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(54) TEMPERATURE CONTROL FOR LIGHT-EMITTING DIODE STABILIZATION

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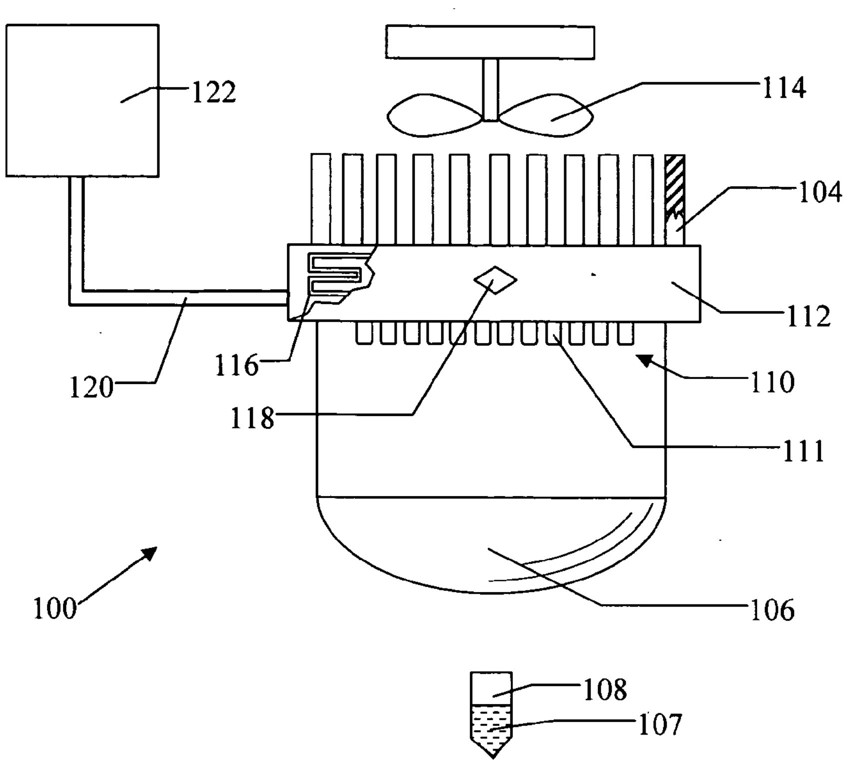
(57) **ABSTRACT**

A system is provided that includes a light-emitting diode (LED); a temperature sensor in thermal contact with the LED and capable of measuring an operating temperature and generating an operating temperature signal; and a temperature regulating system capable of receiving the operating temperature signal and regulating the operating temperature based on the operating temperature signal. A method for stabilizing the temperature of an LED is provided. A method is provided that includes providing a system comprising an LED, a reaction region, and a sample in the reaction region; generating excitation beams with the LED; directing excitation beams to the sample; detecting an optical property of the sample to obtain detection data; measuring the operating temperature of the light emitting diode; and adjusting the detection data of an excitation beam characteristic shift related to the operating temperature, when the LED is operated at the operating temperature to generate the excitation beams.



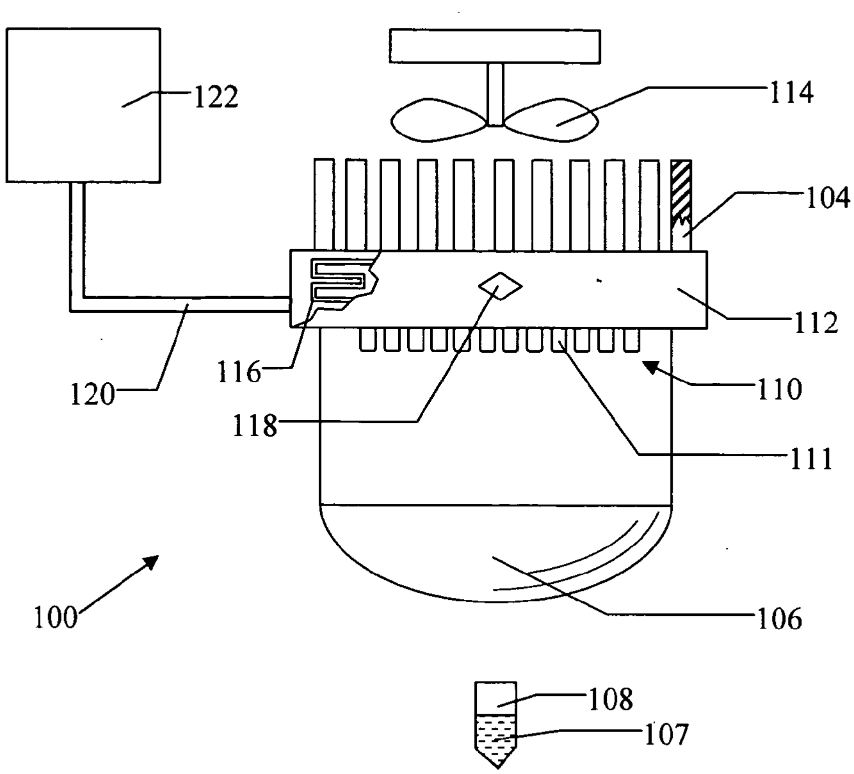

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FIG. 1

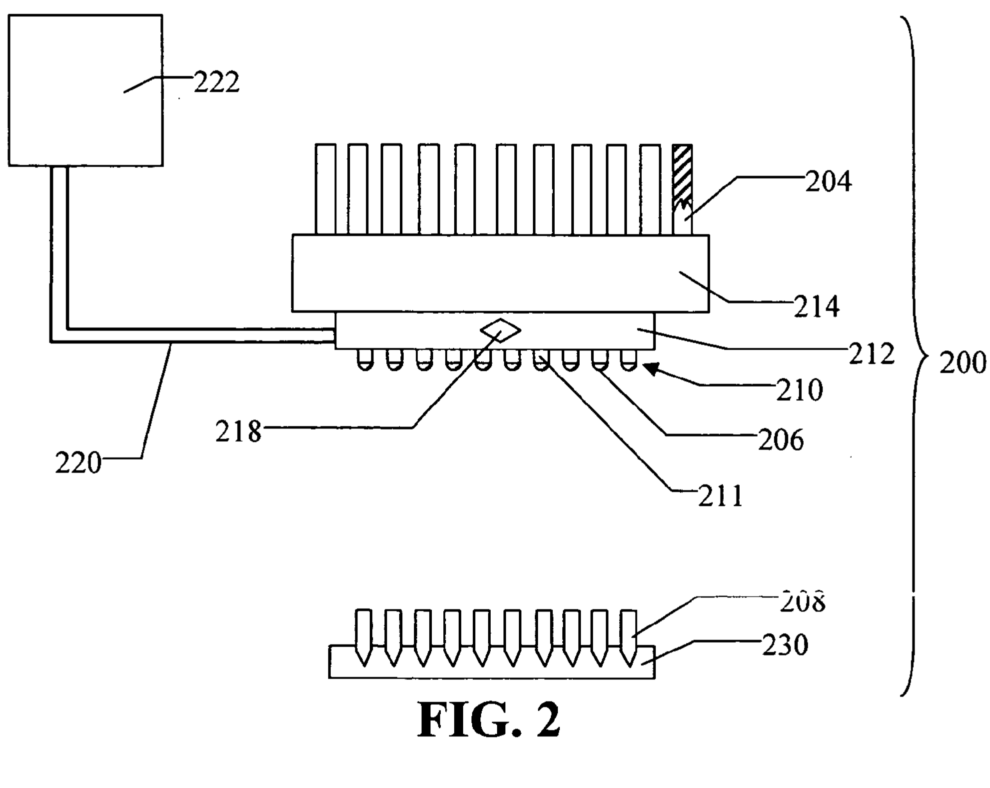
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FIG. 2

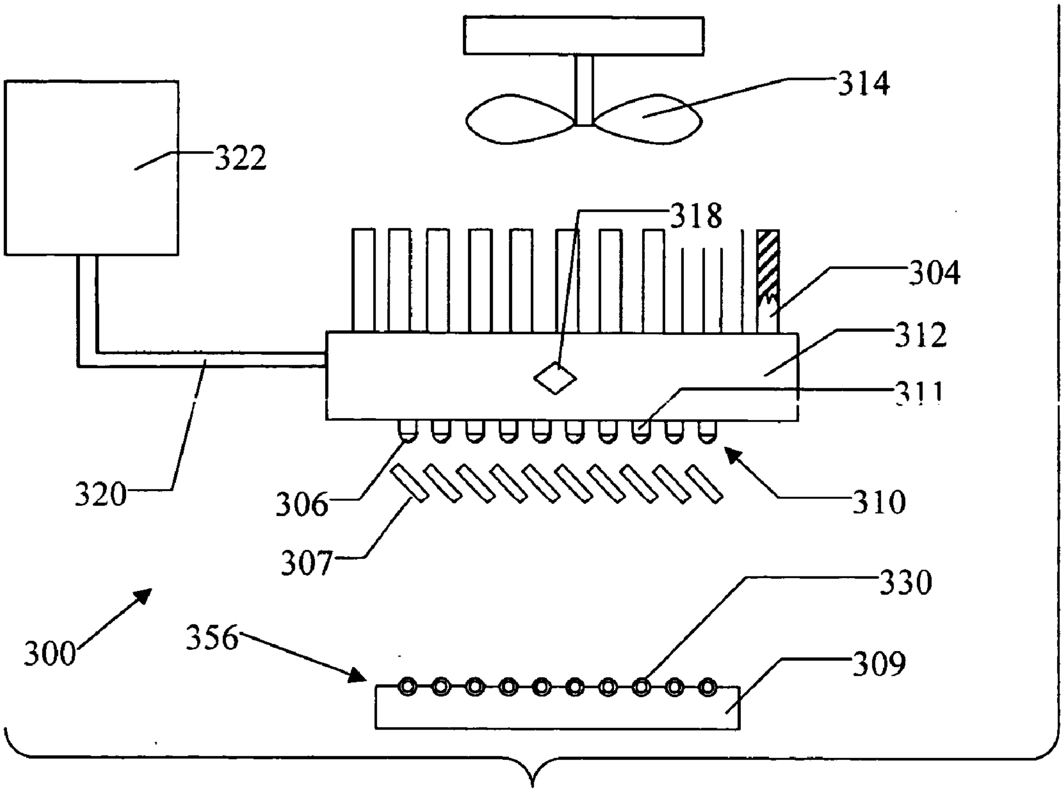
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FIG. 3a

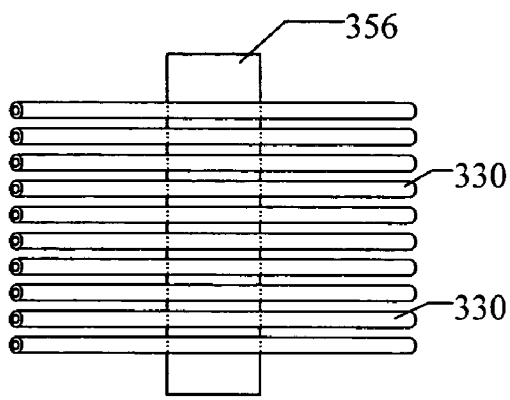
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FIG. 3b

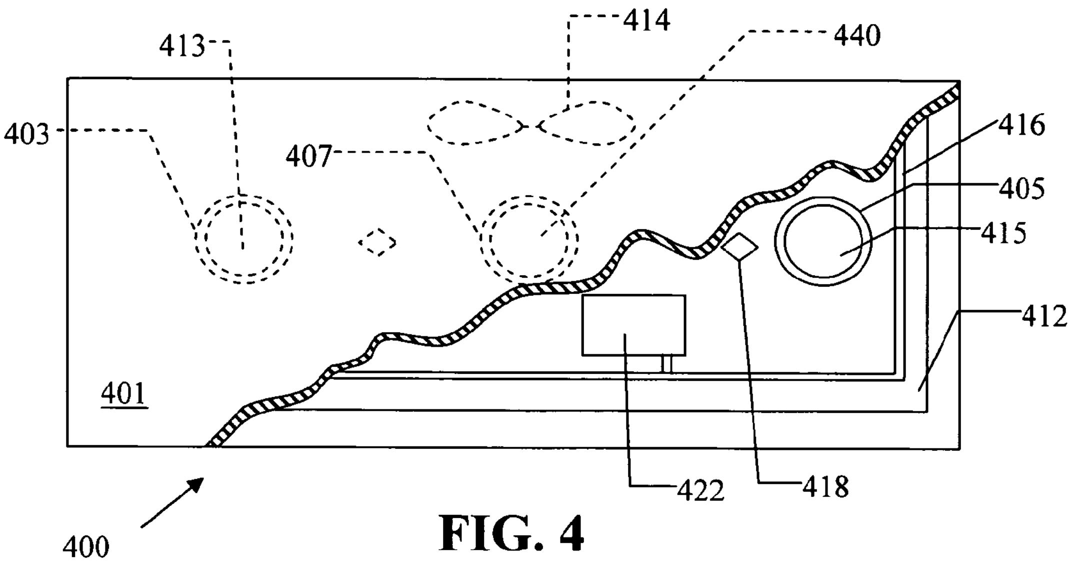
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FIG. 4

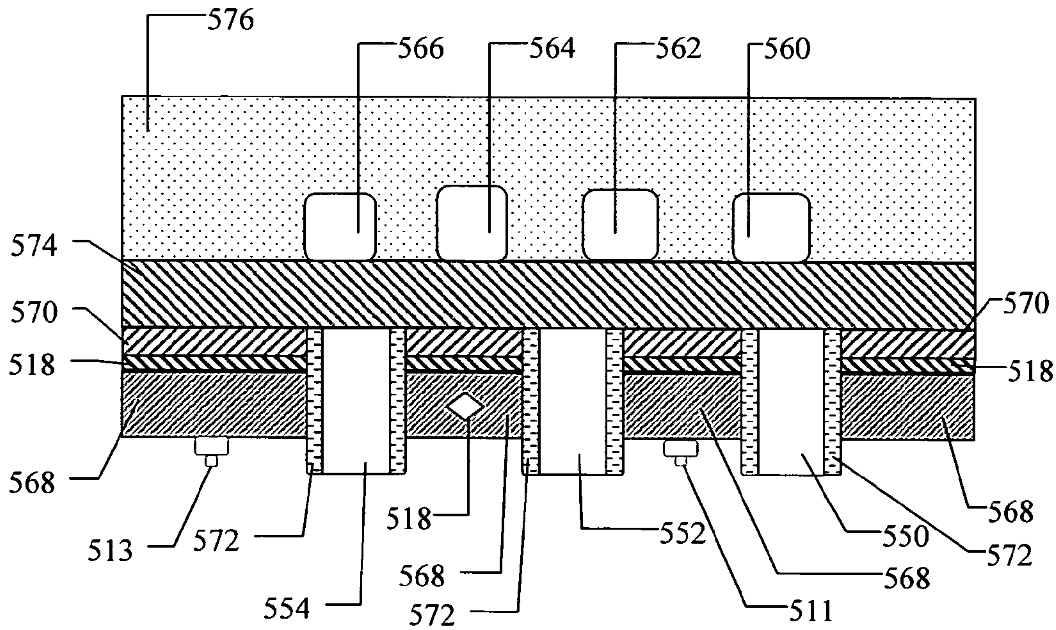
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FIG. 5

TEMPERATURE CONTROL FOR LIGHT-EMITTING DIODE STABILIZATION

CROSS-REFERENCE TO RELATED APPLICATION

[0001] The present application is a continuation-in-part of co-pending U.S. patent application Ser. No. 10/440,719, filed May 19, 2003, which in turn is a continuation-in-part of co-pending U.S. patent application Ser. No. 10/216,620, filed Aug. 9, 2002, which in turn is a continuation of co-pending U.S. patent application Ser. No. 09/700,536, filed Nov. 29, 2001, which claims priority to PCT/US99/11088, filed May 17, 1999, which published as publication number WO 99/60381 on Nov. 29, 1999, all of which are incorporated herein in their entireties by reference. Cross-reference is made to co-pending U.S. patent application Ser. No. 10/440,920 entitled "Optical Instrument Including Excitation Source" to Boege et al. (Attorney Docket No. 5010-027-01), co-pending U.S. patent application Ser. No. 10/440,852 entitled "Apparatus And Method For Differentiating Multiple Fluorescence Signals By Excitation Wavelength" to King et al. (Attorney Docket No. 5010-047-01), both filed on May 19, 2003, and to U.S. patent application Ser. No. 10/735,339, filed Dec. 12, 2003, all of which are incorporated herein in their entireties by reference.

FIELD

[0002] The present teachings relate to an optical instrument using excitation beams generated by a light-emitting diode.

BACKGROUND

[0003] Light-Emitting Diodes (LEDs) can be used as an excitation source for optical detection, for example, in fluorescent measurement. There is a need for providing an LED excitation beam source that does not exhibit beam intensity changes and/or spectral shift. A device compatible with nucleotide amplification reactions, detecting such reactions, and capable of processing a relatively large number of amplification reactions is desirable.

SUMMARY

[0004] According to various embodiments, a system is provided that includes one or more light-emitting diode (LED), a temperature sensor, and a temperature regulator. The temperature sensor can be in thermal contact with the LED, can be capable of measuring an operating temperature, and can be capable of generating an operating temperature signal. The temperature regulator can be capable of receiving an operating temperature signal of the LED and regulating the operating temperature based on the operating temperature signal. Herein, it is to be understood that by LED what is meant is at least one LED, and that a group or array of LED's can be included in an "LED" as described herein.

[0005] According to various embodiments, a method for illuminating a reaction region with excitation beams is provided. The method can include providing a system that includes an LED and a reaction region. The method can include generating excitation beams with the LED; directing the excitation beams toward the reaction region; measuring an operating temperature of the LED; and regulating the operating temperature by transferring heat away from and/or into the LED, based on the measured operating temperature. The reaction region can include a sample retained therein.

[0006] According to various embodiments, a method for illuminating a reaction region with excitation beams is provided. The method can include providing a system that includes an LED and a reaction region. The method can include generating excitation beams with the LED; directing excitation beams to the sample; detecting an optical property of the sample to obtain detection data; measuring the operating temperature of the light emitting diode; and adjusting the detection data based on the operating temperature. The adjustment can be made, for example, by shifting the detection data. The shifting of the detection data can include, for example, a shift in intensity, spectra, or both.

[0007] Additional features and advantages of various embodiments will be set forth in part in the description that follows, and in part will be apparent from the description, or can be learned by practice of various embodiments. Other advantages of the various embodiments will be realized and attained by means of the elements and combinations exemplified in the application.

BRIEF DESCRIPTION OF THE DRAWINGS

[0008] Various embodiments of the present teachings are exemplified in the accompanying drawings. The teachings are not limited to the embodiments depicted in the drawings, and include equivalent structures and methods as set forth in the following description and as would be known to those of ordinary skill in the art in view of the present teachings. In the drawings:

[0009] **FIG. 1** is a side view in partial cross-section of a system including a heater providing temperature stabilization for an LED array according to various embodiments;

[0010] **FIG. 2** is a view in partial side cross-section of a system including a thermoelectric device providing temperature stabilization for an LED array according to various embodiments;

[0011] **FIG. 3*a*** is a side view in partial side cross-section of a system including a fan and cooling fins providing temperature stabilization for an LED array according to various embodiments;

[0012] **FIG. 3*b*** is a top plan view of a capillary sample holder according to various embodiments;

[0013] **FIG. 4** is a top view in partial cross-section of a system including a fan and heating element providing temperature stabilization for an LED according to various embodiments; and

[0014] **FIG. 5** is a side view in a partial cross-section of a system providing a strong thermal conductive path according to various embodiments.

[0015] It is to be understood that both the foregoing general description and the following detailed description are exemplary and explanatory only and are intended to provide a further explanation of the various embodiments of the present teachings.

DESCRIPTION OF VARIOUS EMBODIMENTS

[0016] According to various embodiments, a system is provided that includes one or more light-emitting diode

(LED), a temperature sensor in thermal contact with the LED, and a temperature regulator. The temperature sensor can be capable of measuring an operating temperature and generating a signal. The signal can include an operating temperature signal. The signal can be a digital signal. The digital signal can be indicative of whether the temperature being sensed is above or below a set point. The temperature sensor can generate a signal without thermal contact with the LED. The temperature sensor does not have to directly generate an operating temperature signal but rather can simply indicate whether a temperature is above or below a set point. The temperature regulator can be capable of receiving the operating temperature signal and regulating the operating temperature based on the operating temperature signal.

[0017] According to various embodiments, the system can include a heat-transfer device and a control unit capable of controlling the heat-transfer device. The heat-transfer device can include a fan capable of forming an air current in thermal contact with the LED. The heat-transfer device can include a cooling fin in thermal contact with the LED. The heat-transfer device can include a heater in thermal contact with the LED. The heat-transfer device can include a thermoelectric device in thermal contact with the LED. The thermoelectric device can be connected to a reversible-DC-power supply. According to various embodiments, the temperature regulator can include a temperature system that can be capable of increasing and/or decreasing the operating temperature of the LED.

[0018] According to various embodiments, the temperature regulator can comprise a system adapted to control excitation of one or more fluorescent dyes. The temperature regulator can be adapted such that it is capable of maintaining the operating temperature within an operating temperature range including a minimum temperature and a maximum temperature separated by, for example, about 15° C., about 5° C., about 1° C., or about 0.5° C. The operating temperature range can also be specified as a nominal temperature and an acceptable deviation value range.

[0019] According to various embodiments, the temperature regulator can be a temperature regulating system that can include a user input device that is capable of being programmed to maintain an operating temperature range including a minimum temperature and a maximum temperature. The system can include a display capable of displaying the operating temperature signal.

[0020] According to various embodiments, the system can include an error signaling device capable of signaling an alarm when the operating temperature is greater than a maximum temperature. The error signaling device can signal an alarm when the operating temperature is less than a minimum temperature.

[0021] According to various embodiments, the system can include a substrate in contact with the LED. The substrate can include, for example, a Printed-Circuit Board (PCB). According to various embodiments, the reaction region can include a sample retained therein.

[0022] The sample can include one or more nucleotide. The reaction region can include reagents necessary to perform a nucleic acid sequence amplification reaction. The sample can include fluorescent dyes, labels, or markers. The system can include a detector capable of detecting an optical property of the reaction region.

[0023] According to various embodiments, the temperature sensor can include a thermister, a thermocouple, a resistance temperature detector (RTD), a bandgap semiconductor temperature sensor, a non-contact temperature sensor, a bandgap semiconductor resistive temperature detector, a platinum resistive temperature detector, a bi-metallic temperature detector, a combination thereof, or the like.

[0024] **FIG. 1** is side cross-sectional view of a system **100**, according to various embodiments, including an LED array **110** that includes a plurality of LEDs **111**. The system can also include a focal lens **106**. The focal lens **106** can focus excitation beams emitted by the LED array **110**. The LED array **110** can be in physical and/or thermal contact with a substrate **112**. The LED array **110** can include one or more rows or patterns of individual LEDs. The substrate **112** can be a PCB. A heating device **116**, for example, a resistive heating element, can be provided in thermal contact with the LED array **110**. The heating device **116** can be included in, on, or in and on the substrate **112**. The system **100** can include a temperature sensor **118** in thermal contact the LED array **110**. The temperature sensor can be centrally located with respect to the LED array **110**. The temperature sensor **118** can be included on the substrate **112**. A temperature regulator or temperature regulating system **122** can be provided that is capable of receiving a signal from the temperature sensor **118**. The temperature sensor **118** and temperature regulating system **122** can be integrated and/or can be of a unitary construction. The temperature regulating system **122** can control the heating device **116**. The temperature regulating system **122** can control a fan **114**. The temperature regulating system **122** can control the fan **114** and the heating device **116**. For example, the temperature regulating system **122** can be used to control the heating device **116** to reach or maintain a nominal operating temperature while the fan **114** prevents the operating temperature from getting too high. This optimization can be used, for example, if the LED array **111** is not continuously on. The fan **114** can direct an air current over one or more cooling fins **104**. The cooling fins **104** can be in thermal contact with the LED array **110**, with the substrate **112**, or with both. The temperature regulating system **122** can send signals to and/or receive signals from the temperature sensor **118**, the heating device **116**, and/or the fan **114**. The temperature regulating system **122** can send and receive signals using wires **120**. Excitation beams can be emitted from LED array **110** and directed to one or more reaction region **108**. The reaction region **108** can include a sample **107**. The reaction region can be a microtiter tray.

[0025] **FIG. 2** is a side cross-sectional view of a system **200**, according to various embodiments, that includes a temperature stabilization device for an LED array **210**, for example, by including a plurality of LEDs **111**. A focal lens **206** can be included to focus excitation beams emitted from each of the individual LEDs **211**. The LED array **210** can be in physical and/or thermal contact with a substrate **212**. The system **200** can include a temperature sensor **218** in thermal contact with the LED array **210**, the substrate **212**, or both. The temperature sensor **218** can be included in or on the substrate **212**. A temperature regulating system **222** can receive a signal from the temperature sensor **218**. The temperature regulating system **222** can control a thermoelectric device **214**, for example, a Peltier device. The thermoelectric device **214** can be in thermal contact with the LED array **210**, with substrate **212**, or with both. The thermoelectric device **214** can transfer thermal energy from an ambient environment to the LED array **210**. The thermoelectric device **214** can transfer thermal energy to an ambient environment from the LED array **210**. The thermoelectric device **214** can include a temperature sensor. A plurality of cooling fins **204** can be in thermal contact with the LED array **210** and/or with the thermoelectric device **214**. The temperature regulating system **222** can send signals to and/or receive signal from the temperature sensor **218**, and/or the thermoelectric device **214**, for example, through wires **220**. Excitation beams can be emitted from LED array **210** and can be directed to a plurality of reaction regions **208**, for example, held in a thermal cycling block **230**. The thermoelectric device **214** can be used to maintain a lower temperature than could be otherwise achieved under operating conditions. This can permit the LED array **210** to operate more efficiently, with a higher total flux output. The thermoelectric device **214** can be used in a heating mode, for example, to reach or maintain a temperature when the LED array **210** is not on. The thermoelectric device **214** can be used in a cooling mode when the duty cycle of the LED array **210** is high enough to require cooling.

[0026] **FIG. 3***a* is a side cross-sectional view of a system **300** according to various embodiments and capable of providing temperature stabilization for an LED array **310** including a plurality of individual LEDs **311**. A focal or collimating lens **306** can be included to focus excitation beams emitted from each of the individual LEDs **311**. The collimating lens can be a Fresnel lens. A beam splitter **307** can be included to separate excitation beams from emission beams. The beam splitter **307** can be replaced by a filter or beam splitter as described, for example, in U.S. patent application Ser. No. 10/735,339, filed Dec. 12, 2003, which is incorporated herein in its entirety by reference. The LED array **310** can be in contact with a substrate **312**. The system **300** can include a temperature sensor **318** in thermal contact with the LED array **310**. The temperature sensor **318** can be included in, on, or in and on the substrate **312**. A temperature regulating system **322** can receive a signal from the temperature sensor **318**. The temperature regulating system **322** can control a fan **314**. The fan **314** can direct an air current over a plurality of cooling fins **304**. The cooling fins **304** can be in physical and/or thermal contact with the LED array **310**. The temperature regulating system **322** can communicate with the temperature sensor **318**, and/or the fan, through wires **320**. Excitation beams can be emitted from LED array **310** and directed to a reaction region **308** formed or disposed in, on, or in and on a substrate **309**. The reaction regions can include capillaries **330** of a capillary array. The capillaries **330** can each have a portion that passes through a detection zone **356**.

[0027] According to various embodiments, the temperature control system can include a heater. The system can include a cooler. The system can include both a heater and a cooler. Cooling and heating rates can be augmented by using a plurality of heaters and/or coolers as desired. If a heater is provided, it can comprise a plurality of different types of heating devices. If a cooler is provided, it can comprise a plurality of different types of cooling devices.

[0028] **FIG. 3***b* is a top plan partial view of the array of capillaries **330** shown in **FIG. 3***a*, and the detection zone **356**. The capillaries can traverse the detection zone **356**, where excitation beams from the LED array **310** (**FIG. 3***a*) can be directed. For example, the excitation beams can be used for fluorescence detection of analytes in capillaries of a capillary electrophoresis device. Such can be the case in DNA sequencing and fragment length analysis applications.

[0029] An LED illumination system can provide consistent illumination, can be light in weight, and can require minimal cooling and/or heating. The LED can be a standard semi-conductor device, an organic LED, or an inorganic LED. Examples of organic LEDs are QDOT-based LEDs and a nanotube-based LEDs. The LED can be a stack of LED's such as a stack of organic LEDs or a stack of organic LED layers.

[0030] According to various embodiments, LEDs producing several different excitation wavelengths can be used, either simultaneously or sequentially. The use of a plurality of different excitation wavelengths can improve the calibration matrix necessary to distinguish fluorescence emissions of various dyes.

[0031] According to various embodiments, a system can comprise LEDs, photodiodes, operational amplifiers, and LED-current control circuits. The components of the system can change properties with operating temperature variations. A temperature regulating system can maintain these components at a constant temperature. The constant temperature can be elevated from an ambient temperature. The constant temperature can be lower than an ambient temperature. For example, the system components can be held at a constant temperature above an ambient temperature using a resistive heating element as a heat source under the control of the temperature regulating system. A strong or high thermal conductivity pathway can be used between the system components, for example, to the temperature sensor from a heat source and/or a heat sink.

[0032] The temperature sensor can be used to measure directly, indirectly, or by calculation, the temperature of the system components. The temperature sensor can measure an operating temperature for various components of the system. The temperature sensor can provide feedback to a temperature regulating system. The temperature regulating system can monitor the amount of heating or cooling provided by a heat source or a heat sink to maintain the system components at a nominal temperature within an acceptable deviation value range.

[0033] The temperature control characteristics of a temperature regulating system can be improved by enclosing the system components in a thermally isolated environment. For example, the system components and the temperature sensor, and/or the temperature regulating system, can be placed in an enclosure or housing. The enclosure can have openings for allowing illumination from the LEDs to illuminate a detection zone. Heat exchange pathways can be disposed in the enclosure to allow for thermal transfer between the system and an ambient environment. The heat exchange pathway can be a vent in the enclosure. A cooling fan can cool the thermally isolated environment provided by the enclosure. The heat exchange pathway can include, for example, a high conductivity thermal surface included in the enclosure and in thermal contact with a thermoelectric device. The system components can be separated from the enclosure using a thermal insulator to lower a heat exchange rate between the enclosure and the temperature control components. The temperature sensor can be in thermal contact with, in heat-transfer communication with, or otherwise thermally coupled to, the substrate. Known methods of heat transfer include, but are not limited to conduction, convection, and thermal radiation.

[0034] According to various embodiments, a heat conductive adhesive or compliant pad can be used to attain good thermal conductivity between a heat sink or heat source, and other system components, for example, to maintain temperature stability in the system. A heat exchange pathway can be established for system components such as photodiodes and LEDs using a ground path to the same metal or layer plate, for example, in a PCB. The plate can be a metal, for example, aluminum, copper, or other electrically conductive metals. The system can thus maintain temperature stability and keep various system components at substantially the same temperature. The heat exchange pathway can exchange heat with the ground plate. Other temperature interface materials, for example, adhesive backed resistive elements, can be used to achieve good contact with the system components. A resistive heater can be disposed in or on a common substrate shared with other electrical circuits included in the system.

[0035] **FIG. 4** is a top plan cross-sectional view of a system **400**. A housing **401**, also known as a cave, an oven, or an enclosure, can include openings such as **403** and **407** as shown. LEDs **413**, **415** can irradiate through respective openings (**403**) to illuminate one or more reaction regions (not shown). The opening **407** can allow transmission or passing of emission beams from a reaction region to a detector **440**. One or more temperature sensor **418** can be disposed in or on a housing substrate **412**. The substrate **412** can include a heating device **416**. The temperature sensor **418** can be disposed on or in the housing substrate **412**. LEDs **413** and **415**, and detector **440**, can be disposed on or in the housing substrate **412**. A temperature regulator or temperature regulating system **422**, capable of receiving a signal from the temperature sensor **418**, can be included, for example, in the housing **412**. The temperature regulating system **422** can control the heating device **416** and/or a cooling fan **414**, as desired, for example, to maintain the system **400** within a desired or pre-set temperature range. The housing **401** can provide a relatively small, thermally isolated, volume to be temperature-regulated by the temperature regulating system **422**. Control circuits (not shown) necessary to utilize the LEDs **413**, **415** and the detector **440** can be housed within the housing **401**. Excitation beams can be emitted from the LEDs **413**, **415** and directed toward one or more reaction regions. LED **413** can produce excitation beams of a different wavelength range than LED **415**, for example, LED **413** can produce blue light and LED **415** can produce green light. LED **413** can be operated simultaneously or sequentially with LED **415**.

[0036] **FIG. 5** is a side cross-sectional view of a system **500** according to various embodiments. The system **500** can include photodiode detectors **550**, **552**, and **554** disposed on a substrate **574**. The substrate **574** can have control circuits **560**, **562**, **564**, and **566** disposed on a first surface or back side **575** thereof. The system **500** can include an LED **513** mounted on a plate **568** having a thermal conductivity of about 0.1 w/cm·k or higher, for example, about 0.3 w/cm·k or higher or about 0.5 w/cm·k or higher. For example, the plate **568** can comprise, for example, aluminum, steel, stainless steel, another metal or alloy, a printed circuit board, or a combination thereof. An elastomer pad **570** having a high thermal conductivity can be disposed between the substrate **574** and the plate **568**. The substrate can be a multi-layer structure including a layer having a thermal conductivity of about 0.1 w/cm·k or higher. The elastomer pad **570** can electrically isolate an electric resistive heater **518** from the substrate **574**. The photodiode detectors **550**, **552**, and **554** can be adhered or bonded to the substrate **574** using, for example, an adhesive **572**. A temperature sensor **519** can be disposed in thermal contact with the system **500**, for example, the temperature sensor **519** can be disposed in contact with the plate **568**. Thermal insulation **576** can be disposed adjacent the second surface or backside **575** of the substrate **574** to thermally isolate the system **500** from an ambient environment. The system can maintain the control circuits **560**, **562**, **564**, **566**, the photodiode detectors **550**, **552**, **554**, and the LEDs **511**, **513**, at the same temperature. Accordingly, a constant and uniform temperature can be maintained across the system **500**.

[0037] Various embodiments depicting configurations of LED's, reaction regions, and intervening devices that can be used to direct excitation beams from light sources, for example, LEDs, toward reaction regions, can be found, for example, in U.S. patent application Ser. No. 10/440,920, filed May 19, 2003, entitled "Optical Instrument Including Excitation Source" to Boege et al., U.S. patent application Ser. No. 10/440,719, filed May 19, 2003, entitled "Optical Instrument Including Excitation Source" to Boege et al., U.S. patent application Ser. No. 10/440,852, filed May 19, 2003, entitled "Apparatus And Method For Differentiating Multiple Fluorescence Signals By Excitation Wavelength" to King et al., U.S. patent application Ser. No. 10/735,339, filed Dec. 12, 2003, and International Publication No. PCT/US01/35079. All Patents, Patent Applications, and publications mentioned herein are incorporated herein in their entireties by reference.

[0038] The LED or the LED array can include a plurality of LEDs mounted on a substrate. The LED can thermally contact a temperature regulating system. The temperature regulating system can control a heat-transfer device and/or a temperature sensor. The temperature regulating system can maintain the operating temperature of the LED such that the operating temperature does not change appreciably, by not more than 0.5° C., that is, does not fluctuate by more than 10° C. during operation, for example, by not more than 5° C., by not more than 1° C., by not more than 0.5° C., or by not more than 0.1° C. or less. The temperature regulating system can maintain the operating temperature of the LED such that the operating temperature does not exceed the bounds of a programmed temperature range. According to various embodiments, a temperature regulating system and a temperature sensor can be included in a single-unit or can be included in an integrated device, for example, a Maxim DS1620 device available from Maxim Integrated Products, Inc. of Sunnyvale, Calif.

[0039] The temperature sensor and the LED do not necessarily have to be in physical contact. The temperature regulating system can adjust a monitored temperature of the LED to compensate for any thermal masses intervening between the LED and the temperature sensor and to thus derive, calculate, or estimate an operating temperature.

[0040] According to various embodiments, the LED can be cooled to maintain life and illumination uniformity requirements of a system. According to various embodiments, a forced air cooling system or a thermoelectric device, for example, a Peltier device, can be used to cool the LED and to keep the LED from exceeding a maximum operating temperature.

[0041] According to various embodiments, the temperature of the LED can be monitored, for example, with a temperature sensor, and thermal characteristics of a system and spectral characteristics of any LEDs embedded within the system, can be recorded. With understanding and reproducibility of the spectral coefficients of the LED as a function of an operating temperature, the effects of a spectral shift can be mitigated upon detection of optical properties of a sample. According to various embodiments, a dye matrix or detection data can be altered in accordance with the conditions under which the dye matrix or detection data was gathered or detected. Thermal effects on excitation beams emitted by LEDs, including spectral shifts and intensity changes, can thus be minimized or effectively eliminated. According to various embodiments, the temperature of an LED can be monitored and a computing apparatus can adjust the detection data to compensate for the spectral shifts and/or intensity changes of excitation beams emitted from the LED. The compensation for the shifting can be varied across wavelength ranges, for example, different compensations can be provided for different wavelengths of LEDs. A system can be provided that can include a data adjustment unit comprising a memory adapted to store at least two operating temperatures and at least one respective excitation beam characteristic shift for each operating temperature. A plurality of respective excitation beam characteristic shifts can be stored in the memory. The adjustment data can be in the form of a plurality of respective coefficients. Each coefficient can correspond to a respective LED of an LED array. An exemplary range of coefficients can be from about 0.4 nm/° C. to about 4.0 nm/° C., for example, based on deviation from a set or average operating temperature. The coefficients can include two or more nominal temperature coefficients corresponding to two or more LEDs. The coefficients can be determined or designated based on the position of a respective LED in an LED array.

[0042] According to various embodiments, optical detection instruments utilizing LEDs can obtain very stable intensity or spectral characteristics by stabilizing an operating temperature of an LED. Illumination stability can be important to minimize the signal noise in the system. Illumination stability can improve the sensitivity of the instrument to detect low concentration dyes. Spectral stability can be used to maintain values for the deconvolution matrix associated with a set of dyes to prevent quantification errors. Similarly, variations in intensity resulting from temperature changes can be different for different wavelengths of LEDs, resulting in apparent spectral instability.

[0043] According to various embodiments, illumination stability can be improved by allowing the illumination source to warm-up. According to various embodiments, shutters can block excitation beams from reaching a sample to prevent bleach out. According to various embodiments, shutters can block excitation beams from reaching a sample to prevent bleach out during illumination source warm-up. The illumination source can be brought to a desired operating temperature range prior to enabling or turning on the illumination source, using a heater and/or a cooler. Regulating the temperature of the illumination source prior to enabling the illumination source can prevent the need for a shutter and/or can reduce the warm-up time period. According to various embodiments, samples can be subjected to a reaction or a series of reactions, for example, temperature cycled in a nucleic acid sequence amplification or in a sequencing process. According to various embodiments, the shutter can be unblocked in co-ordination with the reaction or the series of reactions, to detect and collect data at an appropriate time, for example, during a fluorescence detection reading of the sample.

[0044] According to various embodiments, sensitivity of the instrument to detect low concentration dyes can be used. An LED can shift, for example, 5% over a 15° C. ambient temperature range maintained by some optical instrumentation. According to various embodiments, a spectral shift of an LED can vary depending on a center wavelength of the LED, for example, blue LEDs can shift less than red LEDs. The spectral shift can be from about 0.04 nm/° C. to about 0.4 nm/° C. The spectral shift can be different for different temperatures. The spectral shift can be calculated. The spectral shift can be obtained from a look-up table. The table can be sorted by temperature, for example. The table can be provided in a long-term storage of a computer system, for example. According to various embodiments, some optical instrumentation can be sensitive to a dye shift of about 1 nm or less. According to various embodiments, laboratory instrumentation utilizing a relatively more robust dye matrix can be less susceptible to the spectral shift of an LED than a system with a relatively less robust dye matrix. The AB **7500** system available from Applied Biosystems of Foster City, Calif., can have a very good dye matrix and can have little susceptibility to spectral shift for at least most dyes.

[0045] According to various embodiments, the LED radiation source can contain one Light Emitting Diode (LED) or an array of individual LEDs. According to various embodiments, each LED can be a high power LED that can emit greater than or equal to about 1 mW of excitation energy. In various embodiments, a high power LED can be used that can emit at least about 5 mW of excitation energy. In various embodiments wherein the LED or array of LEDs can emit, for example, at least about 50 mW of excitation energy, a cooling device such as, but not limited to, a heat sink or fan can be used with the LED. An array of high-powered LEDs can be used that draws, for example, about 10 watts of energy or less, about five watts of energy or less, or about 3 watts of energy or less. Exemplary LED array sources are available, for example, from Stocker Yale (Salem, N.H.) under the trade name LED AREALIGHTS, and from Lumileds Lighting, LLC (San Jose, Calif.) under the trade name Luxeon Star. According to various embodiments, LED light sources can use about 1 microwatt (μW) of power or more, for example, about 5 mW, about 25 mW, about 50 mW, about 1 W, about 5 W, about 50 W, or about 100 W or more, each individually or collectively when used in an array.

[0046] According to various embodiments, the light source can include a combination of two, three, or more

LEDs, laser diodes, and the like, such as, for example, an LED that can emit radiation at about 475 nm, an LED that can emit radiation at about 539 nm, and an LED that can emit radiation at about 593 nm. The LED can be, for example, an Organic Light Emitting Diode (OLED) an inorganic Light Emitted Diode, that can be polymer-based or small-molecule-based (organic or inorganic), an edge emitting diode (ELED), a Thin Film Electroluminescent Device (TFELD), or a Quantum dot based inorganic “organic LED.” The LED can include a phosphorescent OLED (PHOLED). Super bright LEDs can be used and can be arranged in a light array. According to various embodiments, separate LEDs or a packaged set of LEDs can be used in an array. Spectral emissions of the light sources can be effected by an operating temperature of the light source. Other suitable light sources will be apparent to practitioners in the art given the teachings herein. OLEDs can be used as an array while being designed as a single part. As used herein, the terms “excitation source,”“irradiation source,” and “light source” are used interchangeably.

[**0047**] According to various embodiments, excitation beams emitted from the light source can diverge from the light source at an angle of divergence. The angle of divergence can be, for example, from about 5° to about 75° or more. The angle of divergence can be substantially wide, for example, greater than 45°, yet can be efficiently focused by use of a lens, such as the focusing lens **106** (**FIG. 1**), **206** (**FIG. 2**), and **306** (**FIG. 3**). The lens can be a collimating lens, for example, a Fresnel lens.

[**0048**] According to various embodiments, a quantum dot can be used as a source for luminescence and as a fluorescent marker. Quantum dots can be used for both. The quantum dot based LED can be tuned to emit light in a tighter emission bandpass, for example, an emission bandpass including a full-width of half-max of about 10 nm or less, about 20 nm or less, or about 50 nm or less. The quantum dot based LED can increase the efficiency of the fluorescent system. The efficiency of a quantum dot based LED can theoretically be higher than that of conventional LEDs, potentially over 90% when sandwiched directly between two conductive films with each film directly touching each quantum dot as opposed to the present 20% efficiency for standard LEDs. Quantum dot based LEDs can be made utilizing a slurry of quantum dots, where current flows through an average of several quantum dots before being emitted as a photon. This conduction through several quantum dots can cause resistive losses in efficiency. Quantum dots can provide many more colors than conventional LEDs.

[**0049**] A Quantum dot based LED can emit light in an emission band that is narrower than an emission band of a normal LED, for example, about 50% narrower or about 25% narrower. The emission band of the quantum dots can be a function of the size distribution of the quantum dots, and thus can theoretically be extremely narrow. The Quantum dot based LED can also emit light at an electrical energy conversion efficiency of about, 90% or more, for example, approaching 100%. OLED films, including Quantum dot based LEDs, can be applied to a thermal block, used for heating and cooling samples, in a fluorescence system without interfering with the operation of the thermal block.

[**0050**] According to various embodiments, an OLED can be used and/or produced on a flexible substrate, on an optically clear substrate, on a substrate of an unusual shape, or on a combination thereof. Multiple OLEDs can be combined on a substrate, wherein the multiple OLEDs can emit light at different wavelengths. Multiple OLEDs on a single substrate or multiple adjacent substrates can form an interlaced or a non-interlaced pattern of light of various wavelengths. The pattern can correspond to, for example, a sample reservoir arrangement or array. One or more OLEDs can form a shape surrounding, for example, a sample reservoir, a series of sample reservoirs, an array of a plurality of sample reservoirs, or a sample flow path. The sample flow path can be, for example, a channel, a capillary, or a micro-capillary. One or more OLEDs can be formed to follow the sample flow path. One or more OLEDs can be formed in the shape of a substrate or a portion of a substrate. For example, the OLED can be curved, circular, oval, rectangular, square, triangular, annular, or any other geometrically regular shape. The OLED can be formed as an irregular geometric shape. The OLED can illuminate one or more sample reservoirs, for example, an OLED can illuminate one, two, three, four, or more sample reservoirs simultaneously, or in sequence. The OLED can be designed, for example, to illuminate all the wells of a corresponding multi-well array.

[**0051**] According to various embodiments, an OLED can be used and can be formed from one or more stable, organic materials. The OLED can include one or more carbon-based thin films and the OLED can be capable of emitting light of various colors when a voltage is applied across the one or more carbon-based thin films. Various LEDs can use different films, for example, quantum dot based LEDs can use Indium tin oxide.

[**0052**] According to various embodiments, an operating temperature of an LED can be controlled with a Peltier-effect thermoelectric device, a heat pump, an electrical resistance heating element (Joule heater), fluid-flow through channels in a metal block, reservoirs of fluid at different temperatures, tempered air impingement, a combination thereof, or the like. According to various embodiments, the thermal device can include a fan to direct air-flow over cooling fins, or a cold bar to assist in a heat transfer between an LED and another thermal mass, such as air. According to various embodiments, the thermal conductivity of the LED and/or a platform supporting the LED can be greater than that of a surrounding ambient environment, for example, the surrounding air.

[**0053**] According to various embodiments, a thermoelectric device can be used as a heat-transfer device, for example, an XLT module available from Marlow Industries, Inc. of Dallas, Tex. Controls for the thermoelectric device can include an adjustable-bipolar DC output current power supply. The power supply can provide programmable PID control/ramp to set point control, deviation alarms, and automatic and manual operating modes. In reactions, for example, real-time monitoring of Polymerase Chain Reaction (PCR) reactions, thermoelectric devices can both heat and cool, as desired, the LED by using a bi-directional power supply under programmable and/or logic control. The programmable and logic control can be provided by using a general purpose computer, or custom built hardware, for example, a field programmable gate array (FPGA). Thermoelectric devices can be specifically designed to withstand the continuous temperature excursions required in PCR use.

[0054] According to various embodiments, a heat-transfer device can include a vapor-cycle device, for example, a Freon-based vapor compression or absorption refrigerator. In such units, thermal energy can be extracted from a region, thereby reducing its temperature, then rejected to a "heat sink" region of higher temperature. Vapor-cycle devices can include moving mechanical parts and can include a working fluid, while thermoelectric elements can be totally solid state.

[0055] According to various embodiments, a thermal interface material (TIM) can provide a good thermal contact between two surfaces, for example, between an LED support and a substrate, and/or between an LED housing and a thermoelectric device. The TIM can include silicone-based greases, elastomeric pads, thermally conductive tapes, thermally conductive adhesives, or a combination thereof. Zinc-oxide silicone can be used as a TIM. According to various embodiments, Gap-Pad products, for example, GAP PAD VO ULTRA SOFT materials or SIL-PAD, materials available from Berquist Company of Chanhassen, Minn., can be used as thermal interface materials. A TIM is described in U.S. Pat. No. 5,679,457 to Bergerson, which is incorporated herein in its entirety. According to various embodiments, a TIM can be disposed between a heat-transfer device and an LED.

[0056] According to various embodiments, a method can be provided for maintaining emssion intensity and spectral stability of an LED. The method can comprise: providing a system comprising an LED; generating excitation beams with the LED; measuring an operating temperature of the LED; and regulating the operating temperature by at least one of transferring heat from the LED and transferring heat to the LED, based on the operating temperature, to maintain emssion intensity and spectral stability of the LED. The regulating can comprise retrieving from a memory source adjustment data corresponding to a desired operating temperature or temperature range at which emission intensity and spectral stability of the LED are maintained.

[0057] Other embodiments will be apparent to those skilled in the art from consideration of the present specification and practice of various embodiments disclosed herein. It is intended that the present specification and examples be considered as exemplary only.

What is claimed is:

1. A system comprising:

a light-emitting diode (LED);

a temperature sensor in thermal contact with the LED and capable of measuring an operating temperature of the LED and generating an operating temperature signal; and

a temperature regulating system capable of receiving the operating temperature signal and regulating the operating temperature based on the operating temperature signal, and adapted to control excitation of one or more fluorescent dyes.

2. The system of claim 1, wherein the temperature sensor and the temperature regulating system comprise a single unit.

3. The system of claim 1, wherein the temperature regulating system includes a heat-transfer device and a control unit capable of controlling the heat-transfer device.

4. The system of claim 3, wherein the heat-transfer device includes a fan capable of forming an air current in thermal contact with the LED.

5. The system of claim 3, wherein the heat-transfer device includes one or more cooling fins in thermal contact with the LED.

6. The system of claim 3, wherein the heat-transfer device includes a heater in thermal contact with the LED.

7. The system of claim 3, wherein the heat-transfer device includes a thermoelectric device in thermal contact with the LED.

8. The system of claim 7, wherein the heat-transfer device includes a reversible power supply capable of supplying power to the thermoelectric device.

9. The system of claim 1, wherein the temperature regulating system is capable of increasing the operating temperature of the LED.

10. The system of claim 1, wherein the temperature regulating system is capable of decreasing the operating temperature of the LED.

11. The system of claim 1, wherein the temperature regulating system is capable of maintaining the operating temperature to be within an operating temperature range including a minimum temperature and a maximum temperature separated by about 5° C.

12. The system of claim 1, wherein the temperature regulating system is capable of maintaining the operating temperature to be within an operating temperature range including a minimum temperature and a maximum temperature separated by about 1° C.

13. The system of claim 1, further comprising a user input device in communication with the temperature regulating system and capable of inputting an operating temperature range including a nominal temperature and an acceptable deviation value rage.

14. The system of claim 1, wherein the temperature sensor comprises at least one of a thermistor, a thermocouple, a bandgap semiconductor resistive temperature detector, a platinum resistive temperature detector, or a bi-metallic temperature detector.

15. The system of claim 1, wherein the temperature sensor comprises at least one of a thermistor, or a bandgap semiconductor resistive temperature detector.

16. The system of claim 1, further comprising an error signaling device capable of signaling an alarm when at least one condition of (1) the operating temperature is greater than a maximum temperature, and (2) the operating temperature is less than a minimum temperature, is met.

17. The system of claim 1, further comprising a substrate contacting the LED.

18. The system of claim 17, wherein the substrate has a thermal conductivity of about 0.1 w/cm·k or greater.

19. The system of claim 17, wherein the LED comprises a plurality of LED's.

20. The system of claim 17, wherein the substrate is a multi-layer laminate and at least one layer of the laminate has a thermal conductivity of about 0.1 w/cm·k or greater.

21. The system of claim 17, wherein the substrate includes a printed-circuit board.

22. The system of claim 1, wherein the LED includes an organic LED.

23. The system of claim 1, wherein the LED includes a quantum-dot based light source.

24. The system of claim 1, wherein the LED comprises a plurality of light-emitting diodes that are capable of emitting emissions of different respective wavelength ranges.

25. The system of claim 24, wherein the plurality of LEDs comprises at least one Blue LED and at least one Green LED.

26. The system of claim 1, further comprising a reaction region, and wherein the LED is capable of generating and directing excitation beams toward the reaction region.

27. The system of claim 26, wherein the reaction region comprises a plurality of spaced-apart reaction regions.

28. The system of claim 26, wherein the reaction region comprises a sample disposed therein, and the sample comprises at least one nucleic acid sequence.

29. The system of claim 26, further comprising a detector capable of detecting at least one optical property of the reaction region.

30. The system of claim 26, wherein the reaction region includes at least one nucleic acid sequence amplification reaction reagent.

31. The system of claim 1, further comprising at least one separation region, and wherein the LED is capable of generating and directing excitation beams toward the at least one separation region.

32. The system of claim 31, wherein the LED comprises a plurality of light-emitting diodes that are capable of emitting different respective wavelength ranges.

33. The system of claim 31, wherein the at least one separation region comprises an electrophoresis separation medium.

34. The system of claim 1, wherein the reaction region includes at least one fluorescent reporter dye.

35. A system comprising:

a reaction region;

a light-emitting diode (LED) capable of generating and directing excitation beams toward the reaction region;

a temperature sensor in thermal contact with the LED and capable of measuring an operating temperature of the LED and generating an operating temperature signal;

a detector adapted to detect emission signals from the reaction region and capable of generating detection data; and

a data adjustment unit capable of receiving the operating temperature signal and the detection data, and adapted to adjust the detection data for an excitation beam characteristic shift related to the operating temperature, to form shifted detection data.

36. The system of claim 35, wherein the data adjustment unit comprises a memory adapted to store at least two operating temperatures and at least one respective excitation beam characteristic shift for each operating temperature.

37. The system of claim 35, wherein the temperature sensor comprises at least one of a thermistor, a thermocouple, a bandgap semiconductor resistive temperature detector, a platinum resistive temperature detector, or a bi-metallic temperature detector.

38. The system of claim 35, wherein the LED comprises a plurality of light-emitting diodes that are capable of emitting different respective wavelength ranges.

39. The system of claim 38, wherein the data adjustment unit comprises a memory adapted to store at least two operating temperatures, and at least one respective excitation beam characteristic shift for each operating temperature, and wherein the at least one respective excitation beam characteristic shift comprises at least one respective excitation beam characteristic shift for each different wavelength range.

40. The system of claim 38, wherein the plurality of LEDs comprises at least one blue LED and at least one green LED.

41. The system of claim 35, further comprising at least one separation region, and wherein the LED is capable of generating and directing excitation beams toward the at least one separation region.

42. The system of claim 35, wherein the LED comprises a plurality of LEDs disposed adjacent one another and each LED has a respective operating temperature and a respective excitation beam characteristic shift.

43. The system of claim 35, wherein the LED comprises a plurality of LEDs stacked along a path for directing excitation beams towards the reaction region, and each LED has a respective operating temperature and a respective excitation beam characteristic shift.

44. The system of claim 35, further comprising a temperature regulating system capable of receiving the operating temperature signal and regulating the operating temperature based on the operating temperature signal.

45. The system of **35**, wherein the excitation beam characteristic comprises a spectrum of the generated excitation beams.

46. The system of **35**, wherein the excitation beam characteristic comprises an intensity of the generated excitation beams.

47. A method for illuminating a reaction region with excitation beams, the method comprising:

providing a system comprising an LED, a reaction region, and a sample in the reaction region;

generating excitation beams with the LED;

directing excitation beams at the sample;

measuring an operating temperature of the LED; and

regulating the operating temperature by at least one of transferring heat from the LED and transferring heat to the LED, based on the operating temperature.

48. The method of claim 47, wherein said regulating the operating temperature includes maintaining the operating temperature to be within an operating temperature range including a minimum temperature and a maximum temperature separated by about 5° C.

49. The method of claim 47, wherein said regulating the operating temperature includes maintaining the operating temperature to be within an operating temperature range including a minimum temperature and a maximum temperature separated by about 1° C.

50. The method of claim 47, wherein said regulating the operating temperature occurs prior in time to said generating the excitation beams.

51. The method of claim 47, wherein said regulating the operating temperature comprises setting the operating temperature to be greater than an ambient environment temperature.

52. The method of claim 47, wherein the sample includes reagents necessary to perform a nucleic acid sequence amplification reaction.

53. The method of claim 47, wherein the LED comprises a plurality of light-emitting diodes that are capable of emitting different respective wavelength ranges.

54. A method for illuminating a reaction region with excitation beams, the method comprising:

providing a system comprising an LED, a reaction region, and a sample in the reaction region;

generating excitation beams with the LED;

directing excitation beams to the sample;

detecting an optical property of the sample to obtain detection data;

measuring the operating temperature of the light emitting diode to determine a measured temperature; and

adjusting the detection data of an excitation beam characteristic shift related to the measured temperature.

55. The method of **54**, wherein the excitation beam characteristic comprises a spectrum of the generated excitation beams.

56. The method of **54**, wherein the excitation beam characteristic comprises an intensity of the generated excitation beams.

57. The method of claim 54, wherein the sample includes reagents necessary to perform a nucleic acid sequence amplification reaction.

58. The method of claim 54, wherein the LED comprises a plurality of light-emitting diodes that are capable of emitting different respective wavelength ranges.

59. The method of claim 58, adjusting the detection data comprises picking the excitation beam characteristic shift based on the position of an activated LED from the plurality of LEDs.

60. The method of claim 54, wherein the LED comprise a plurality of LEDs disposed adjacent to one another and each LED has a respective operating temperature and a respective excitation beam characteristic shift.

61. The method of claim 60, wherein the LED's comprise inner LEDs and outer LEDs, and the method comprises making a greater adjustment of excitation beam characteristic shift for one or more of the inner LEDs compared to the adjustment for one or more of the outer LED's.

62. The method of claim 60, wherein adjusting the detection data comprises retrieving from a memory source a respective excitation beam characteristic shift.

63. The method of claim 54, wherein the LED comprises a plurality of LEDs stacked in along a path for directing excitation beams towards the reaction region and each LED has a respective operating temperature and a respective excitation beam characteristic shift.

64. The method of claim 54, further comprising profiling the plurality of LEDs to obtain a calculated operating temperature based on a position of each LED in the plurality of LEDs.

65. A method for maintaining emission intensity and spectral stability of an LED, the method comprising:

providing a system comprising an LED;

generating excitation beams with the LED;

measuring an operating temperature of the LED; and

regulating the operating temperature by at least one of transferring heat from the LED and transferring heat to the LED, based on the operating temperature, to maintain emssion intensity and spectral stability of the LED.

66. The method of claim 65, wherein the regulating the operating temperature comprises retrieving from a memory source adjustment data corresponding to a desired operating temperature or temperature range at which emission intensity and spectral stability of the LED are maintained.

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