

particular FIG. 21 illustrates the increased dynamic range of a barcoded array when it is utilized with DEAL technology. The data show measurements of hCG, a pregnancy test marker, in human serum using the DEAL bar-code immunoassay that can cover the huge dynamic range >4 orders of magnitude.

[0197] In particular, the results illustrated in FIG. 21, show that an expanded range of concentrations that can be detected from a single DEAL-based bio-barcode, demonstrated here for the detection of hCG. hCG is a pregnancy test marker, as well as a serum cancer marker. By varying the primary DNA oligomer concentration that binds the 1^o antibody capture agent during the initial flow patterning step, a single set of bar-code can distinguish the hCG concentration spanning from 25000 mIU/mL to 0.25 mIU/mL (not shown) in a single step.

Example 9

Barcoded Array for Detecting a Biological Profile: Detection of Human Chorionic Gonadotropin (hCG) Over a Period of Time

[0198] Applicants performed a test on a series of standard human chorionic gonadotropin (hCG) spiked human serum samples provided by the National Cancer Institute (NCI). hCG is widely used for pregnancy testing, and also serves as a biomarker for gestational trophoblastic tumors and germ cell cancers of the ovaries and testes.

[0199] The results from these hCG assays are shown in FIG. 22, which illustrate measurement of human chorionic gonadotropin (hCG) spiked in sera using a microfluidic DEAL barcode chip on an integrated platform including a barcoded array manufactured as described in U.S. Application entitled "Microfluidic Devices, Methods and Systems for Detecting Target Molecules" Serial No. to be assigned filed on Jul. 16, 2008, Docket Number P235-US herein incorporated by reference in its entirety.

[0200] In Panel a of FIG. 22, fluorescence images of DEAL barcodes used in measuring standard hCG samples and two unknowns, are shown. The bars used to measure hCG were patterned with DNA strand A at different concentrations. TNF- α encoded by strand B was employed as a negative control. The lane indicated with REF represents the reference marker, while the other lanes indicate hCG test results in which the DNA was patterned from solutions at concentrations that varied from 2 μ M-200 μ M. A negative control using TNF- α was also included.

[0201] ELISA-like sensitivity (\sim 1 mIU/mL), but with a broader detectable concentration range (\sim 10⁵), was demonstrated by quantitating fluorescence intensity. Moreover, even without photon integration, the analyte concentrations over a large range can be readily estimated by eye through pattern-recognition of the full barcode (See also indication in Example 5).

[0202] Quantitation of fluorescence signals obtained at different DNA loading was also performed as indicated in panel (b) of FIG. 22. In such a barcoded array, the bar with high DNA-loading rendered great sensitivity at low analyte concentrations, whereas the bar with low DNA-loading was used to readily discriminate samples with high analyte concentrations. The two unknowns were also assayed and the results are in good agreement with ELISA tests run at NCI Laboratories.

[0203] Applicants noted that the hCG level is tracked during pregnancy, with concentrations in the blood increasing

from \sim 5 mIU/mL in the first week of pregnancy to \sim 2 \times 10⁵ mIU/mL in ten weeks. The microfluidic barcoded arrays used in the experiments herein described can accurately cover such a broad physiological hCG range.

Example 10

Barcoded Array for Detecting a Biological Profile: Protein Profiling in Cancer Patients

[0204] A barcoded array was used to detect a biological profile as illustrated in FIG. 23. In particular, FIG. 23 shows the use of an integrated microfluidic DEAL barcoded device for human serum protein profiling. The serum samples from 12 cancer patients were measured in such prototype clinic test platform.

[0205] The protein profile obtained from this experiment exhibits unique patterns for individual patients, suggesting the efficacy of DEAL bar-code assay for serum-based cancer diagnosis and personalized medicine. This result displays a great indication for using a barcoded device and in particular an integrated DEAL barcode device for diagnostics and in particular human disease diagnostics.

[0206] In particular, the results of FIG. 23 show that the integrated DEAL Bio bar-code device can be used for rapid, sensitive and high-throughput protein measurements out of cancer patient sera. Panel A illustrates the design of the integrated microfluidic device that can conduct a dozen of serum assays at the same time in a highly automated fashion. Blue denotes the microfluidic channels for delivery of all reagents and samples. Magenta shows the control channel for pressure-actuated valves where they intersect the microfluidic channels. Overlay is a representative image of DEAL barcode chip visualized by Cy5 fluorescence probes.

[0207] Measurement of 12 proteins out of 11 cancer patient serum samples and reference serum is illustrated in Panel B. The number denotes each individual lanes used for protein detection out of a patient sample.

[0208] Statistics of 12 protein level present in the serum samples from 12 different patients (S1-S 12), among which S1-5 are breast cancer patients while S6-S11 are prostate cancer patients, is shown in Panel C. Each patient displays a unique pattern of serum proteins that are thought to be associated with their unique molecular origin of cancer.

[0209] A chart listing the specification and medical history of cancer patients is shown in panel D. Several unique signatures can be seen by comparing the medical record and the serum protein profile measured from DEAL bar-code assay.

Example 11

Barcoded Array for Detecting a Biological Profile: Additional Protein Profiling in Cancer Patients

[0210] To further assess the utility and reproducibility of barcoded array for clinical blood samples, applicants measured a panel of 12 proteins from small amounts of serum from 24 cancer patients in a DEAL barcoded microfluidic device. The proteins in this panel included prostate specific antigen (PSA), as well as eleven proteins secreted by various white blood cells. Each barcode was measured many times for each serum sample.

[0211] The stored serum samples from 11 breast cancer patients (all female) and 11 prostate cancer patients (all male) were acquired from Asterand. Two unknowns were acquired from Sigma-Aldrich. Nineteen out of 22 patients were Cau-