SENSOR CHIP FOR USE IN OPTICAL SPECTROSCOPY

Technical Field

The present invention generally relates to a sensor chip for use in an optical spectroscopic device, such as a surface plasmon resonance spectrometer.

Background

Optical spectroscopic devices, such as Surface Plasmon Resonance (SPR) devices, provide a label-free, real-time measurement of a substance by using an optical method that detects changes in the refractive index of a dielectric film adjacent to a metal surface. There are several ways to excite the surface plasmons. One way is by using the Kretschmann configuration which is commonly employed in a number of commercially available instruments that takes advantage of a prism's high refractive index to increase the momentum of light and achieve the required resonance. The Kretschmann configuration utilized a prism coupling arrangement to focus incident light on a reflective material layer. However, the prism coupling arrangement has led to technical constraints in miniaturization of SPR devices, use of sensor substrate materials and in the formation of micro-arrays. For example, the Kretschmann configuration poses strict requirements on the optical properties of the support substrate as well as the need for the layer of the reflective material to be within a specific range.

To overcome the above problems, use of a grating-coupled surface plasmon resonance device is suggested as an alternative to the Kretschmann configuration. This allows for the miniaturization of the device. Furthermore, there is no restriction on the optical quality of the support substrate used in the grating-

coupled surface plasmon resonance and less restriction on the requirement for the reflective material layer to be within a specific thickness range if grating coupled surface plasmon resonance is used instead of the Kretschmann configuration. For example, in the Kretschmann configuration, the thickness range of the reflective material layer, such as gold, is from 46 nm to 50 nm. If the thickness of the gold layer is not within this range, it will lead to a decrease in the surface plasmon resonance coupling efficiency and results in decreasing sensitivity of surface plasmon resonance. On the other hand, in grating coupled surface plasmon resonance devices, the thickness of the reflective material layer is typically about 100 nm. The surface plasmon resonance coupling efficiency is not substantially affected even if the thickness of the reflective material layer is above this value.

It is suggested that micro-fluidic devices can be employed with surface plasmon resonance in order to detect small volumes of sample fluid and to develop assays for simultaneous analysis of parallel interactions. One known sensor chips for use in SPR are FLEXCHIP™ sensor chips from Biacore AB of GE Healthcare, United Kingdom. The FLEXCHIP™ sensor chip consists of a flat sensor chip substrate having grating formations disposed on a surface of the substrate. The flat sensor chip substrate is attached to a micro-fluidic device which has only one fluid channel. The FLEXCHIP™ sensor chips are expensive and the price increases if they have been modified by deposition of biomolecules on the surface. The device can be somewhat cumbersome to manufacture because it requires accurate alignment of the sensor chip to the micro-fluidic device during manufacture. This can be problematic because accurate

attachment of the flow regions of the micro-fluidic device relative to the reflective metal film is necessary to obtain an accurate measurement in SPR. This renders manufacturing and assembly somewhat cumbersome and requires a number of manufacturing steps and extended manufacturing time.

Furthermore, in use, separate equipment such as a microarray machine may be required to deposit the test samples, usually in the magnitude of a few hundred, onto the surface of the sensor chip before it is fixed to the microfluidic device. This machine is expensive and may be complicated to use. Although it is possible that the SPR machine can be configured to align the molecules onto the surface of the sensor chip, the fixed alignment means that it is inflexible to use. If a surface plasmon resonance sensor chip is not readily available, there exists a need for a reflective material layer to be deposited onto the sensor chip substrate in order for it to be used for surface plasmon resonance studies. Before the deposition step, the surface of the sensor chip substrate must be cleaned to a very high degree for the SPR device to function. After deposition, the SPR device must be used immediately as the deposited reflective material layer is still 'fresh' and substantially without contaminants. Accordingly, extra steps involved in cleaning the surface of the sensor chip substrate and then depositing the reflective material layer are needed. This translates to additional costs, processing steps and equipment for a user. For example, it may be necessary to purchase an evaporator for evaporating metal onto the sensor chip substrate. Such metal evaporators are expensive and are complicated to operate.

In some cases, cleaning of commercially available SPR sensor chips is required as the reflective material

layer may be contaminated during storage and transportation. A clean or 'fresh' reflective material surface is a pre-requisite for SPR experiments as any contaminant present, even in trace amounts, may affect the results obtained. Therefore, as mentioned above, an extra cleaning step is required, which adds to the complication of using commercially available SPR sensor chips.

There is a need to provide a sensor chip for optical spectroscopy, such as surface plasmon resonance, that overcomes, or at least ameliorates, one or more of the disadvantages described above.

Summary

According to a first aspect, there is provided a sensor chip having a substrate body comprising at least one fluid channel integrally formed on the surface thereof and a reflective material layer being integrally formed on at least part of said fluid channel.

Advantageously, the fluid channel is integrally formed with said body and said reflective material layer is integrally formed on said fluid channel so that the sensor chip can function as a combined micro-fluidic device in optical spectrometry. More advantageously, alignment problems associated with sensor chips having micro-fluidic channels are avoided because in the sensor chip of the first disclosed aspect, the fluid channels are integrally formed with said body and said reflective material layer is integrally formed on said fluid channels.

According to a second aspect, there is provided a process for making a sensor chip comprising the step of applying, to a substrate body, an imprint forming surface of an imprinting stamp to the substrate body under

conditions to form at least one fluid channel extending through and along the surface of said substrate body.

According to a third aspect, there is provided a method of using the sensor chip of the first aspect, comprising the steps of:

directing a light source that is incident on the reflective material layer;

transmitting a sample fluid through at least one fluid channel;

detecting the light reflected from the reflective material layer; and

measuring the intensity of the reflected light,

wherein a change in the intensity of the reflected light during flow of sample fluid through at least one channel indicates a change in the refractive index of the reflective material layer.

This change in the refractive index of the reflective material layer may be the result of a molecular interaction between the molecules in the sample fluid and the reflective material layer.

According to a fourth aspect, there is provided a surface plasmon resonance system comprising:

a light source;

a sensor chip as defined in the first aspect;

a light detector for receiving light reflected from the reflective material layer of the sensor chip; and

an optical modulator for directing modulated light the said sensor chip.

In one embodiment, the optical modulator may be a chopper. In another embodiment, the optical modulator may be a polarizer.

In one embodiment, there is disclosed a process for making a micro-fluidic device for use in an optical

spectroscopic device, the process comprising the steps of:

providing an imprinting stamp having an imprint forming surface provided thereon covered with a layer of reflective material; and

applying said imprint forming surface of said imprinting stamp to a substrate under conditions to form one or more fluid channels thereon and to transfer, at least part of said reflective material layer, onto at least part of said formed fluid channels, wherein in use said fluid channels allow sample fluid to be transmitted therethrough while said transferred reflective material layer reflects light transmitted thereon from a light source of the optical spectroscopic device.

Advantageously, the process allows for the deposition of a reflective material layer into the channels of a micro-fluidic device as the channels are being formed in one imprinting step. That is, formation of both the channels and the reflective layer can be performed in a single processing step. It is therefore not necessary to use other reflective material deposition apparatus (ie such as a metal evaporator) to deposit the reflective material layer on the channels of the micro-fluidic device. More advantageously, because the reflective layer is deposited during fluid channel formation, a high degree of accuracy is obtained with respect to the deposition of the reflective material layer relative to the fluid channels.

Advantageously, the process allows for the production of a flow cell that can be used in a micro-fluidic device as well as a surface plasmon resonance sensor cell that can be used in experiments that require optical analysis.

In another embodiment there is provided a micro-fluidic device for use in an optical spectroscopic device, the micro-fluidic device comprising:

a substrate having one or more channels disposed on a surface thereon;

a reflective material layer disposed on the surface of at least part of said one or more channels, said reflective material layer having been deposited onto said channels during imprint formation of said channels in an imprint forming step.

Definitions

The following words and terms used herein shall have the meaning indicated:

The term "integrally formed" means that the body of the sensor chip, which comprises the at least one fluid channel and the reflective material layer is a single unitary body. Typically, the integrally formed body may be made in an imprint stamping method in which an imprint stamp having a reflective material layer disposed on an imprint forming surface is applied to a substrate body under conditions to form the channels while at the same time transferring the reflective material layer thereon.

The term "Surface Plasmon Resonance" or SPR is to be interpreted to refer to a resonance condition when the tangential component of the wave vector of a light incident on a reflective material layer matches to the wave vector of the surface plasmons. At this condition, energy is transferred from the incident light to the surface plasmons. This is indicated by a drop in the intensity of light reflected from the surface of the reflective material layer. The resonance condition depends on the wavelength of the incident light, the frequency of the incident light, the refractive index of

all materials used in the surface plasmon resonance device and the angle at which the light is incident on the reflective material layer.

The term "reflective material" is to be interpreted broadly to include any metals that are able of providing free charges, ions or valence electrons. Any metal that is capable of resonating with light at a particular wavelength to produce surface plasmon resonance may be used as the reflective material.

The term "milliscale" is to be interpreted to include any dimensions that are in the range of about 1 mm to about 10 mm.

The term "microscale" is to be interpreted to include any dimensions that are in the range of about 1 (μm) to about 1000 μm. The term "microstructures" as used herein, refers to structures comprising "microscale" features.

The term "nanoscale" is to be interpreted to include any dimensions that are below about 1 μm. The term "nanostructures" as used herein, are structures comprising "nanoscale" or "submicron" features.

The term "three dimensional" is to be interpreted broadly to include any structures, structural features, imprints or patterns that have both lateral variations (thickness) as well as variations with depth.

The terms "gratings" and "grating formations" are to be used inter-changeably, and are typically a series of parallel disposed grooves or slit formations on the surface of a solid surface having dimensions in the nano-scale range.

The term "grating constant" is to be interpreted broadly as the distance between adjacent gratings that are separated by a depression. As shown in Fig. 3B, the grating constant of gratings in a square-wave form is the

distance represented by a-a'. In embodiments where the gratings are in a sinusoidal wave shape or a blazed grating shape, the grating constant is the distance between the adjacent peaks.

The term "glass transition temperature" (T_g) is to be interpreted to include any temperature of a polymer at which the polymer lies between the rubbery and glass states. This means that above the T_g, the polymer becomes rubbery and can undergo elastic or plastic deformation without fracture. Above this temperature, such polymers can be induced to flow under pressure. When the temperature of the polymer falls below the T_g, generally, the polymer will become inflexible and brittle such that it will break when a stress is applied to the polymer. It should be noted that the T_g is not a sharp transition temperature but a gradual transition and is subject to some variation depending on the experimental conditions (e.g., film thickness, tacticity of the polymer, etc.). The actual T_g of a polymer film will vary as a function of film thickness. The T_g will be defined herein as being the bulk glass-transition temperature of the polymer substrate. The bulk glass transition temperature is a specific value that is widely agreed upon in the literature. Glass transition temperature values of polymers may be obtained from *PPP Handbook™* software edited by *Dr D. T. Wu, 2000.*

The word "substantially" does not exclude "completely" e.g. a composition which is "substantially free" from Y may be completely free from Y. Where necessary, the word "substantially" may be omitted from the definition of the invention.

Unless specified otherwise, the terms "comprising" and "comprise", and grammatical variants thereof, are intended to represent "open" or "inclusive" language such

that they include recited elements but also permit inclusion of additional, unrecited elements.

As used herein, the term "about", in the context of concentrations of components of the formulations, typically means +/- 5% of the stated value, more typically +/- 4% of the stated value, more typically +/- 3% of the stated value, more typically, +/- 2% of the stated value, even more typically +/- 1% of the stated value, and even more typically +/- 0.5% of the stated value.

Throughout this disclosure, certain embodiments may be disclosed in a range format. It should be understood that the description in range format is merely for convenience and brevity and should not be construed as an inflexible limitation on the scope of the disclosed ranges. Accordingly, the description of a range should be considered to have specifically disclosed all the possible sub-ranges as well as individual numerical values within that range. For example, description of a range such as from 1 to 6 should be considered to have specifically disclosed sub-ranges such as from 1 to 3, from 1 to 4, from 1 to 5, from 2 to 4, from 2 to 6, from 3 to 6 etc., as well as individual numbers within that range, for example, 1, 2, 3, 4, 5, and 6. This applies regardless of the breadth of the range.

Detailed Disclosure of Embodiments

Exemplary, non-limiting embodiments of a process for making a sensor chip for use in an optical spectroscopic device, such as a surface plasmon resonance device, will now be disclosed. The sensor chip has a substrate body comprising at least one fluid channel integrally formed on the surface thereof and a reflective material layer

being integrally formed on at least part of said fluid channel.

The sensor chip may be made in a process comprising the steps of providing an imprinting stamp having an imprint forming surface provided thereon covered with a layer of reflective material and applying the imprint forming surface of the imprinting stamp to a substrate body under conditions to form one or more fluid channels thereon and to transfer, at least part of the reflective material layer, onto at least part of the formed fluid channels, wherein in use, the fluid channels allow sample fluid to be transmitted therethrough while the transferred reflective material layer reflects light transmitted thereon from a light source of the optical spectroscopic device.

The reflective material layer may be partially or completely transferred to the channels of the sensor chip during the imprint forming step.

The reflective material may be a metal selected from the group consisting of Group IB and Group IIIA of the Periodic Table of Elements, as well as their alloys and combinations thereof.

In one embodiment, the reflective material may be a metal selected from the group consisting of aluminum, copper, gold, silver and combinations thereof.

In one embodiment, the reflective material is gold. In another embodiment, the reflective material is silver.

The reflective material may be deposited on the surface of the imprinting stamp by thermal evaporation or sputtering.

As the reflective material is deposited on the sensor chip at the time of formation of the fluid channels, the sensor chip can be supplied as a complete unit to the end user (ie operator of an SPR device),

without the need for the user to purchase or operate the reflective material deposition apparatus.

The thickness of the reflective material layer covered on the surface of the imprinting stamp may be in the range selected from the group consisting of about 100 nm to about 500 nm, about 100 nm to about 400 nm, about 100 nm to about 300nm, about 100 nm to about 200 nm, about 200 nm to about 500 nm, about 300 nm to about 500nm and about 400 nm to about 500nm. In one embodiment, the thickness of the reflective material layer is about 150 nm.

The substrate body may be a polymer substrate although any material that can be molded by a stamp may be used.

In one embodiment, the polymer substrate is a thermoplastic polymer.

The monomers to form the thermoplastic polymer may be selected from the group consisting of olefins, acrylics, phthalamides, acrylonitriles, cellulosics, styrenes, alkyls, alkyls methacrylates, alkenes, halogenated alkenes, amides, imides, aryletherketones, butadienes, ketones, esters, acetals, carbonates and co-monomers thereof.

In one embodiment, the thermoplastic polymer is poly(ethylene-co-acrylic acid).

In one embodiment, the polymer substrate is a photoresist material. In another embodiment, the photoresist material is SU-8™ photoresist obtained from MicroChem Corp of Newton, Massachusetts of the United States of America.

The imprinting stamp may be made of any suitable material that is chemically inert and may be harder than the substrate body during the applying step. The molds may be made of silicon, metal, glass, quartz, ceramic,

hard polymeric materials with Tg above 200°C or combinations thereof.

The imprinting stamp may have an imprint forming surface provided thereon and may be patterned such that some patterns may protrude from the surface of the imprinting stamp. The patterns may comprise holes, columns, projections or trenches. The patterns may have defined heights and widths in the microscale or in the nanoscale. The patterns may be spaced apart from each other. The patterns may be three-dimensional structures.

The patterns may be formed on the imprinting stamp by a method selected from the group consisting of photolithography, deep reactive ion etching, holographic lithography, e-beam lithography, ion-beam lithography and combinations thereof.

In one embodiment, the patterns on the imprinting stamp may comprise projections and trenches. The projections and trenches may be placed parallel to each other. The projections and trenches may extend along respective longitudinal axes. The width and height of the projections and trenches may be either in the milliscale or in the microscale.

The width of the projections and corresponding channels may be in the range selected from the group consisting of about 100 micron to about 1000 micron, about 100 micron to about 900 micron, about 100 micron to about 800 micron, about 100 micron to about 700 micron, about 100 micron to about 600 micron, about 100 micron to about 500 micron, about 100 micron to about 400 micron, about 100 micron to about 300 micron, about 100 micron to about 200 micron, about 200 micron to about 1000 micron, about 300 micron to about 1000 micron, about 400 micron to about 1000 micron, about 500 micron to about 1000 micron, about 600 micron to about 1000 micron, about 700

micron to about 1000 micron, about 800 micron to about 1000 micron and about 900 micron to about 1000 micron. In one embodiment, the width of the projections and corresponding channels may be in the range of about 200 micron to about 500 micron.

The surface of the projections on the imprinting stamp may be patterned. The pattern may comprise a plurality of gratings and depressions. The plurality of gratings and depressions may be placed parallel to one another. The plurality of gratings and depressions may extend along respective longitudinal axes.

The longitudinal axes of the gratings may be disposed at an angle relative to the longitudinal axis of the projections. The angle between the two longitudinal axes may be selected from the range of about 0° to about 90°. In one embodiment, the angle between the two longitudinal axes is about 90° such that the gratings are substantially disposed perpendicularly to the projections.

The grating constant of the grating formations on the projections of the imprinting stamp may be in the range selected from the group consisting of about 100 nm to about 1000 nm, about 100 nm to about 900 nm, about 100 nm to about 800 nm, about 100 nm to about 700 nm, about 100 nm to about 600 nm, about 100 nm to about 500 nm, about 100 nm to about 400 nm, about 100 nm to about 300 nm, about 100 nm to about 200 nm, about 200 nm to about 1000 nm, about 300 nm to about 1000 nm, about 400 nm to about 1000 nm, about 500 nm to about 1000 nm, about 600 nm to about 1000 nm, about 700 nm to about 1000 nm, about 800 nm to about 1000 nm and about 900 nm to about 1000 nm. In one embodiment, the grating constant is selected from the group consisting of about 450 nm, about 500 nm, about 650 nm and about 750 nm.

The height of the gratings on the projections may be in the range selected from the group consisting of about 10 nm to about 100 nm, about 10 nm to about 90 nm, about 10 nm to about 80 nm, about 10 nm to about 70 nm, about 10 nm to about 60 nm, about 10 nm to about 50 nm, about 10 nm to about 40 nm, about 10 nm to about 30 nm, about 10 nm to about 20 nm, about 20 nm to about 100 nm, about 30 nm to about 100 nm, about 40 nm to about 100 nm, about 50 nm to about 100 nm, about 60 nm to about 100 nm, about 70 nm to about 100 nm, about 80 nm to about 100 nm and about 90 nm to about 100 nm.

During the applying step, the patterns on the imprinting stamp may be substantially transferred to the sensor chip to form patterns on the sensor chip that are complementary to those on the imprinting stamp. As an example, if the pattern on the imprinting stamp comprises projections, the projections may result in channels on the sensor chip when the imprinting stamp is applied to the substrate.

The patterns disposed on the surface of the projections may result in complementary patterns being formed in the channels. For example, the gratings on the projections of the imprinting stamp may result in depressions formed in the channels of the sensor chip; the depressions on the projections of the imprinting stamp may result in gratings formed in the channels of the sensor chip.

The gratings and depressions formed in the channels of the sensor chip, as seen in a cross-sectional view, may follow a sinusoidal wave shape, a square-wave, a trapezoidal shape, a blazed grating shape or a triangular shape. It is to be appreciated that the shape of the gratings and depressions are not limited to those defined above, and may include any periodic but random profile.

It is to be appreciated that due to the imprinting conditions, the shape of the gratings and depressions in the channels of the sensor chip may have some deviation from the shape of the corresponding depressions and gratings on the projections on the imprinting stamp.

It is to be appreciated that the dimensions of the patterns formed on the sensor chip after the imprint forming step should be substantially similar to the dimensions of the patterns on the imprinting stamp.

The imprinting stamp may be applied to the polymer substrate at a temperature that is above the glass transition temperature (Tg) of the polymer substrate. At this temperature, the polymer softens and may conform to the shape of the patterns on the imprinting stamp. Furthermore, the mold may be applied preferably at a predetermined pressure for a certain period of time to form an imprint on the surface of the polymer substrate. The temperature and pressure to be applied will be dependent on the polymer used.

In one embodiment, the imprinting stamp may be applied to the polymer substrate at a temperature that is above the melting point of the polymer substrate.

The temperature used during the applying step may be selected from the group consisting of about 80°C to about 200°C, about 80°C to about 180°C, about 80°C to about 160°C, about 80°C to about 140°C, about 80°C to about 120°C, about 80°C to about 100°C, about 100°C to about 200°C, about 120°C to about 200°C, about 140°C to about 200°C, about 160°C to about 200°C, about 180°C to about 200°C, about 120°C to about 140°C, about 120°C to about 160°C and about 120°C to about 180°C. In one embodiment, the temperature used during the applying step may be in the range of about 95°C to about 140°C.

The temperature used during the applying step may be in the range of from about 5°C to about 50°C above the glass transition temperature of the polymer substrate.

In one embodiment, the temperature used during the applying step may be in the range of from about 5°C to about 30°C above the melting point of the polymer substrate.

The pressure used during the applying step may be selected from the group consisting of about 10 bars to about 50 bars, about 20 bars to about 50 bars, about 30 bars to about 50 bars, about 40 bars to about 50 bars, about 10 bars to about 40 bars, about 10 bars to about 30 bars and about 10 bars to about 20 bars.

The time period used during the applying step may be in the range of about 1 minute to about 20 minutes.

In one embodiment, an imprinting stamp may be applied to the PEAA substrate for about 1 minute at a temperature of about 95°C and pressure of about 10 bars.

When a light beam is transmitted from a light source into the channels of the substrate body, the gratings disposed in the channels may aid in the generation of surface plasmons. This may be due to the diffraction of the light as it contacts the gratings into higher orders, each with a unique phase velocity and momentum that differ by a multiple of the grating wave vector. Waves that match the momentum of the plasmon can resonantly couple with electrons of the reflective material to thereby generate a surface plasmon.

The disclosed sensor chip offers a number of advantages. Firstly, any material may be used for the body of the sensor chip without any restrictions as compared to that used in the Kretschmann configuration. Secondly, by changing the grating constant, the resonance angle for measurement may be shifted accordingly and

conveniently. Thirdly, there is no need for a prism or index matching fluids. Accordingly, a grating-coupled surface plasmon resonance device incorporating said disclosed sensor chip greatly facilitates minimization and multi-array measurements.

The sensor chip may be used for detecting changes that take place at the molecular level at the surface of the reflective material layer. This may be due to binding of test samples to the reflective material layer, interaction and capture of target molecules to the test samples or deposition of at least one layer of molecules at the surface of the reflective material layer.

The sensor chip may be used in a surface plasmon resonance set-up comprising a light source and a light detector.

The light source may be any device that is capable of generating and emitting electromagnetic radiation. The light source may be a device selected from the group consisting of halogen lamps, light emitting diodes, fluorescent lamps and diode laser. The electromagnetic radiation may be in the infrared, visible light or ultraviolet region. The electromagnetic radiation may be a laser beam.

The wavelength of the electromagnetic radiation may be in the range of about 400 nm to about 1200 nm. In one embodiment, the wavelength of the electromagnetic radiation is about 632.8 nm.

The light detector may be any device that is capable of sensing or detecting light. The light detector may be a photodetector such as a photocell, a photodiode, a phototransistor, a charge-coupled device, an image sensor, a photo-electric tube or a photomultiplier. As the light is detected, the intensity of the light reflected from the reflective material layer may be

transmitted from the light detector to a computer or reader that is capable of analyzing the results and hence, generate a surface plasmon resonance profile.

It is to be appreciated that auxiliary units known in the art to be associated with a surface plasmon resonance set-up such as, but not limited to, optical modulators, polarizers, light filters, mechanisms for controlling the amount of light emitted or moving means to move the sensor chip are to be incorporated herein.

Brief Description Of Drawings

The accompanying drawings illustrate a disclosed embodiment and serve to explain the principles of the disclosed embodiment. It is to be understood, however, that the drawings are provided for purposes of illustration only, and not as a definition of the limits of the invention.

Fig. 1 is a schematic diagram of a process for making a sensor chip for surface plasmon resonance according to a disclosed embodiment.

Fig. 2 is a schematic diagram of a process for making a sensor chip for surface plasmon resonance according to a second embodiment.

Fig. 3A is a schematic diagram of a process for making the imprinting stamp with an imprint forming surface.

Fig. 3B is a cross sectional view of the gratings and depressions present on the surface of the projections.

Fig. 4A is a top view of the sensor chip made according to a disclosed embodiment.

Fig. 4B is a cross-sectional view of the sensor chip denoted by line X-X' of Fig. 4A.

Fig. 5 is a schematic layout of a surface plasmon resonance set-up incorporating a sensor chip made in accordance to a disclosed embodiment.

Fig. 6 is a perspective view of a sensor chip used in a surface plasmon resonance technique.

Fig. 7A is a top view optical microscope image at 5x magnification of an imprinting stamp used in a disclosed process.

Fig. 7B is a top view optical microscope image at 5x magnification of a sensor chip after imprinting with an imprinting stamp.

Fig. 8A is an atomic force microscope image at an area of 5 microns x 5 microns of the grating profile on the projections of the imprinting stamp.

Fig. 8B is an atomic force microscope image at an area of 5 micron x 5 micron of the grating profile on the channels of the sensor chip after imprinting with an imprinting stamp.

Fig. 9A is a surface plasmon resonance measurement result of the grating profile on the projections of the imprinting stamp.

Fig. 9B is a surface plasmon resonance measurement result of the grating profile on the channels of the sensor chip.

Fig. 10 is a surface plasmon resonance profile of a sensor chip being used to detect a sample relative to a control in which the sample was absent.

Detailed Description of Drawings

Non-limiting embodiments will be further described in greater detail by reference to the following Examples described further below, which should not be construed as in any way limiting the scope of the invention.

Fig. 1 is a schematic diagram of a process 100 for making a sensor chip for surface plasmon resonance according to a disclosed embodiment. The process 100 comprises the steps of applying an imprinting stamp 2 to a surface of a substrate body made of poly(ethylene-co-acrylic acid) (PEAA) substrate 12.

In step (a), the imprinting stamp 2 is prepared and has a patterned surface consisting of a series of parallel projections 6 disposed between pairs of trenches 8.

A grating pattern 4 is disposed on the surface of projections 6. The grating pattern 4 will be described in further detail below.

In step (b), a layer of a reflective material in the form of gold layer 10 is deposited onto the surface of the imprinting stamp 2 such that the gold layer 10 is deposited onto the projections 6 and in trenches 8. It is to be appreciated that in order to reduce the amount of gold, a mask (not shown) may be used to cover the trenches 8 such that the gold is deposited on the projections 6 but not on the trenches 8 of the imprinting stamp 2.

In step (c), the imprinting stamp 2 is applied using a compressive clamp that functions as a nanoimprinter onto the surface of PEAA polymer substrate 12 at a temperature of 95°C, which is about 45°C above the Tg, and at a pressure of 10 Bars and for about 1 minute to result in the PEAA polymer 12A with a three-dimensional imprint

consisting of channels 14 bounded by walls 16 that are partially covered by the gold layer 10 as shown in step (d). It is to be appreciated that an Obducat nanoimprinter can be used to apply the imprint onto the PEAA polymer substrate 12.

Fig. 2 is a schematic diagram of a process 101 for making a sensor chip for surface plasmon resonance according to a disclosed embodiment. Here, the process 101 is similar to the process 100 and thus like elements are labeled with like reference numerals followed by a prime (') symbol.

In process 101, the shapes of the channels 14' and their respective walls 16' of PEAA polymer 12A' differ slightly when compared to those of PEAA polymer 12A of Fig. 1. This is due to gaps 54 between the imprinting stamp 2' and the surface of the PEAA polymer substrate 12'. The walls 16' of the channels 14' are gradually curved as the PEAA polymer substrate 12' flows into the regions bounded by the trenches 8' of the imprinting stamp 2' during the step of applying an imprinting stamp 2' to the surface of the PEAA polymer substrate 12' at a temperature above the glass transition temperature of the PEAA polymer substrate 12'.

Here, the gold layer 10' is deposited in the channels 14' only due to the lack of contact between the trenches 8' of the imprinting stamp with the surface of the PEAA polymer substrate 12'. Hence, the imprinting stamp 2', having the projections 6' of a dimension to avoid contact with the surface of the PEAA 12', saves on gold having to be deposited onto the top surface of the walls 16'. This therefore saves on cost of production as less gold is used in the device.

Fig. 3A is a schematic diagram of a process for making the imprinting stamp 2" with an imprint forming

surface 4″. The imprinting stamp 2″ can be used in the process 100 of Fig. 1 or process 101 of Fig. 2 and is functionally similar to the imprinting stamps 2 and 2′ of these processes respectively. Accordingly, the imprinting stamp in this Figure is denoted with like reference numerals but with a double prime symbol (″).

In step (a), a grating pattern 4″ is disposed on the surface of an imprinting stamp 2″ by laser holography. In step (b), a variety of methods may be employed to create the projections 6″ and trenches 8″ to form the imprint forming surface of the imprinting stamp 2″. As is known in the art, any methods to create a pattern of imprints on a stamp may be used. For example, a photoresist material may be spun-coated onto the surface of the imprinting stamp 2″ and a series of photoresist imprint patterns can be created in the positive term after processing steps (ie such as baking and photolithography). Following which, etching such as deep reactive ion etching may be used to remove the unwanted materials from the imprinting stamp 2″ to thereby form projections 6″ and trenches 8″.

As is seen in Fig. 3A, the grating pattern 4″ is present on the surface of the projections 6″. Accordingly, as the imprinting stamp 2″ is applied to a PEAA polymer substrate (12,12′) while the PEAA polymer substrate (12,12′) is at a temperature higher than its glass transition temperature, the grating profile as well as the pattern of projections 6″ and trenches 8″ are imprinted onto the PEAA polymer substrate (12,12′).

Fig. 3B is a cross sectional view of the grating pattern 4″ consisting of a series of parallel gratings 52 disposed between pairs of depressions 50 present.

The grating constant or grating period is defined as the distance between adjacent gratings 52 as depicted by the line a-a' of Fig. 3B. As mentioned above, the use of gratings in a surface plasmon resonance device offers several advantages over known surface plasmon resonance device that do not employ gratings.

Fig. 4A is a top view of a sensor chip 30 made according to a disclosed embodiment. The sensor chip 30 comprises a PEAA polymer 12A"' placed on a glass substrate (not shown). The PEAA polymer 12A"' and related features are similar to those discussed above and are denoted by like reference numerals but with a triple prime ("') symbol.

As seen in Fig. 4A, a grating profile 56 is formed in the channels 14"' of PEAA polymer 12A"'. It is to be appreciated that the grating profile 56 is complementary to that of the grating pattern 4" of Fig. 3B.

Fig. 4B is a cross-sectional view of the sensor chip denoted by line X-X' of Fig. 4A. Here, a reflective material layer 10"' is deposited into the channels 14"' of the PEAA polymer 12A"'.

Fig. 5 is a schematic layout of a system 120 for performing surface plasmon resonance. The system 120 incorporates the sensor chip 30 of Fig. 4A.

A light source in the form of laser 20 projects a beam of laser light through an optical modulator in the form of chopper 32 and a pair of polarizers (36A,36B) before it is projected onto the gold layer (10"'), disposed in the sensor chip 30. The pair of polarizers (36A,36B) polarize the laser beam such that it has a selected polarization orientation. The laser beam may be substantially p-polarized or substantially s-polarized.

The sensor chip 30 is placed on a goniometer 38 to allow the control and measurement of the angle of incidence θ and the positioning of a light detector in the form of a photodiode 40 at an angle of 2θ with respect to the incident beam. The photodiode 40 detects the reflected light intensity. The reflected light intensity is read by a reader in the form of lock-in amplifier 42 which is then phase-coupled to the frequency of the chopper 32 and a computer 48 generates the surface plasmon resonance profile. The observed minimum reflectivity value corresponds to the state of a surface plasmon resonance.

The computer 48 is also connected to a steering device in the form of motor-steering device 44. The motor-steering device 44 serves to move the goniometer 38 as necessary in order for the sensor chip 30 to receive the laser beam and to thereby reflect the laser beam.

Fig. 6 is a perspective view of a sensor chip 30^ used in a surface plasmon resonance to detect changes in the refractive index of the reflective material layer 10^ as sample fluids (58A, 58B, 58C) pass through the channels 14^. The components used here are similar to those depicted in Fig. 5 and are thereby depicted by like reference numerals but with the symbol (^).

Here, a light source 20^ such as a diode laser emits light that is incident to and reflected from individual channels 14^ as indicated by arrows 24A-24A~, 24B-24B~ and 24C-24C~. The reflected light is detected by a photodiode 22^. A cover 18 is placed on top of the channel walls.

Sample fluids (58A, 58B, 58C) are passed through the channels 14^ as indicated by the arrows 26A-26A~, 26B-26B~ and 26C-26C~. It is to be appreciated that the

sample fluid (58A, 58B, 58C) in each channel 14^ may be the same or may be different from each other. The sample fluid (58A, 58B, 58C) may contain compounds of interest that preferentially bind to capture molecules on the surface of the gold layer 10^. The sample fluid (58A, 58B, 58C) may contain molecules that are capable of binding to the gold layer 10^ and participate in a molecular self-assembly process.

Fig. 6 indicates that a sensor chip made in accordance to a disclosed embodiment may be suitable as a flow cell for micro-fluidic experiments. This is due to the deposition of the gold layer 10^ into the channels 14^ of the sensor chip in one step to generate both the flow cell and sensor cell concurrently.

Example 1

The process 100 of Fig. 1 is used in this example. The imprinting stamp 2 with a grating pattern 4 on the surface of the projections 6 was provided in step (a).

The imprinting stamp 2"' comprises a silicon substrate that is available commercially and has a grating pattern 4"' fabricated on the surface as shown in Fig. 3A, step (a). The grating constant used in this example is 500 nm. A layer of AZ4620 photoresist from Hoechst Celanese Corporation of Somerville of New Jersey, United States of America was spin coated onto the silicon substrate. The photoresist imprint pattern in the positive term was obtained via a photolithography process. This involves baking the photoresist, exposing the desired pattern to light and removing areas of the photoresist that have not been exposed to light by rinsing the photoresist with an etching solvent.

Following which, deep reactive ion etching was used to produce the imprint pattern corresponding to projections 6‴ and trenches 8‴ as depicted in step (b) of Fig. 3A. The height of the projections 6‴ used here is 80 micron.

After the imprint pattern is disposed onto the surface of the silicon substrate to form a silicon stamp, the silicon stamp was cleaned with acetone before being rinsed in isopropyl alcohol and deionized water. In step (b) of Fig. 1, the silicon stamp 2 was coated with a layer of gold 10 with a thickness of about 150nm by thermal evaporation.

In step (c), the silicon stamp 2 was applied onto the surface of a PEAA substrate 12. The PEAA was obtained from Sigma-Aldrich of St. Louis of Missouri of the United States of America (catalog number: 18,104-8, Chemical Abstract Service Registry Number of 9010-77-9) in the form of beads 20wt% acrylic acid. When used, PEAA beads were heated onto a glass substrate (not shown in Fig. 1) at a temperature of 95°C until completely molten and then contacted with the gold-coated silicon stamp using a compressive clamp under a pressure of 10 bars for about 1 minute.

In step (d), the silicon stamp 2 and PEAA substrate 12 were separated after being cooled to room temperature so that the resulting PEAA polymer 12A has an imprint pattern that is complementary to that on the silicon stamp 2. For example, channels 14 bounded by walls 16 are formed after step (c). Importantly, due to the weak adherence between the gold layer 10 and the silicon stamp 2, the gold layer 10 transfers easily to the PEAA substrate 12 at the same time as the imprint pattern is formed on the PEAA polymer 12A. The stamped gold adhered

to the PEAA polymer 12A well. The inventors have found that this process is versatile and may allow for substantially accurate imprint formations over large areas.

Fig. 7A is a top view optical microscope image at 5x magnification of the silicon stamp 102 used in this example. Fig. 7B is a top view optical microscope image at 5x magnification of the PEAA polymer 112A after imprinting step (c). It can be seen from these images that the pattern on the silicon stamp 102 consisting of projections 106 and trenches 108 are transferred onto the PEAA polymer 112A as corresponding channels 114 and walls 116. The channels 114 of the PEAA polymer 112A allows for a plurality of sample fluids to pass through the channels 114 whereby changes to the gold layer during the sampling process can be detected in a surface plasmon resonance set-up. The width of the walls is about 10 micron here.

Fig. 8A is an atomic force microscope image at an area of 5 micron x 5 micron of the grating profile on the projections of the silicon stamp. Fig. 8B is an atomic force microscope image at an area of 5 micron x 5 micron of the grating profile on the channels of the PEAA polymer after the imprinting step (c). By comparing Fig. 8A and Fig. 8B, it can be seen that the molten PEAA substrate and the silicon stamp conformed to each other such that the grating profile was replicated. It can be seen that the grating profile on the PEAA polymer was not so homogenous as compared to that of the silicon stamp. Without being bound by theory, it is thought that the following has occurred. Firstly, the profile of the gratings' top part on the PEAA polymer is the reflection of the profile of the gratings' bottom part on the

silicon stamp which may not be smooth after deep reactive ion etching. Therefore, part of the roughness is the result of the polymer conforming to the roughness of the stamp. Secondly, some damage to the grating profile may occur when the silicon stamp is removed from the PEAA substrate. It is to be noted that there was substantially no difference in the grating profile amplitude between the silicon stamp and the PEAA polymer.

Example 2

In this example, a PEAA substrate 12 with channels 14 of 200 micron width and walls 16 with width of 3 mm was made in accordance with a disclosed process. The thickness of the gold layer used here is about 150 nm. The PEAA substrate 12 was employed in a sensor chip 30 in a surface plasmon resonance set-up 120 as depicted in Fig. 5.

A p-polarized He-Ne-laser (JDS Uniphase, USA) with a wavelength of 632.8nm and laser point of 2 mm was used to excite surface plasmons. The chopped laser beam was reflected from the grating profile within the channels 14 and the angular reflectivity was monitored by means of the photodiode 40. The reflectivity signal was read out by the lock-in amplifier 42 which is phase-coupled to the frequency of the chopper 32 to obtain a typical surface plasmon spectrum. The observed minimum of reflectivity corresponds to the state of a surface plasmon resonance.

Fig. 9A is a surface plasmon resonance measurement result of the grating profile on the projections 6 of the silicon stamp 2. Fig. 9B is a surface plasmon resonance measurement result of the grating profile on the channels 14 of the PEAA polymer 12A. It can be seen that both of the surface plasmon resonance measurement results have

sharp minimum angles, which means that the grating profile on the projections 6 on the silicon stamp 2 or within the channels 14 of the PEAA polymer 12A can couple to the surface plasmon with a high efficiency. This data demonstrates that the gold layer 10 was efficiently transferred to the PEAA polymer 12A during formation of the channels 14 because clear and distinct minima reflectivity angles were obtained.

In this case, the surface plasmon resonance profile was the result from one single channel and not the average result of several channels.

Example 3

The sensor chip 30 and surface plasmon resonance set-up 120 of Example 2 was used in a molecular bonding self-assembly process.

1 mM Mecapto-terminated polyethylene glycol (mercapto-PEG) in phosphate-buffered saline (PBS) was used as the sample fluid to flow through the channels 14 of the sensor chip 30 for 20 minutes to enable the self assembling process. The surface plasmon resonance measurement results before and after the self-assembly film adsorption to the gold surface are shown in Fig. 10. As shown in Fig. 10, a significant increase in the minimum reflectance value, a broadening of the surface plasmon resonance profile and an approximately 0.5° shift in the plasmon angle were observed after 20 minutes of the self-assembly process. This means that the mercapto-PEG chains bonded to the gold surface. This experiment demonstrates that the quality of the gold surface was good enough to work as a sensing surface.

Applications

The sensor chip in a surface plasmon resonance set-up may be used for a variety of applications in pharmaceutical industries, research, medical diagnostic testing, detection of biologics or microorganisms for food safety or security purposes (such as bio-terrorism monitoring) or in environmental monitoring. These applications may comprise ligand screening, immunology, cell biology, signal transduction, chemical interactions and nucleotide-nucleotide, nucleotide-protein, protein-protein and protein-lipid interactions.

Advantageously, the fluid channel is integrally formed on the surface of the sensor chip and a reflective material layer is integrally formed on at least part of said fluid channel in one step. As discussed above, a reflective material layer deposited on the surface of an imprinting stamp is transferred to the surface of a polymer substrate when the imprinting stamp is applied to the polymer substrate at a temperature that is above the glass transition temperature of the polymer substrate. Hence, the imprinting stamp results in formation of imprints as well as transfer of a reflective material layer to the polymer substrate in one step.

Advantageously, gratings may be incorporated into the sensor chip in order to aid in the generation of surface plasmons and to substantially obviate the need for a substrate surface with a defined optical quality as well as the need for a reflective material layer to have a defined thickness.

Advantageously, the sensor chip may not require additional processing steps such as cleaning and depositing the reflective material layer after the sensor chip is manufactured.

Advantageously, the sensor chip may not require cleaning after it is packed and transported to an end-user. A protective stamp covering may be provided over the reflective material layer during storage and transportation. The protective stamp covering may substantially protect the reflective material layer from contamination. Accordingly, the stamp covering can be removed from the sensor chip before the sensor chip is used.

Advantageously, additional equipment to deposit test substances on the surface of the reflective material layer may not be necessary. This is because sample fluids containing the test substances can transmit through the formed fluid channels to thereby deposit test substances on the surface of the reflective material layer.

Advantageously, the sensor chip can be manufactured in a reduced amount of time as compared to known processes.

It will be apparent that various other modifications and adaptations of the invention will be apparent to the person skilled in the art after reading the foregoing disclosure without departing from the spirit and scope of the invention and it is intended that all such modifications and adaptations come within the scope of the appended claims.

Claims

1. A sensor chip having a substrate body comprising at least one fluid channel integrally formed on the surface thereof and a reflective material layer being integrally formed on at least part of said fluid channel.

2. A sensor chip as claimed in claim 1, comprising a plurality of grating formations disposed on the surface of said at least one fluid channel.

3. A sensor chip as claimed in claim 2, wherein the width of said grating formations is in the nanoscale range.

4. A sensor chip as claimed in claim 3, wherein the grating constant of said grating formations is in the range of 100 nm to 1000 nm.

5. A sensor chip as claimed in claim 4, wherein the height of said grating formations is in the range of 10 nm to 100 nm.

6. A sensor chip as claimed in claim 5, wherein said grating formations have a shape, when viewed in cross-section, selected from the group consisting of sinusoidal wave shape, square-wave, trapezoidal shape, blazed shape and triangular shape.

7. A sensor chip as claimed in claim 1, wherein said reflective material is a metal selected from the group consisting of Group IB and Group IIIA of the

Periodic Table of Elements, alloys and combinations thereof.

8. A sensor chip as claimed in claim 7, wherein said metal is selected from the group consisting of aluminum, copper, gold, silver and combinations thereof.

9. A sensor chip as claimed in claim 1, wherein the thickness of said reflective material layer is in the range selected from 100 nm to 500 nm.

10. A sensor chip as claimed in claim 1, comprising a plurality of said fluid channels, each of said adjacent channels being disposed between a trench formation.

11. A sensor chip as claimed in claim 1, wherein the width of said at least one fluid channel is in the range of 100 micron to 1000 micron.

12. A sensor chip as claimed in claim 1, wherein said substrate body is a thermoplastic polymer.

13. A sensor chip as claimed in claim 12, wherein said thermoplastic polymer comprises monomers selected from the group consisting of olefins, acrylics and co-monomers thereof.

14. A sensor chip as claimed in claim 13, wherein said thermoplastic polymer is poly(ethylene-co-acrylic acid).

15. A sensor chip as claimed in claim 1, wherein said substrate body is a photoresist material.

16. A process for making a sensor chip comprising the step of applying, to a substrate body, an imprint forming surface of an imprinting stamp to said substrate body under conditions to form at least one fluid channel extending through and along the surface of said substrate body.

17. A process for making a sensor chip as claimed in claim 16, comprising the step of providing a reflective material layer on the imprint forming surface of said imprinting stamp during said applying step to transfer at least part of said reflective material layer to said at least one fluid channel.

18. A process for making a sensor chip as claimed in claim 16, wherein said substrate body is a thermoplastic polymer.

19. A process as claimed in claim 18, comprising, during said applying step, the step of selecting a temperature that is above the glass transition temperature (Tg) of said thermoplastic polymer.

20. A process according to claim 19, wherein the temperature during said applying step is up to 50°C above said (Tg).

21. A process as claimed in claim 18, comprising, during said applying step, the step of selecting a temperature that is in the range of 5°C to 30°C above the melting point temperature of said thermoplastic polymer.

22. A process according to claim 16, wherein the pressure applied to said thermoplastic polymer by said

imprinting stamp during said applying step is in the range of 10 Bar (1 MPa) to 50 Bar (5 MPa).

23. A process according to claim 16, wherein the time of said applying step is in the range of 1 minute to 20 minutes.

24. A method of using the sensor chip of claim 1, comprising the steps of:

directing a light source that is incident on said reflective material layer;

transmitting a sample fluid through said at least one fluid channel;

detecting the light reflected from said reflective material layer; and

measuring the intensity of said reflected light,

wherein a change in the intensity of said reflected light during flow of said sample fluid through said at least one channel indicates a change in the refractive index of said reflective material layer.

25. A surface plasmon resonance system comprising:

a light source;

a sensor chip as claimed in claim 1;

a light detector for receiving light reflected from said reflective material layer of said sensor chip; and

an optical modulator for directing modulated light onto said sensor chip.