure 5 is shown. This illustrates the scattering efficiency of spheres for front (represented by the dashed line) and back
15 (represented by the solid line) surface reflections. In Figure 6(b), a correlation function obtained from ratio of back surface reflections is shown. The peak occurs at the round trip optical path through individual microspheres, permitting the size of the spheres to be determined with sub-wavelength accuracy.

For comparison, the same ratio for the front surface reflections (dashed line in Figure
20 6(a)) shows only a small linear variation. Taking the Fourier transform of $S(k)$ yields a clear correlation peak (Figure 6(b)), at a physical distance of $z = 5.24 \mu\text{m}$. This can be related to the optical path length through the sphere by $z = 2n/$ with the index of the microspheres $n = 1.59$.

The diameter of the microspheres to be $l = 1.65 \mu\text{m} \pm 0.33 \mu\text{m}$, with the uncertainty given by the correlation pixel size. Thus with fLCI, we are able to determine the size of the microspheres with sub-wavelength accuracy, even exceeding the resolution achievable with this white light source and related art LCI imaging.

5 There are many applications of the various exemplary embodiments of the present invention. One exemplary application of fLCI is in determining the size of cell organelles, in particular the cell nucleus, in epithelial tissues. In biological media, for example, the relative refractive indices are lower for organelles compared to microspheres and thus, smaller scattering signals are expected. The use of a higher power light source will permit the smaller signals to be
10 detected. Other examples include detection of sub-surface defects in manufactured parts, including fabricated integrated circuits, detection of airborne aerosols, such as nerve agents or biotoxins, and detection of exposure to such aerosols by examining epithelial tissues within the respiratory tract.

 Additionally, the larger the size of the nucleus (compared to the microspheres in this
15 experiment), the higher the frequency modulation of the spectrum. Those skilled in the art will appreciate that higher frequency oscillations are detected at a lower efficiency in Fourier transform spectroscopy techniques. Therefore, in order to detect these higher frequency oscillations, a higher resolution spectrograph is used.

 Figure 7 illustrates a generalized embodiment of the fLCI system shown in Figure 1 and
20 discussed in greater detail above. In Figure 7, a light source 700 (e.g. a multi-wavelength light) is coupled into an fLCI system 702. Within the fLCI system 702, a sample portion 704 and a reference portion 706 are located. The sample portion 704 includes a light beam and light scattered from a sample. For example, the sample portion 704 may include a sample holder, a

free space optical arm, or an optical fiber. The reference portion 706 includes a light beam and light that is reflected from a reference. For example, the reference portion 706 may include an optical mirror. A cross-correlator 708 receives and cross-correlates light from the sample with light from the reference.

5 Figure 8 illustrates another exemplary embodiment of the present invention. In Figure 8, a method is disclosed where a first reference light is received 800 and a second reference light is output 802. A first sample light is received 804 and a second sample light is output 806. The second sample light contains light scattered from a sample when at least a portion of the first sample light is scattered from a sample. The second reference light with the second sample light
10 are received and cross-correlated 808.

 Figure 9 illustrates another exemplary embodiment of the present invention. In Figure 9, a method is disclosed where light is received 900 from a sample that has been illuminated. At least a portion of the light is split into reference light and sample light 902. At least a portion of said reference light is reflected from a reference surface to yield reflected reference light 904. At
15 least a portion of the sample light is scattered from a sample to yield scattered sample light 906. The scattered sample light and the reflected reference light are mixed 908. Spectral information is recovered about the scattered sample light 910.

 The foregoing example illustrates how the exemplary embodiments of the present invention can be modified in various manners to improve performance in accordance with the
20 spirit and scope of the present invention.

 From the foregoing detailed description, it should be apparent that fLCI can recover structural information with sub-wavelength accuracy from sub-surface layers based on measuring elastic scattering properties. The simplicity of the system makes it an excellent candidate for

probing cellular morphology in tissue samples and may one day serve as the basis for a biomedical diagnostic device.

The foregoing embodiments and advantages are merely exemplary and are not to be construed as limiting the present invention. The present teaching can be readily applied to other
5 types of apparatuses. The description of the present invention is intended to be illustrative, and not to limit the scope of the claims. Many alternatives, modifications, and variations will be apparent to those skilled in the art. In the claims, means-plus-function clauses are intended to cover the structures described herein as performing the recited function and not only structural equivalents but also equivalent structures.

WHAT IS CLAIMED IS:

1. An apparatus for determining size information from a sample, comprising:
a first receiver that receives a first reference light and outputs a second reference
light;
5 a second receiver that receives a first sample light and outputs a second sample
light, wherein said second sample light contains light scattered from a sample; and
a cross-correlator that receives and cross correlates said second reference light
with said second sample light to yield depth resolved structure and spectroscopic information
about the light scattered from the sample as a function of depth.

10 2. The apparatus of claim 1, wherein the cross-correlator is a spatial
cross-correlator.

3. An apparatus for determining size information from a sample, comprising:
15 a reference arm portion that receives a first reference light and outputs a second
reference light;
a sample arm portion that receives a first sample light and outputs a second
sample light, wherein said second sample light contains light scattered from a sample; and
a spatial cross-correlator that receives and cross correlates said second reference
20 light with said second sample light to yield said size information.

4. The apparatus of claim 3, wherein said spatial cross-correlator
comprises a detector and a processing unit.

5. The apparatus of claim 4, wherein said detector outputs an interference term to said processing unit.

5 6. The apparatus of claim 5, wherein said processor unit processes said interference term to yield size information in the form of depth resolved cross-correlation reflection profiles of the sample.

7. The apparatus of claim 5, wherein said processor unit first applies a
10 Gaussian window or other window function and then Fourier transforms said interference term to yield size information in the form of depth resolved cross-correlation reflection profiles of the sample.

8. The apparatus of claim 7, wherein said Fourier transform obtains an
15 axial spatial cross-correlation between a signal field(s) and a reference field(s).

9. The apparatus of claim 3, further comprising a light source for outputting light which contains said first sample light and said first reference light.

20

10. The apparatus of claim 3, wherein the reference arm portion, the sample arm portion and the spatial cross-correlator are optically coupled with optical fiber.

11. The apparatus of claim 3, wherein a beamsplitter is used to separate a light beam
5 into a reference beam and an input beam.

12. The apparatus of claim 3, wherein a fiber splitter is used to separate a light beam into a reference beam and an input beam.

10 13. A method comprising:
receiving a first reference light and outputting a second reference light;
receiving a first sample light and outputting a second sample light and wherein
said second light contains light scattered from a sample when at least a portion of said first
sample light is scattered from a sample; and
15 receiving and cross correlating said second reference light with said second sample
light to yield depth resolved structure and spectroscopic information about the light scattered
from the sample as a function of depth.

14. A method for determining size information from a sample, comprising:
20 receiving light and splitting at least a portion of said light into reference light and
sample light;
reflecting at least a portion of said reference light from a reference surface to yield
reflected reference light;

scattering at least a portion of said sample light from a sample to yield scattered sample light;

mixing said scattered sample and said reflected reference light; and

recovering said size information from said scattered sample light and said reflected

5 reference light.

15. The method of claim 14, wherein said mixing comprises detecting an intensity of said scattered sample light and said reflected reference light.

10 16. The method of claim 14, wherein said recovering comprises extracting an interference term from a total intensity.

17. The method of claim 14, wherein said recovering step comprises applying a mathematical operator to said interference term to recover said spectral information about said scattered sample light at a particular depth to yield depth resolved cross-correlation reflection points of the sample.

18. The method of claim 17, wherein said mathematical operator is a Gaussian or other window.

20 19. An apparatus for determining a size of objects on a substrate having a first substrate surface and a second substrate surface, said objects on one of the first or second surface of the substrate, comprising:

a reference arm portion that receives a first reference light and outputs a second reference light;

a sample arm portion that receives a first sample light and outputs a second sample light, wherein said second sample light contains light scattered from the first surface of the substrate and light scattered from the second surface of the substrate;

a detector that detects the second reference light and the second sample light and outputs a detected signal; and

a processor unit coupled to the detector that receives the detected signal and determines the size of the objects using a first portion of the signal due to the light scattered from the first surface of the substrate and a second portion of the signal due to light scattered from the second surface of the substrate.

20. The apparatus of claim 19, wherein said processor unit determines the size of the objects by determining a ratio of the first portion of the signal due to the light scattered from the first surface of the substrate and the second portion of the signal due to the light scattered from the second surface of the substrate.

21. The apparatus of claim 19, wherein said processor unit determines the size of the objects by determining a scattering property of the objects using the first portion of the signal due to the light scattered from the first surface of the substrate and the second portion of the signal due to the light scattered from the second surface of the substrate.

22. The apparatus of claim 19, wherein the wavelength of the first sample light and first reference light is greater than the size of objects.

23. The apparatus of claim 19, wherein the wavelength of the first sample light and first reference light is greater than the size of objects and the objects include biological structures.

24. The apparatus of claim 19, wherein the wavelength of the first sample light and first reference light is greater than the size of the objects and the objects include cell organelles.

25. An apparatus for determining scattering properties of objects on a substrate having a first substrate surface and a second substrate surface, said objects on one of the first or second surface of the substrate, comprising:

a reference arm proportion that receives a first reference light and outputs a second reference light;

a sample arm portion that receives a first sample light and outputs a second sample light, wherein said second sample light contains light scattered from the first surface of the substrate, light scattered from the second surface of the substrate, and light scattered from the objects;

a detector that detects the second reference light and the second sample light and outputs a detected signal; and

a processor unit coupled to the detector that receives the detected signal and determines the scattering properties of the objects using a first portion of the signal due to the

light scattered from the first surface of the substrate and a second portion of the signal due to light scattered from the second portion of the substrate.

26. The apparatus of claim 25, wherein said processor unit determines the scattering
5 properties of the objects by determining a ratio of the first portion of the signal due to the light scattered from the first surface of the substrate and the second portion of the signal due to the light scattered from the second surface of the substrate.

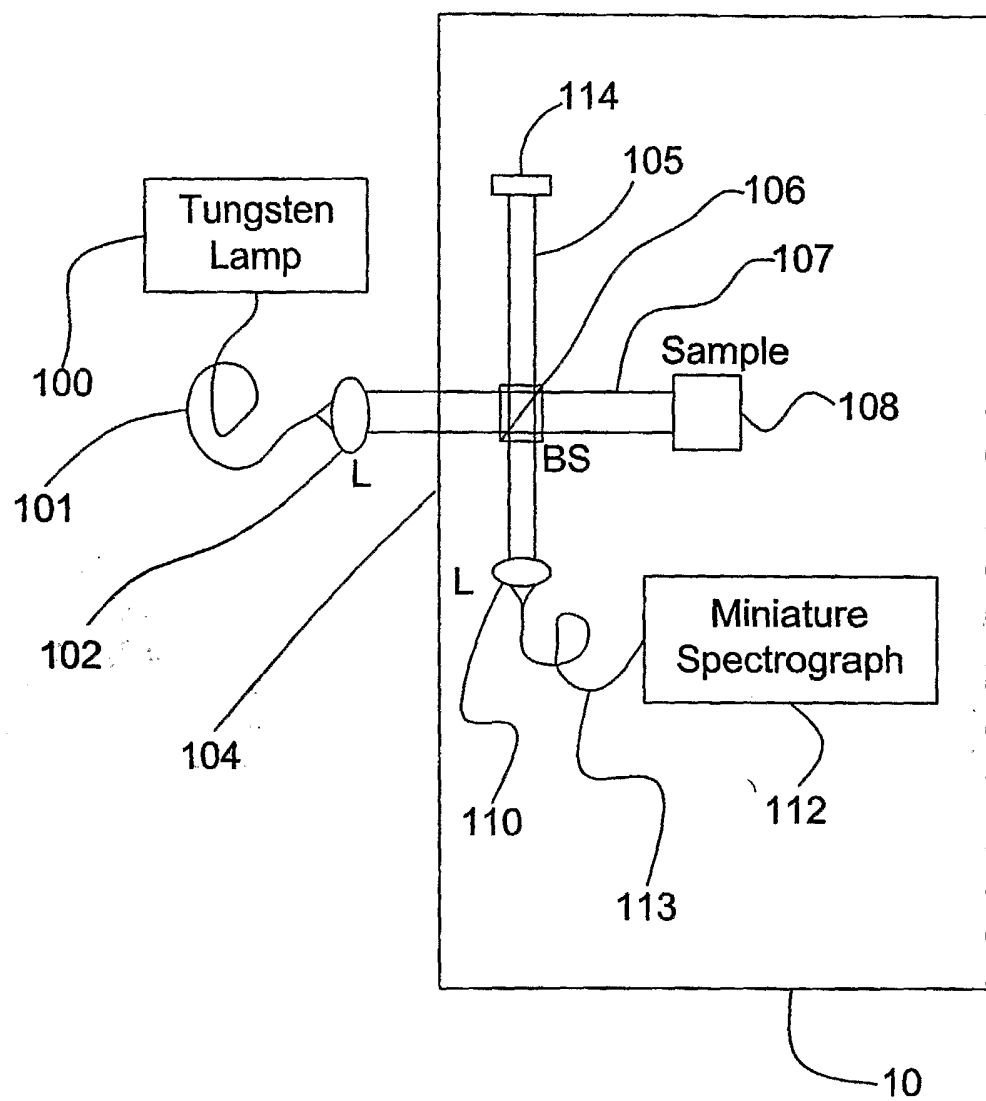


FIG. 1A

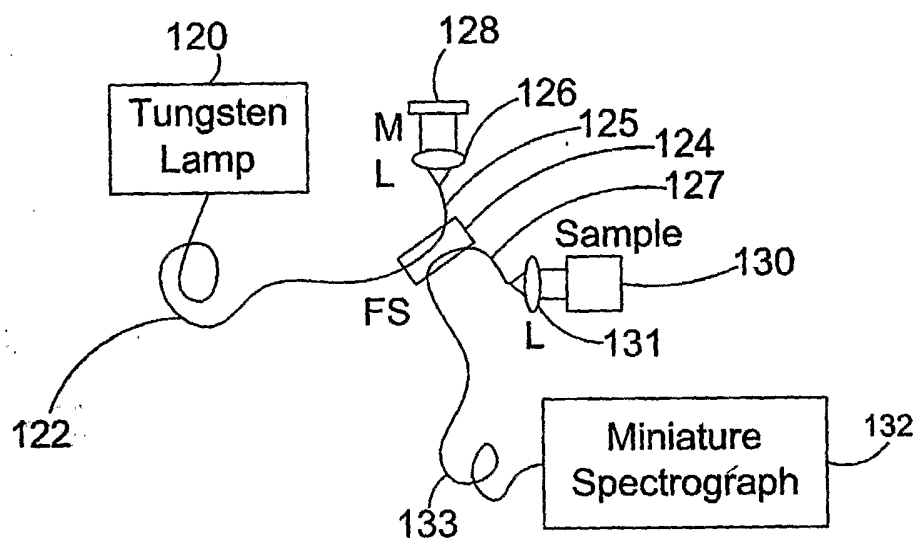


FIG. 1B

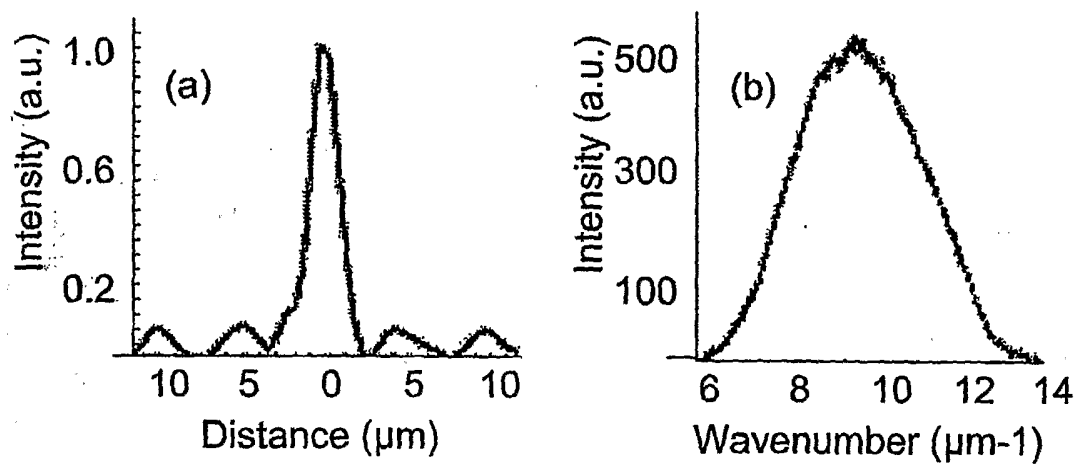


FIG. 2

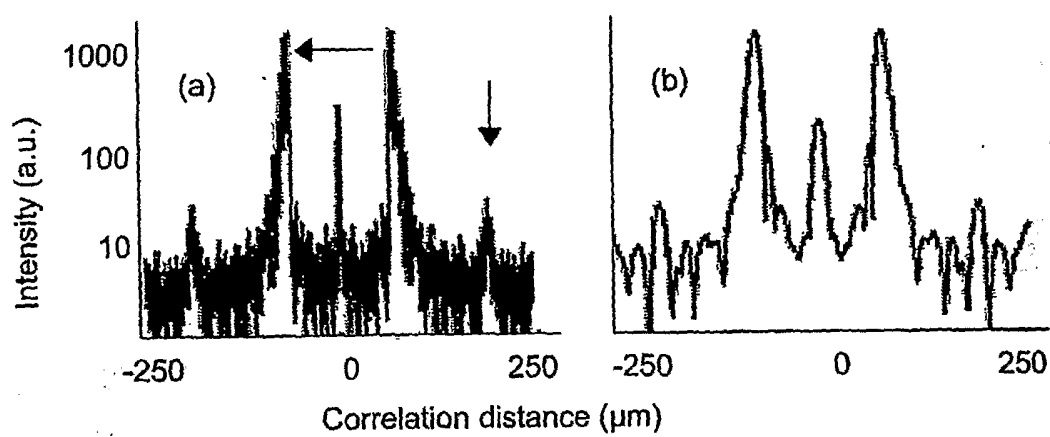


FIG. 3

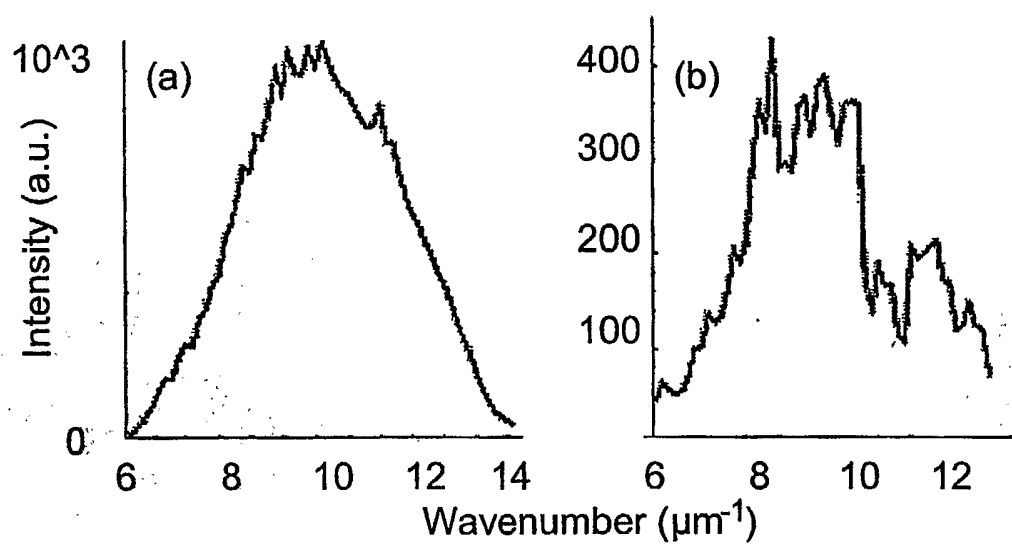


FIG. 4

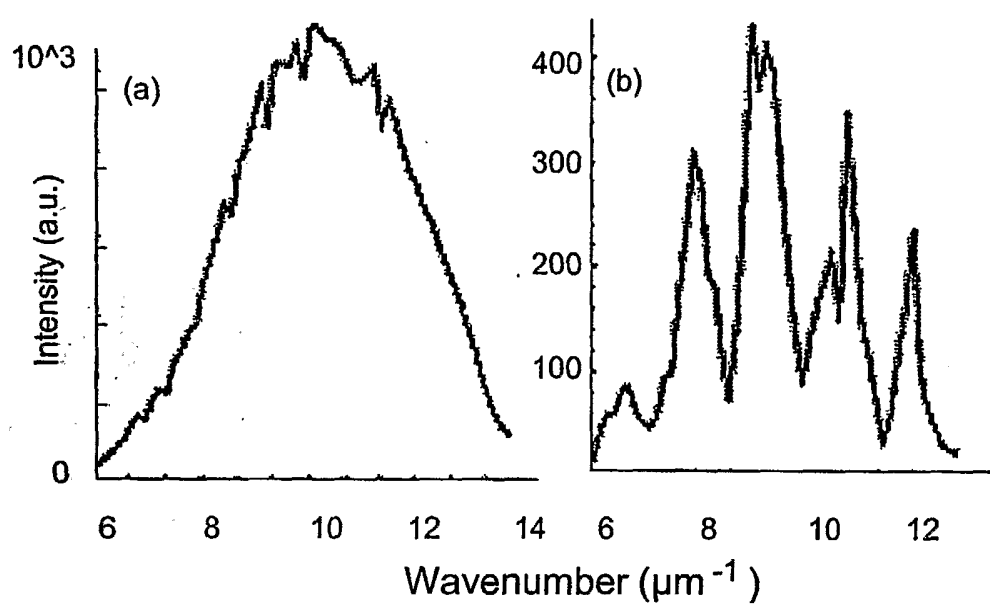


FIG. 5

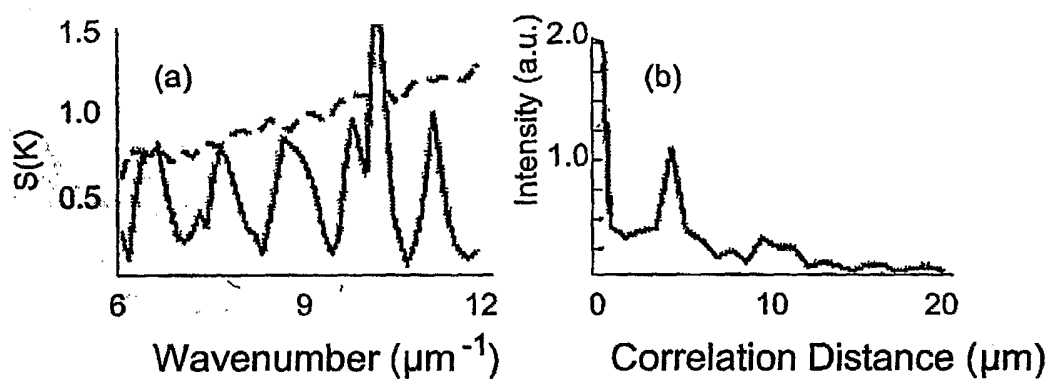


FIG. 6

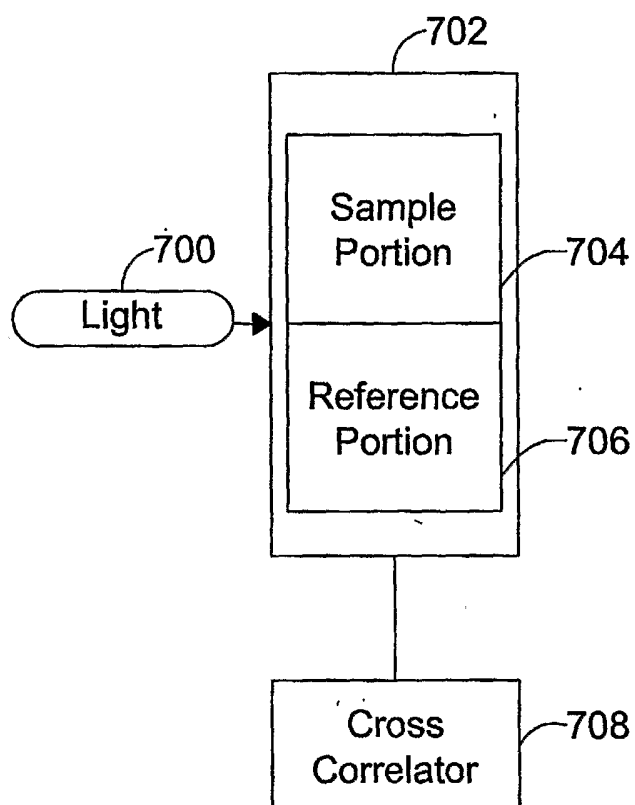


FIG. 7

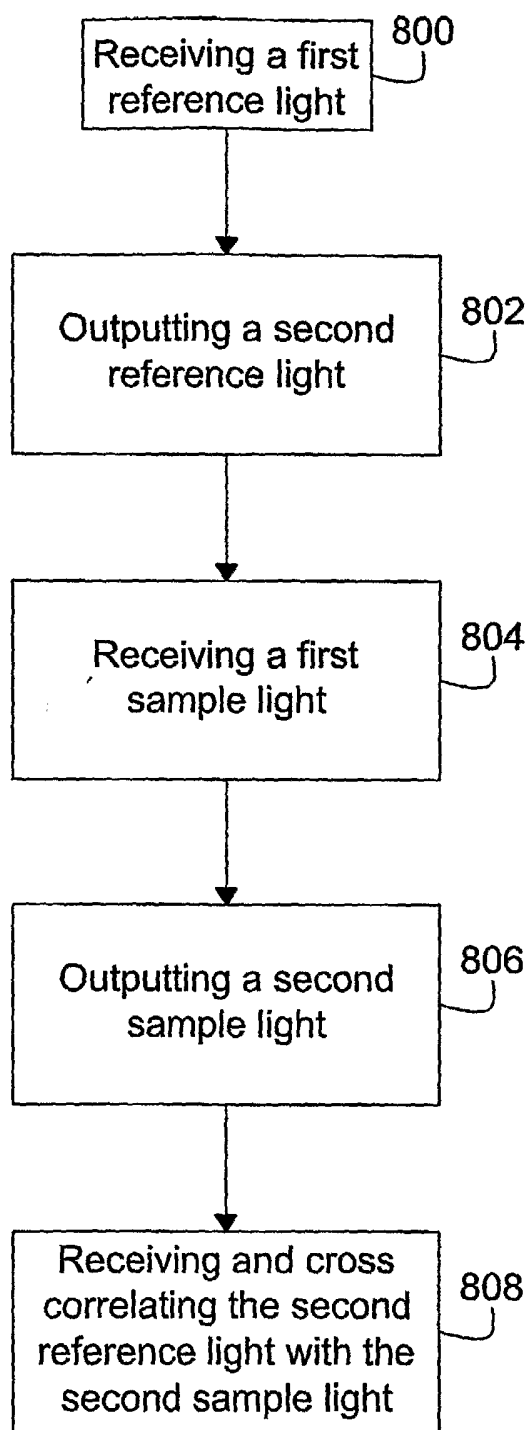


FIG. 8

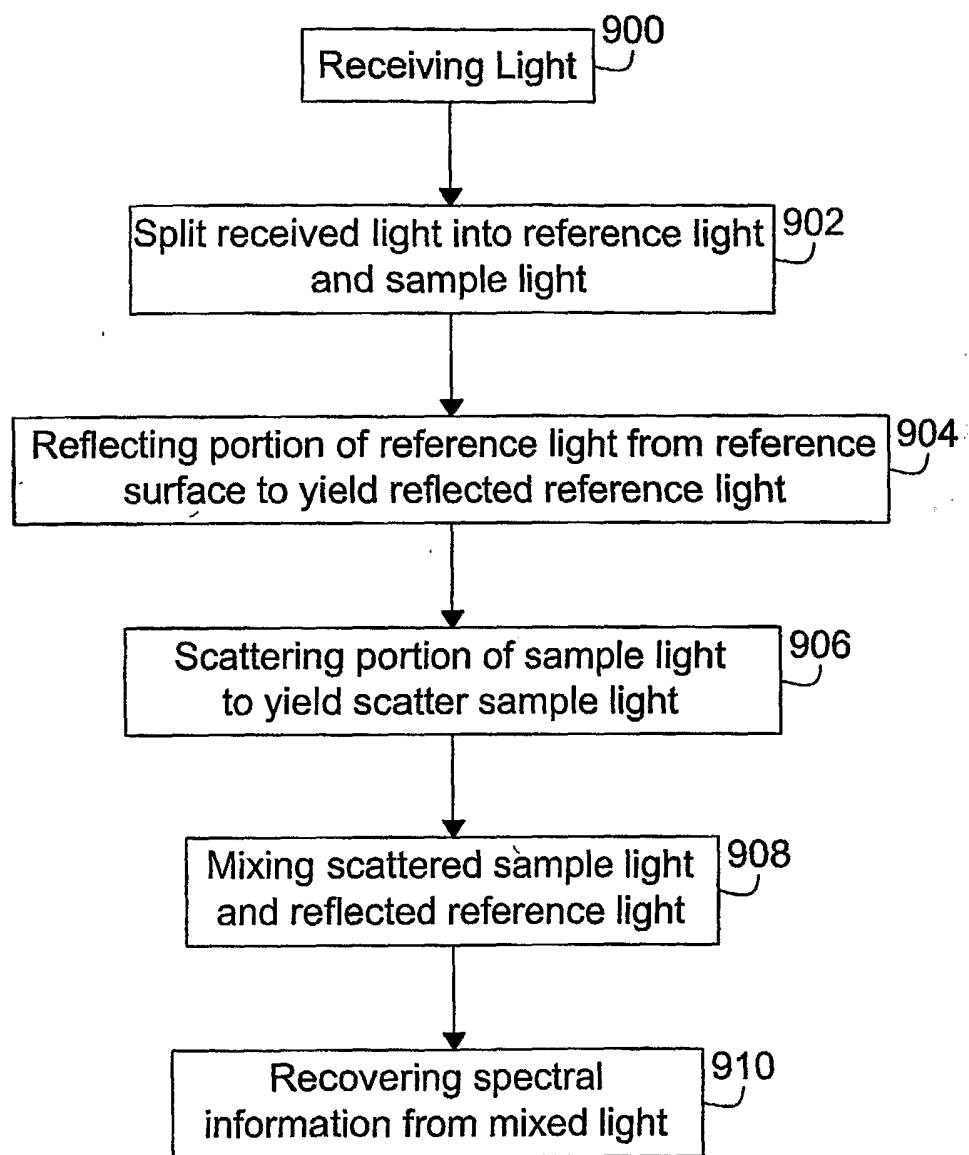


FIG. 9