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FLUORESCENCE SCANNING SYSTEM FOR ANALYTICAL ULTRACENTRIFUGATION

Abstract

The present disclosure provides various embodiments of a fluorescence scanning system having a sample holder with a sample suspended within that is rotated by a centrifuge such that the sample is illuminated at various angles by an excitation beam by operation of a galvanometer and such that the sample emits a fluorescence emission that is detected through a narrow window of exposure defined along the travel path of rotation taken by the sample holder when rotated by the centrifuge. A stationary fluorescence detector is in operative communication with the sample holder along the narrow window of exposure for detecting the fluorescence emissions emitted by the sample from the sample holder while also separating the excitation beam from the fluorescence emissions.

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Background/Summary

CROSS-REFERENCE TO RELATED APPLICATIONS [0001] This application is a continuation of U.S. Non-Provisional patent application Ser. No. 17/192,623, entitled “FLUORESCENCE SCANNING SYSTEM FOR ANALYTICAL ULTRACENTRIFUGATION,” and filed Mar. 4, 2021. U.S. Non-Provisional patent application Ser. No. 17/192,623 claims the benefit of priority of U.S. Provisional Application No. 62/985,222, entitled “FLUORESCENCE SCANNING SYSTEM FOR ANALYTICAL ULTRACENTRIFUGATION,” and filed Mar. 4, 2020. The entire contents of each of the above-cited applications are hereby incorporated by reference for all purposes.

FIELD
[0002] The present disclosure relates to a fluorescence detector. More particularly, the present disclosure relates to a fluorescence scanning system for analytical ultracentrifugation having a novel sample holder and scanning arrangement.
BACKGROUND
[0003] Analytical ultracentrifugation is a classical technique for the study of macromolecules and particulates suspended in a solution. An analytical ultracentrifuge (AUC) functions by measuring the radial concentration distribution of particles in real-time after application of a centrifugal field, and then mathematically analyzing the concentration distributions as they evolve over time. Samples in an AUC are enclosed in centrifuge holders with transparent windows, such that the radial concentration distributions are measured in real-time through synchronized axial optical systems based on interferometry, absorbance, or fluorescence scanners. Analytical ultracentrifugation occurs in a rotor chamber under high vacuum and with little space for optical elements; consequently, detection of fluorescence emissions from a sample can be difficult in an AUC due to space constraints. In view of the foregoing, there is a need for improved analytical ultracentrifugation systems and methods that enable detection of fluorescence emissions.
SUMMARY
[0004] The present disclosure provides an analytical ultracentrifuge (AUC) system including movable galvanometer mirrors that may be used to manipulate the spatial location (e.g., radial position of the sample volume) probed by an excitation beam in an analytical ultracentrifuge (AUC), thereby greatly simplifying integration of a fluorescence detector into an AUC system. In embodiments, the galvanometer mirror may change the angle of a collimated excitation beam, thereby modulating the position it probes in a rotating sample with the AUC system, thereby allowing sample to be radially scanned. In this regard, the excitation beam does not need to traverse the sample parallel to the axis of sample rotation, and deviations may be accounted for computationally during data analysis.
[0005] In an aspect, the disclosure provides a fluorescence scanning system that includes a centrifuge defining a chamber and a rotor extending laterally across the chamber, wherein the rotor is operable for rotation about an axis of the centrifuge; a sample holder engaged to the rotor for rotation of the sample holder along a travel path, the sample holder defining an inner chamber wherein a proximal plano-convex lens window is positioned at a first end of the sample holder and a distal plano-convex lens window is positioned at an opposite second end of the sample holder; a

sample disposed inside the inner chamber of the sample holder; an illumination source for generating and transmitting a collimated excitation beam; a galvanometer in operative association with the illumination source for scanning the collimated excitation beam at different angles along the sample disposed within the sample holder; a stationary mirror in operative communication with the galvanometer for transmitting the collimated excitation beam into the sample holder through the proximal plano-convex lens window and illuminating the sample to generate fluorescence emissions; and a stationary fluorescence detector in operative communication with the sample holder at a point along the travel path of rotation of the sample holder for detecting the fluorescence emissions and collimated excitation beam emitted from the sample holder.

[0006] In some embodiments, the stationary fluorescence detector comprises a dichroic mirror operable for separating the collimated excitation beam from the fluorescence emissions emitted from the sample holder such that the collimated excitation beam is deflected by the dichroic mirror and the fluorescence emissions passes through the dichroic mirror.

[0007] In some embodiments, the proximal plano-convex lens window is configured to focus the collimated excitation beam along different angles along the sample.

[0008] In some embodiments, the distal plano-convex lens window is configured to focus the fluorescence emissions emitted by the sample to the stationary fluorescence detector.

[0009] In some embodiments, the proximal plano-convex lens window and the distal plano-convex lens window comprise a sapphire lens.

[0010] In some embodiments, the proximal plano-convex lens window and the distal plano-convex lens window each comprise a convex portion that allows the excitation beam to have a more parallel orientation relative to the axis of rotation of the rotor of the centrifuge.

[0011] In some embodiments, the stationary mirror is oriented at a 45 degree angle relative to the galvanometer.

[0012] In some embodiments, the sample is suspended in a liquid within the inner chamber of the sample holder.

[0013] In some embodiments, the stationary fluorescence detector comprises a first detection lens for focusing the excitation beam and the operable for separating the excitation beam from the fluorescence emissions.

[0014] In some embodiments, the dichroic mirror is operable for deflecting the excitation beam off the dichroic mirror while allowing the fluorescence emissions to pass through the dichroic mirror.

[0015] In some embodiments, the proximal plano-convex lens window is configured to modulate the angle of entry of the excitation beam into the sample.

[0016] In some embodiments, the sample is scanned radially by the excitation beam.

[0017] In some embodiments, the centrifuge defines opposing apertures for permitting illumination of the sample by the excitation beam and detection of the fluorescence emissions along a one to three degree window defined along the travel path of the sample holder.

[0018] In some embodiments, the sample is exposed for illumination and detection in a 1 microsecond period of time along the travel path of the sample holder.

[0019] In some embodiments, the proximal plano-convex lens window defines a convex portion and an opposite plano portion and wherein the distal plano-convex lens window also defines a convex portion and an opposite plano portion, wherein the plano portions of the distal and proximal plano-convex lens windows communicate with the inner chamber of the sample holder and the convex portions of the distal and proximal plano-convex lens windows communicate with the exterior of the sample holder.

Description

DESCRIPTION OF THE DRAWINGS

[0020] The accompanying drawings, which are incorporated in and constitute a part of the specification, illustrate various example methods, and other example embodiments of various aspects of the invention. It will be appreciated that the illustrated element boundaries (e.g., boxes, groups of boxes, or other shapes) in the figures represent one example of the boundaries. Corresponding reference characters indicate corresponding elements among the view of the drawings. One of ordinary skill in the art will appreciate that in some examples one element may be designed as multiple elements or that multiple elements may be designed as one element. Furthermore, elements may not be drawn to scale.

[0021] FIG. **1** is a simplified illustration of an illustrative embodiment of the fluorescence scanning system showing the use of a galvanometer to scan the sample with an excitation beam.

[0022] FIG. **2** is a simplified illustration of the fluorescence scanning system of FIG. **1** showing the detection of the fluorescence emissions from the sample by a stationary fluorescence detector.

[0023] FIG. **3** is a photograph showing an illustrative prototype for testing the operation of the fluorescence scanning system.

[0024] FIG. **4** is a photograph showing an illustrative prototype in which the excitation beam is shown exiting the laser collimator, reflected on a mirror and onto a galvanometer to a final mirror positioned underneath the rotor of the ultracentrifuge.

DETAILED DESCRIPTION

[0025] The present disclosure is based, at least in part, on the discovery that movable galvanometer mirrors may be used to manipulate the spatial location (e.g., radial position of the sample volume) probed by an excitation beam in an analytical ultracentrifuge (AUC), thereby greatly simplifying integration of a fluorescence detector into an AUC system. In embodiments, the galvanometer mirror may change the angle of a collimated excitation beam, thereby modulating the position it probes in a rotating sample with the AUC system, thereby allowing sample to be radially scanned. In this regard, the excitation beam does not need to traverse the sample parallel to the axis of sample rotation, and deviations may be accounted for computationally during data analysis.

[0026] Fluorescence detection in analytical centrifugation has been shown to greatly extend the concentration range of analytical ultracentrifugation, thereby permitting novel applications to the characterization of ultra-high affinity protein interactions, such as antibody-antigen interactions. In addition, fluorescence detection in analytical centrifugation has been applied to applications related to the study of tracer proteins in highly concentrated solutions, which facilitates the characterization of protein pharmaceuticals in serum or formulation conditions. Since analytical ultracentrifugation occurs in a rotor chamber with high vacuum and little space for optical elements, it has been found that the use of mirrors to manipulate the spatial location (e.g., radial position of the sample volume) probed by the excitation beam greatly simplifies the detector design as disclosed herein.

[0027] In one aspect, the fluorescence scanning system includes a novel sample holder that rotates around the axis of centrifugation and includes a distal plano-convex lens window at one end of the sample holder for directing a plurality of excitation beams along a sample suspended within the chamber of the sample holder and an opposite proximal plano-convex lens window that focuses fluorescence emissions emitted by the sample after excitation to a stationary fluorescence detector for detection.

[0028] In another aspect, the sample within the sample holder is excited by the excitation beams and the fluorescence emissions emitted by the sample are detected along about a one to three degree window (e.g., one microsecond of exposure) as the sample holder is rotated along a 360 degree path by the centrifuge. In a further aspect, a single excitation beam generated by an illumination source is converted to a plurality of excitation beams by a galvanometer such that the sample is excited along different angles by the plurality of excitation beams as the sample holder is

rotated along the centrifugal pathway.

[0029] Referring to the drawings, embodiments of a fluorescence scanning system for analytical ultracentrifugation are illustrated and generally indicated as **100** in FIGS. **1**-**4**. As shown in FIGS. **1** and **2**, in some embodiments the fluorescence scanning system **100** includes a centrifuge **102** (FIG. **1**) having a chamber **119** that generally encompasses a sample holder **103**, a rotor **104**, and a 360 degree travel path **204**. The centrifuge **102** rotates the sample holder **103** by operation of the rotor **104** that rotates the sample holder **103** around the 360 degree travel path **204** about an axis **200** of the centrifuge **102** in a lateral direction. As shown in FIG. **1**, the sample holder **103** and the rotor **104** are within the chamber **119** of the centrifuge **102**. The rotor **104** extends laterally across the chamber **119**. The sample holder **103** is rotated within an enclosed chamber at about 60,000 rpms by the centrifuge **102**. In some embodiments the centrifuge **102** may define opposing apertures **120**A and **120**B at a point along the 360 degree travel path **204** such that the sample holder **103** passes by the aperture **120**A for excitation of a sample **108** and aperture **120**B for detection of the fluorescence emissions **118** as shall be discussed in greater detail below.

[0030] In addition, the fluorescence scanning system **100** includes a stationary fluorescence detector **101** for detection of fluorescence emissions **118** emitted by the sample **108** and detected through the sample holder **103**. In this arrangement, the stationary fluorescence detector **101** that detects the fluorescence emissions **118** and an illumination source **105** that excites the sample **108** are located outside the chamber **119** of the centrifuge **102** rather than being located within the limited confines of the chamber **119**. In addition, the stationary fluorescence detector **101** is in periodic communication with the sample holder **103** as it travels within the chamber **119** of the centrifuge **102** through the opposite apertures **120**A/**120**B. In some embodiments, the centrifuge **102** may be a Beckman Coulter ultracentrifuge having an analytical rotor (e.g., holes to receive the sample holder **103**).

[0031] In some embodiments, the illumination source **105** emits a collimated excitation beam **106**, for example a collimated laser beam, onto a galvanometer **107** that is operable for changing the angle of the collimated excitation beam **106**, thereby modulating the position of the collimated excitation beam **106** relative to the sample **108** within the sample holder **103** and allowing the sample **108** to be scanned radially as it is rotated within the centrifuge **102**. In this arrangement, the collimated excitation beam **106** does not need to traverse the sample **108** parallel to the axis **200** of the sample **108** rotation about the centrifuge **102**. In this manner, deviations can be computationally accounted for in the data interpretation by a processor **128** in operative communication with the fluorescence scanning system. As shown, the collimated excitation beam **106** is reflected off a stationary mirror **112** and through the aperture **120** A such that the sample **108** becomes illuminated and excited as the sample holder **103** travels over the aperture **120** A of the centrifuge **102**.

[0032] As shown, the sample holder **103** defines a chamber **111** having a proximal plano-convex lens window **110** at one end of the chamber **111** and a distal plano-convex lens window **109** at the opposite end of the chamber **111**. In some embodiments, the convex portion of the proximal plano-convex lens window **110** is formed along the exterior of the sample holder **103** in communication with the chamber **111** while the plano portion of the proximal plano-convex lens window **110** is formed along the interior of the sample holder **103**. Similarly, the convex portion of the distal plano-convex lens window **109** is formed along the exterior of the sample holder **103** in communication with the opposite side of the chamber **111**, while the plano portion of the proximal plano-convex lens window **109** is formed along the interior of the sample holder **103**. In one aspect, the slightly curved configuration of the convex portion of the proximal plano-convex lens window **109** allows the excitation beams **106** to have a more parallel orientation relative to the axis **200** of the rotation of the centrifuge **102** for the rotor **104**. In some embodiments, both the distal and proximal plano-convex lens windows **109** and **110** may be sapphire lenses that act as windows into the chamber **111** of the sample holder **103** to provide a seal for the sample **108** in the chamber **111** and to let the collimated excitation beams **106** enter the chamber **111** to sufficiently illuminate

the sample **108** as well as permit the fluorescence emissions **118** emitted by the sample **108** to exit the chamber **111** at the correct orientation for detection by the stationary fluorescence detector **101**.
[0033] In some embodiments, the stationary fluorescence detector **101** includes a first detector lens **113** for focusing the fluorescence emissions **118** emitted from the sample holder **103** by the sample **108** through a dichroic mirror **114** before passing through laser line filter **115**. The fluorescence emissions **118** are then focused onto a photomultiplier tube **117** by a second detector lens **116** for detection as shown in FIG. **2**. In addition, as shown in FIG. **1**, the stationary fluorescence detector **101** separates the excitation beams **106** from the fluorescence emissions **118** that both emit from the sample holder **103** and pass through first detector lens **113**. As noted above, the fluorescence emissions **118** that pass through the first detector lens **113** also pass through the dichroic mirror **114**; however, the dichroic mirror **114** is operable to reflect the excitation beams **106** that pass through the first detector lens **113** such that only fluorescence emissions **118** are received by the photomultiplier tube **117**.
[0034] In some embodiments, a processor **128** is in operative communication with the centrifuge **102**, the fluorescence detector **101**, galvanometer **107**, and/or illumination source **105** for performing the functionalities of the fluorescence scanning system **100** related to sample illumination, fluorescence detecting, and sample imaging.
[0035] Referring to FIGS. **3** and **4**, a prototype was set up to test the efficacy of the fluorescence scanning system **100** in which the sample holder **103** was placed in a centrifuge **102** and an illumination source **105** transmits the excitation beam **106** to the sample holder **103** by an arrangement of the galvanometer **107** and stationary mirror **112** that scans the sample (not shown) for detection by the photomultiplier **117** in the fluorescence detector **101** (FIG. **3**).
REFERENCE NUMBERS
[0036] **100**. Fluorescence Scanning System [0037] **101**. Fluorescence Detector [0038] **102**. Centrifuge [0039] **103**. Sample Holder-Centrifuge [0040] **104**. Rotor-Centrifuge [0041] **105**. Illumination Source [0042] **106**. Excitation Beam [0043] **107**. Galvanometer [0044] **108**. Sample [0045] **109**. Distal Plano-Convex Lens Window-Sample Holder [0046] **110**. Proximal Plano-Convex Lens Window-Sample Holder [0047] **111**. Chamber-Sample Holder [0048] **112**. Stationary Mirror [0049] **113**. First Detector Lens-Fluorescence Detector [0050] **114**. Dichroic Mirror-Fluorescence Detector [0051] **115**. Laser Line Filter-Fluorescence Detector [0052] **116**. Second Detector Lens-Fluorescence Detector [0053] **117**. Photomultiplier Tube-Fluorescence Detector [0054] **118**. Fluorescence Emissions [0055] **119**. Chamber Centrifuge [0056] **120** A/B. Apertures-Centrifuge [0057] **128**. Processor [0058] **200**. Axis of Centrifugal Rotation [0059] **204**. Travel Path
[0060] It should be understood from the foregoing that, while particular embodiments have been illustrated and described, various modifications can be made thereto without departing from the spirit and scope of the invention as will be apparent to those skilled in the art. Such changes and modifications are within the scope and teachings of this invention as defined in the claims appended hereto.
[0061] References to “one embodiment”, “an embodiment”, “one example”, and “an example” indicate that the embodiment(s) or example(s) so described may include a particular feature, structure, characteristic, property, element, or limitation, but that not every embodiment or example necessarily includes that particular feature, structure, characteristic, property, element or limitation. Furthermore, repeated use of the phrase “in one embodiment” does not necessarily refer to the same embodiment, though it may.
[0062] To the extent that the term “includes” or “including” is employed in the detailed description or the claims, it is intended to be inclusive in a manner similar to the term “comprising” as that term is interpreted when employed as a transitional word in a claim.
[0063] Throughout this specification and the claims that follow, unless the context requires otherwise, the words ‘comprise’ and ‘include’ and variations such as ‘comprising’ and ‘including’ will be understood to be terms of inclusion and not exclusion. For example, when such terms are

used to refer to a stated integer or group of integers, such terms do not imply the exclusion of any other integer or group of integers.
[0064] To the extent that the term “or” is employed in the detailed description or claims (e.g., A or B) it is intended to mean “A or B or both”. When the applicants intend to indicate “only A or B but not both” then the term “only A or B but not both” will be employed. Thus, use of the term “or” herein is the inclusive, and not the exclusive use. See, Bryan A. Garner, A Dictionary of Modern Legal Usage 724 (2d. Ed. 1995).
[0065] Ranges can be expressed herein as from “about” one particular value and/or to “about” another particular value. When such a range is expressed, another aspect includes from the one particular value and/or to the other particular value. Similarly, when values are expressed as approximations, by use of the antecedent “about,” it is understood that the particular value forms another aspect. It is further understood that the endpoints of each of the ranges are significant both in relation to the other endpoint, and independently of the other endpoint. It is also understood that there are a number of values disclosed herein, and that each value is also herein disclosed as “about” that particular value in addition to the value itself. It is also understood that throughout the application, data are provided in a number of different formats and that this data represent endpoints and starting points and ranges for any combination of the data points. For example, if a particular data point “10” and a particular data point “15” are disclosed, it is understood that greater than, greater than or equal to, less than, less than or equal to, and equal to 10 and 15 are considered disclosed as well as between 10 and 15. It is also understood that each unit between two particular units are also disclosed. For example, if 10 and 15 are disclosed, then 11, 12, 13, and 14 are also disclosed. In this regard, ranges provided herein are understood to be shorthand for all of the values within the range. For example, a range of 1 to 50 is understood to include any number, combination of numbers, or sub-range from the group consisting 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, or 50 as well as all intervening decimal values between the aforementioned integers such as, for example, 1.1, 1.2, 1.3, 1.4, 1.5, 1.6, 1.7, 1.8, and 1.9. With respect to sub-ranges, “nested sub-ranges” that extend from either end point of the range are specifically contemplated. For example, a nested sub-range of an exemplary range of 1 to 50 may comprise 1 to 10, 1 to 20, 1 to 30, and 1 to 40 in one direction, or 50 to 40, 50 to 30, 50 to 20, and 50 to 10 in the other direction.
[0066] While example systems, methods, and other embodiments have been illustrated by describing examples, and while the examples have been described in considerable detail, it is not the intention of the applicants to restrict or in any way limit the scope of the appended claims to such detail. It is, of course, not possible to describe every conceivable combination of components or methodologies for purposes of describing the systems, methods, and other embodiments described herein. Therefore, the invention is not limited to the specific details, the representative apparatus, and illustrative examples shown and described. Thus, this application is intended to embrace alterations, modifications, and variations that fall within the scope of the appended claims.

Claims

1. A fluorescence scanning system comprising: an illumination source configured to generate and transmit a collimated excitation beam; a galvanometer configured to be in operative association with the illumination source, the galvanometer configured to change an angle of the collimated excitation beam, thereby modulating a position of the collimated excitation beam relative to a sample disposed within a sample holder housed in a rotor of a centrifuge; a stationary mirror configured to be in operative communication with the galvanometer and configured to transmit the collimated excitation beam into the sample holder and illuminate the sample to generate fluorescence emissions; and a stationary fluorescence detector configured to be in operative

communication with the sample holder at a point along a travel path of rotation of the sample holder for detecting the fluorescence emissions.

2. The system of claim 1, further comprising the sample holder, wherein the sample holder comprises a proximal plano-convex lens window positioned at a first end of the sample holder and a distal plano-convex lens window positioned at an opposite second end of the sample holder.

3. The system of claim 2, wherein the proximal plano-convex lens window is configured to focus the collimated excitation beam along different angles along the sample.

4. The system of claim 2, wherein the distal plano-convex lens window is configured to focus the fluorescence emissions emitted by the sample to the stationary fluorescence detector.

5. The system of claim 2, wherein the proximal plano-convex lens window and the distal plano-convex lens window each comprise a sapphire lens.

6. The system of claim 2, wherein the proximal plano-convex lens window and the distal plano-convex lens window each comprise a convex portion that allows the collimated excitation beam to have a more parallel orientation relative to an axis of rotation of the rotor of the centrifuge.

7. The system of claim 2, wherein the proximal plano-convex lens window defines a convex portion and an opposite plano portion and wherein the distal plano-convex lens window also defines a convex portion and an opposite plano portion, wherein the plano portions of the distal and proximal plano-convex lens windows communicate with the inner sample holder chamber of the sample holder and the convex portions of the distal and proximal plano-convex lens windows communicate with the exterior of the sample holder.

8. The system of claim 2, wherein the proximal plano-convex lens window is configured to modulate an angle of entry of the collimated excitation beam into the sample.

9. The system of claim 1, wherein the stationary fluorescence detector comprises a dichroic mirror configured to separate the collimated excitation beam from the fluorescence emissions emitted from the sample holder such that the collimated excitation beam is deflected by the dichroic mirror and the fluorescence emissions pass through the dichroic mirror.

10. The system of claim 9, wherein the stationary fluorescence detector further comprises a first detection lens for focusing the collimated excitation beam through the dichroic mirror.

11. The system of claim 1, wherein the stationary mirror is oriented at a 45 degree angle relative to the galvanometer.

12. The system of claim 1, further comprising the centrifuge, wherein the rotor is operable for rotation about an axis of the centrifuge in order to rotate the sample holder along the travel path, and wherein the centrifuge defines opposing apertures for permitting illumination of the sample by the collimated excitation beam and detection of the fluorescence emissions along a one to three degree window defined along the travel path of the sample holder.

13. A sample holder defining a chamber configured to house a sample, the sample holder comprising: a proximal plano-convex lens window positioned at a first end of the chamber of the sample holder and a distal plano-convex lens window positioned at an opposite second end of chamber of the sample holder, the proximal plano-convex lens window configured to direct one or more excitation beams into the sample within the chamber of the sample holder and the distal plano-convex lens window configured to focus fluorescence emissions emitted by the sample after excitation to a stationary fluorescence detector for detection.

14. The system of claim 13, wherein the proximal plano-convex lens window and the distal plano-convex lens window each comprise a sapphire lens.

15. The system of claim 13, wherein the proximal plano-convex lens window defines a convex portion and an opposite plano portion and wherein the distal plano-convex lens window also defines a convex portion and an opposite plano portion, wherein the plano portions of the distal and proximal plano-convex lens windows communicate with the chamber of the sample holder and the convex portions of the distal and proximal plano-convex lens windows communicate with an exterior of the sample holder.

16. A method for a fluorescence scanning system, comprising: directing a single excitation beam generated by an illumination source to a galvanometer; converting the single excitation beam to a plurality of excitation beams by the galvanometer; directing the plurality of excitation beams to a sample housed within a sample holder as the sample holder is rotated by a centrifuge rotor, such that the sample within the sample holder is excited along different angles by the plurality of excitation beams as the sample holder is rotated; and focusing fluorescence emissions emitted by the sample after excitation to a stationary fluorescence detector for detection.

17. The method of claim 16, wherein directing the plurality of excitation beams to the sample housed within a sample holder as the sample holder is rotated by the centrifuge rotor comprises directing the plurality of excitation beams to the sample via a proximal plano-convex lens positioned at a first end of the sample holder.

18. The method of claim 17, wherein focusing fluorescence emissions emitted by the sample after excitation to the stationary fluorescence detector for detection comprises focusing fluorescence emissions emitted by the sample after excitation via a distal plano-convex lens positioned at a second end of the sample holder, opposite the first end.