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(54) **SENSOR SYTEM FOR DETECTING THE PRESENCE OR CONCENTRATION OF ANALYTES**

(57) The invention relates to a Sensor system (1) for detecting the presence or concentration of an analyte (A) in a medium (M), comprising: a sensor (10) having a sensor head (100) comprising a chamber (101), which sensor head (100) comprises a permeable area (105) through which the analyte (A) can pass into the chamber (101) when the sensor head (100) contacts said medium (M), a cross-linked hydrogel (H) filling said chamber (101), which hydrogel (H) is configured to undergo a change in volume when contacting the analyte (A) passed into the chamber (101) which leads to a change in pressure in said chamber, and a pressure sensor (102) configured to measure the pressure (P) in said chamber (101) for detecting the presence or concentration of said analyte (A).

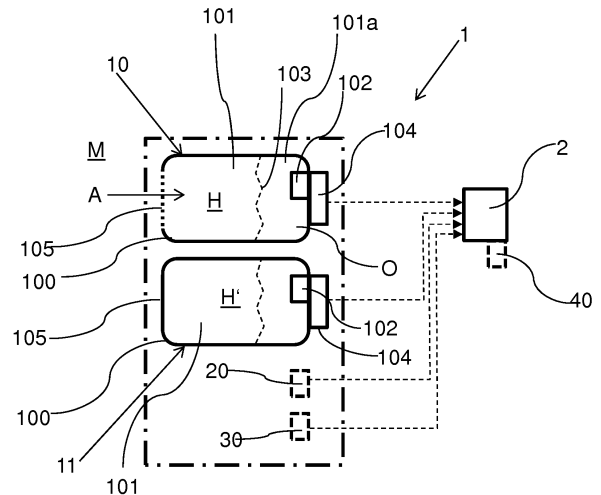


FIG. 2

Description

[0001] The invention relates to a sensor system for detecting the presence or concentration of an analyte.

[0002] Such sensors for detecting the presence/concentration of an analyte in a fluid medium can be based on polymers such as hydrogels which react to a stimulus, here the presence/specific concentration of an analyte with pronounced volume changes, for instance due to incorporation or release of water molecules.

[0003] For realizing such a material and for detecting the presence, particularly concentration of an analyte, hydrogels are typically modified to comprise selective responsive components which are able to selectively bind an analyte.

[0004] In such a hydrogel being maintained at a defined temperature, the binding of the analyte to the hydrogel disturbs the balance between the tendency to capture water and to release water from the hydrogel. In case of electrolyte-sensitive hydrogels comprising crown ether, the amount of bound water per crown ether differs significantly for the free configuration and complex comprising the metal ion (analyte). Therefore, actually small changes in analyte concentration may lead to large changes of the swelling level (i.e. water content and volume). These effects can be drastically amplified when the hydrogel exhibits a critical volume phase transition at a so called LCST (Lower Critical Solution Temperature), at which the volume of the hydrogel changes abruptly from the swollen to the shrunken state (and vice versa).

[0005] As indicated in Fig 1, this transition temperature LCST, here in case of a hydrogel sensitive to K^+ is decreased due to capture of K^+ in a 2:1 crown ether complex. At a constant temperature a reversible release of water and a decrease in volume occurs. In case the potassium concentration is lowered, potassium is removed from the crown ether and water is received in the hydrogel resulting in an increasing volume. It is known, that under these conditions, hydrogels are only sensitive to an analyte in a rather narrow temperature interval around LCST (here between $LCST_a$ und $LCST_b$). This can be explained by the observation that only in this interval the volume-temperature curve comprises a significant slope. Due to the detection of the analyte the shift of this curve thus only results in a vertical distance at these temperatures which corresponds to a change in volume. In the remaining temperature regions (in case of Poly(N-isopropylacrylamid)(PNIPAAm) below $30^\circ C$ and above $33^\circ C$) said curve does not comprise a significant slope, such that the shift of the curve due to potassium does not result in a significant change of volume of the hydrogel.

[0006] Further, particularly, WO98/19787 discloses a sensor device composed of a crystalline colloidal array (CCA) polymerized in a hydrogel. The hydrogels are characterized as being capable of shrinking and swelling in response to specific stimuli applied thereto which

changes the embedded lattice structure of the CCA which is optically monitored.

[0007] In the state of the art, particularly in WO98/19787, hydrogels are usually considered at constant (ambient) pressure and changes of the properties of the respective hydrogel are often characterized using gravimetric or optical methods. However, under isobaric conditions, as described above, the sensitivity of the hydrogel to the respective analyte is limited to a small temperature region.

[0008] Thus, the problem to be solved by the present invention is to provide a sensor system that avoids the above-stated disadvantage of a small useable temperature interval.

[0009] This problem is solved by a sensor system having the features of claim 1.

[0010] Further aspects of the present invention are described below. Specific embodiments of the sensor system according to invention are stated in the sub claims and are also described below.

[0011] According to claim 1, a Sensor system for detecting the presence or concentration of an analyte in a medium is disclosed, comprising:

- a sensor having a sensor head comprising a chamber, which sensor head comprises a permeable area through which the analyte can pass into the chamber when the sensor head contacts said medium,
- a cross-linked hydrogel filling said chamber, which hydrogel is configured to undergo a change in volume (e.g. the hydrogen swells or shrinks) when contacting the analyte passed into the volume which leads to a change in pressure in said chamber (particularly due to the isochoric state of the hydrogel in said chamber), and
- a pressure sensor (particularly arranged in said volume) that is configured to measure the pressure in said chamber for detecting the presence or concentration of said analyte.

[0012] Particularly, in the framework of the present invention, a cross-link is an (e.g. covalent) bond that links one polymer chain to another. A cross-linked hydrogel is a hydrogel comprising a plurality of polymer chains, wherein each of the polymer chain comprises at least one cross-link, particularly a plurality of cross-links, to another polymer chain. Macroscopically, crosslinking has the effect on polymers that they are no longer soluble in solvents, and indeed exhibit swelling upon solvent uptake.

[0013] Particularly, the sensor system can be configured to conduct determining the concentration in a continuous fashion or merely at specific points in time (e.g. at regular intervals).

[0014] Particularly, in an embodiment said permeable area can comprise a membrane or grid that is permeable for the analyte, but not for the hydrogel residing in the chamber, wherein said area forms a counter bearing for

the hydrogel so that a pressure can build up in the chamber when the hydrogel tends to swell in said chamber due to a change in concentration of the analyte in the chamber of the sensor head.

[0015] Surprisingly, it has been found that using a cross-linked hydrogel in isochoric conditions (e.g. in said chamber of the sensor head) significantly increases the temperature interval in which measurements can be performed. This is due to the surprising fact that said hydrogel comprises essentially a linear dependency of the measured pressure over a wide range of temperatures below LCST.

[0016] Furthermore, the cross-linked hydrogel comprises a sufficient stability that allows loading the hydrogel in the isochoric chamber with a pressure. Furthermore, such a cross-linked hydrogel is much less likely to escape from the chamber of the sensor head than a linear hydrogel/polymer, which are not suited for these kinds of pressure measurements.

[0017] Due to the fact that measurements can now be performed over a much wider range of practical temperatures complex mathematical procedures to compensate of a strong temperature dependence of the respective measurement or the need to keep the temperature constant upon measuring can be avoided. Moreover, keeping the temperature constant is hardly possible when the sensor head is implanted into a patient that exhibits a varying body temperature (e.g. due to a fever etc.)

[0018] Particularly, according to an embodiment the cross-linked hydrogel is a thermo-responsive polymer that comprises a lower critical solution temperature (LCST), i.e. a critical temperature below which the components of a mixture, here a polymer and water, are miscible for all compositions, which LCST decreases when the analyte bonds to the hydrogel and which increases when the analyte is released from the hydrogel, wherein below the LCST the measured pressure depends linearly and/or mildly quadratically on the temperature of the hydrogel. This means that the sensor comprises essentially a constant sensitivity for the analyte over a relatively broad range of temperatures below LCST due to the essentially linear and/or mildly quadratically dependence of the pressure signal on temperature.

[0019] At the LCST the volume of the hydrogel changes abruptly with temperature from a swollen to a shrunken state (and vice versa). Increasing the temperature of a swollen hydrogel above the LCST leads to a decreased efficiency of the hydrogen bonds between the polymer and water which finally does not suffice to keep the polymer in the solution. Thus, a phase separation occurs, wherein water is released from the polymer resulting in two phases: a polymer-enriched phase and an aqueous phase that does not comprise the polymer. Alternatively, also hydrogels having an UCST (Upper Critical Solution Temperature) or hydrogels exhibiting both, an LCST and a UCST may be used in the sensor system according to the present invention.

[0020] Further, according to a preferred embodiment

of the present invention, the hydrogel comprises a plurality of a responsive component (e.g. compound), wherein the respective responsive component is responsive to the analyte such that the volume of the hydrogel tends to decrease when the respective responsive compound binds (non-covalently) to a specific analyte such that the pressure in said chamber decreases, and wherein the volume of the hydrogel tends to increase when a specific analyte is released from the respective responsive compound.

[0021] Further, according to a preferred embodiment of the present invention, the responsive component (e.g. compound) comprises one of:

- a crown ether, wherein particularly the analyte is K^+ ,
- a boronic acid moiety, in particular a boronic acid moiety connected to the polymer via a hydrophobic linker such as a phenyl group, wherein particularly the analyte is glucose.

[0022] Regarding the detection of potassium (K) or K^+ , particularly the polymer Poly(N-isopropylacrylamid) (PNIPAAm) is used in the hydrogel residing in the sensor head's chamber. Here, particularly, the size of the crown ether ring determines which positive ions fit into the respective ring. In principle, regarding crown ether, 1:1 complexes (one crown ether per positive ion, e.g. K^+) and 2:1 complexes (two crown ethers per positive ion, e.g. K^+) can be used.

[0023] Generally any type of crown ether can be used being capable of chelating a specific cation to be detected such as 18-crown-6 or 15-crown-5 ethers. The skilled person is familiar with such terms regarding crown ethers. In a particularly preferred embodiment, the hydrogel of the present invention comprises binding site for 2:1 complexes using two 15-crown-5 moieties. In a further preferred embodiment, a hydrogel is suggested on basis of Poly(N-isopropylacrylamid) (PNIPAAm) being crosslinked and having a plurality of binding sites for 2:1 complexes using two 15-crown-5 moieties.

[0024] However, in alternative embodiments, depending on the specific analyte, also other responsive components or compounds can be used, such as one of: Cryptands, spherands, valinomycin, enniatin B, nonactin, gramicidin, FCCP / CCCP, 2,4-dinitrophenol, monensin, ionomycin, podands, cyclodextrins, cyclophanes, calixarenes, resocarene, cavitands, careands, hemicerands, clefts, tweezers, catenanes and rotaxane.

[0025] Particularly cryptands (such as crown ethers) are suitable for metal ions such as K^+ .

[0026] Further, according to an embodiment of the present invention, said crown ether molecules are attached to the hydrogel in such a manner that they form pairs, wherein the two crown ethers of the respective pair are configured to bind an analyte simultaneously, wherein particularly the analyte is arranged between the crown ethers of the respective pair in its bound state (2:1 complex as suggested above).

[0027] However, using suitable responsive components/compounds, the invention can be applied to a vast variety of different analytes other than potassium and glucose, particularly the analyte can also be one of the following: Electrolytes, carbohydrates, fatty acids, lipids, sugars, nucleotides, deoxyribonucleic acids, ribonucleic acids, amino acids, peptides, proteins, antibodies, hormones, neuro-transmitters, metabolites, metabolites, antigens, drugs, medicines, nano-particles, toxins or any other suitable substance known to the person skilled in the art. Further, the analyte can be one of: albumin / globuline, alkaline phosphatase, alpha-1-alpha-1-antitrypsin, alpha-1-fetoprotein, alpha ammonia, antithrombin III, bicarbonate, bilirubin, carbohydrate antigen 19-9, carcino-embryonic antigens, chloride, cholesterol, cholinesterase, chylomicron residues, cobalamin / vitamin B12, coeruloplasmin, C-reactive proteins, Cystatin C, D-dimers, iron, erythropoietin, erythrocytes, ferritin, fetuin A, fibrinogen, folic acid / vitamin B9, free tetraiodothyronine (FT4), free triiodothyronine (FT3), gamma glutamyltransferase, glutamate dehydrogenase, glutamate oxalacetate Transaminase, glutamate pyruvate transaminase, glycohemoglobin, hematocrit, hemoglobin, haptoglobin, uric acid, urea, HDL cholesterol, homocysteine, immunoglobulin A, immunoglobulin E, immunoglobulin G, immunoglobulin M, INR, calcium, creatinine, creatine Kinase, copper, lactate, lactate dehydrogenase, LDL cholesterol, leukocytes, lipase, lipoprotein, magnesium, corpuscular hemoglobins, myoglobin, sodium, NT-proBNP / BNP, phosphate, prostate aspergilligen antigens, reticulocytes, rheumafactors, platelets, thyroid stimulant hormone, transferrin, triglycerides, troponin T, and VLDL cholesterol.

[0028] Furthermore, generally, the analyte can also be an active substance, wherein the notion active substance shall include typical drugs or metabolites, biologicals, or small molecules comprising, which are given for treatment of diseases such as muscarinic receptor antagonists, neuromuscular blocking agents, cholesterol inhibitors, adrenergic receptor agonists, and indirectly acting sympathomimetic agents, methylxanthines, alpha-adrenergic receptor antagonists, ergot alkaloids, beta-adrenoceptor antagonist, inactivation inhibitors, antisymphotonika, 5-HT receptor agonists, histamine receptor agonists, histamine receptorantagonists, analgesics, local anesthetics, sedatives, anticonvulsants, convulsants, muscle relaxants, Antiparkinsonian agents, neuroleptics, antidepressants, lithium, tranquilizers, immunosuppressants, antirheumatics, antiarrhythmics, antibiotics, ACE inhibitors, aldosterone receptor antagonists, diuretics, vasodilators, positive inotropic agents, antithrombotic / thrombolytic substances, laxatives, antidiarrheals, drugs for obesity, uricostats, uricuricurics, lipid-lowering agents, antidiabetics, antihypoglycaemic agents, hormones, iodine salts, thyrostatic agents, iron, vitamins, trace elements, antivirals, antifungals, antituberkulotics, substances for tumor-brain therapy and vaccines.

[0029] Furthermore, according to an embodiment said

medium carrying the analyte is a fluid medium, particularly an aqueous medium, particularly one of: a body fluid, blood, lymph, saliva, urine, gastric juice, secretions of the pancreatic bile, sweat, tears and the interstitial fluid, extra cellular fluid, breast milk, vaginal secretion, nasal secretion, ejaculate, menstrual fluid, aqueous humor of the eye, cerebral water, ascites, pleural fluid, pericardial fluid, synovia, amniotic liquor, cerumen and pus.

[0030] Furthermore, particularly in case the sensor system according to the invention is applied in the field of environmental / water analysis, analytes may be one of: allergens, plant protectant, pathogens, dioxins, temperature, humidity, conductivity, pH, dissolved oxygen, redox potential, total hardness, carbonate hardness, alkalinity, potential to solve chalk, bromine, chloride, sulfate, sulfite, fluoride, orthophosphate, total phosphorus, cyanide, organic nitrogen, ammonium, nitrate, nitrite, Kjeldahl nitrogen, total nitrogen, total nitrogen bound (TNb), aluminum, antimony, arsenic, barium, beryllium, lead, boron, cadmium, calcium, chromium, cobalt, iron, potassium, copper, magnesium, manganese, molybdenum, sodium, nickel, mercury, sulfur, selenium, silver, tellurium, thallium, titanium, vanadium, Zinc, tin, lithium, TOC, DOC, BOD5, COD, permanganate index, phenol total steam, total phenol (phenoline index), AOX, anionic surfactants, nonionic surfactants, lipophilic substances, hydrocarbon index, halogenated hydrocarbons, Mononuclear aromatic hydrocarbons (B, T, X), polychlorinated biphenyls (PCB), polycyclic aromatic hydrocarbons (PAHs), chlorophenols and pesticides (chlorinated pesticides, triazines).

[0031] Furthermore, according to an embodiment of the present invention, when the analyte is a metal ion, e.g. K⁺, the concentration of crown ether in the hydrogel preferably lies within the range from 0 to 40% and most preferably in the range from 3 to 25%.

[0032] Furthermore, according to an embodiment of the present invention, when the analyte is glucose, the concentration of said boronic acid moiety in the hydrogel preferably lies within the range from 0 to 45% and most preferably in the range from 4 to 30%.

[0033] Furthermore, according to an embodiment, the degree of cross-linking in the hydrogel is adjusted in such a manner that the hydrogel exhibits structural integrity entirely when swollen to the maximum extent. Such a degree of crosslinking has the advantage that the hydrogel is not pressed through the supporting structure of the chamber of the sensor.

[0034] Furthermore, according to an embodiment of the present invention, the responsive component or compound is homogeneously distributed in the hydrogel. Alternatively, the distribution of the responsive component may also be inhomogeneously in the hydrogel and thus the responsive component resides in concentrated regions.

[0035] According to yet another embodiment of the present invention related to the analyte potassium, said hydrogel residing in said chamber comprises a temper-

ature coefficient $K_T = \Delta p / \Delta T$ being the slope of the p-T-curve of the hydrogel (e.g. as measured by the pressure sensor measuring the pressure in said chamber) that lies within the range of 200 mbar/K to 600 mbar/K, and/or a hydrogel $K_{HG} = K_{K^+} / K_T$ in the range from 0,001 to 0,2 and most preferably in the range from 0,004 to 0,1. The hydrogel coefficient K_{HG} describes the concentration dependent (left) shift of the p-T-curve.

[0036] Particularly, these hydrogel specific parameters can be derived from the p-T-graph associated to the hydrogel residing in said chamber of the sensor head. These parameters can be used to characterize the hydrogel. Also the sensitivity K_A (or K_{K^+} in case of K^+) can be derived from the p-T-curve. Due to the analyte-dependent shift of the p-T-curve, a pressure change occurs at each given temperature. For instance, in Fig. 6 a pressure reduction of about -400 mbar in the chamber is observed at 28°C when adding 20 mM KNO_3 to the hydrogel (K^+ is captured). Consequently, the sensitivity derived therefrom is

$$K_{K^+} = -400 \text{ mbar} / 20 \text{ mM} = -20 \text{ mbar/mM}$$

for the specific hydrogel.

[0037] In contrast to a free (isobaric) swelling of the hydrogel, the sensitivity in the isochoric process according to the present invention is essentially constant over broad range of temperatures.

[0038] Particularly, according to a preferred embodiment of the present invention, the hydrogel comprises a polymer of the monomer PNIPAAm.

[0039] Alternatively, one of the following polymers may be used:

- Poly(N-isopropylmethacrylamide) (LCST: 38-42 °C),
- Poly(N-ethyl-N-methyl acrylamide) (LCST: 56 °C),
- Poly(N,N-diethyl acrylamide) (LCST: 32 °C),
- Poly(N,N-dimethylaminoethyl methacrylate) (LCST: 50 °C),
- Oligo(ethylenglycol)methacrylate and poly(oligo(ethylene glycol)methacrylate) (LCST: 26-90 °C).

[0040] Particularly, according to an embodiment of the present invention, the hydrogel comprises a network comprising a plurality of cross-linked polymer chains wherein a plurality of non-cross linked (e.g. linear) polymer chains are bound to said network, wherein only said non-crosslinked polymer chains comprise said responsive component/compound. Particularly, according to an embodiment, the molar fraction of the responsive compound within the respective non-cross linked polymer chain is in the region between 30-70%, particularly 50%.

[0041] Furthermore, according to a preferred embodiment of the present invention, the sensor system comprises an analyzing unit that is configured to determine

the presence or concentration of the analyte using at least the pressure measured in the chamber of the sensor head of the sensor by means of the pressure sensor.

[0042] Furthermore, according to a preferred embodiment of the present invention, the sensor system further comprises a telemetry unit that is configured to wirelessly transfer the measured pressure to the (e.g. remote) analyzing unit.

[0043] Furthermore, according to preferred embodiment of the present invention, the sensor head (and particularly also further sensors or reference sensors of the sensor system) is configured to be implantable into a human or animal body.

[0044] Further, according to preferred embodiment of the present invention, for reducing a cross-sensitivity of the sensor, the sensor system comprises at least one reference sensor which comprises a reference hydrogel, which corresponds to said hydrogel in said chamber of the sensor, but without said responsive components (such that the reference sensor / reference hydrogel has the same or at least a similar cross-sensitivity with respect to salts and the same or at least a similar temperature dependence as the hydrogel in the chamber of the sensor). Particularly, also the reference sensor may comprise a sensor head having a chamber filled with said reference hydrogel and a pressure sensor for measuring a reference pressure in said chamber of the reference sensor. Particularly, the analyzing unit is configured to compensate/correct the pressure measured in the chamber of the sensor with the reference pressure. The difference of both pressures eliminates cross-sensitivity.

[0045] Here, the invention makes use of the surprising findings according to which the effects of the components of the hydrogel with respect to the measured pressure of the reference hydrogel are additive also in complex solutions (e.g. cell culture medium). This eases compensation of cross-sensitivities of e.g. potassium sensitive hydrogels by means of a reference sensor.

[0046] Particularly, the reference sensor may be used to determine disturbances/cross-sensitivities such as e.g. ion concentration, temperature dependence and aging of the hydrogel / polymer network due to degrading processes.

[0047] According to an embodiment, the sensor system may comprise a further second sensor sensitive to another analyte, say analyte B, in contrast to analyte A detected by said (first) sensor. The second sensor may also use a thermo-responsive hydrogel. Here, the sensor system may comprise a further (second) reference sensor that is configured to determine cross-sensitivities with respect to the second sensor. This may also be used to determine/eliminate cross sensitivities concerning the first sensor and analyte B and the second sensor and analyte A. Particularly, the sensor system may comprise an arbitrary number of sensors and corresponding reference sensors.

[0048] Particularly, according to an embodiment, the sensor and the at least one reference sensor may com-

prise the same hydrogel, wherein now the sensor head of the reference sensor is not permeable for the analyte. Particularly, a pore size of said permeable area (e.g. switchable membrane) of the sensor head of the reference sensor is selected such that the analyte cannot pass the permeable area. Possible degradation processes and cross sensitivities occur in both sensors. During idle times when no pressure measurements are taken place said permeable areas are preferably closed.

[0049] Furthermore, also the reference sensor may comprise a telemetry unit for transmitting the measured reference pressure to the analyzing unit. However, the reference sensor may also use the telemetry unit of said sensor (e.g. common telemetry unit).

[0050] Furthermore, according to preferred embodiment of the sensor system according to the present invention, the sensor system comprises a temperature sensor and/or a separate pressure sensor (without a hydrogel) in order to sample further data for reducing cross sensitivities, particularly for compensating blood pressure variations.

[0051] Further, according to preferred embodiment of the sensor system according to the present invention, the sensor system comprises an external pressure sensor for reducing the influence of external barometric pressure changes (e.g. in an ambient atmosphere surrounding it is the patient into whom the sensor head and particularly the at least one reference sensor are implanted).

[0052] Said barometric pressure sensor can be integrated into an external patient device or into an external therapy device. Furthermore, said barometric pressure sensor may be integrated into an external device (e.g. patient or therapy device) that also comprises said analyzing unit to which pressure data is sent by the telemetry unit of the sensor.

[0053] Set external device can be configured to be linked to the sensor (e.g. implanted into the patient) via communication connection, particularly a wireless communication connection, for instance a radio communication connection. Here, the measured barometric pressure can be simultaneously detected and offset with the pressure measured by the sensor in said chamber of the sensor.

[0054] Furthermore, according to yet another embodiment of the present invention, said sensor of the sensor system comprises a component which is configured to mechanically decouple a change in the pressure of the hydrogel in said chamber resulting from a change in temperature of the hydrogel from the concentration dependent pressure measured by the pressure sensor of said sensor.

[0055] Particularly, for this, the pressure sensor may be arranged in the compartment separated from said chamber by a flexible pressure membrane (e.g. bellows, polymer film) to which the hydrogel residing in said chamber is coupled. Particularly said compartment may be filled with a material (e.g. a fluid), such as an oil, which material comprises a positive temperature coefficient

and particularly functions as a compensation component. In contrast thereto, the hydrogel comprises a negative temperature coefficient. In case the hydrogels temperature increases, the hydrogel shrinks whereas the material arranged in said compartment expands. Particularly, the amount of said material is selected such, that the increase in volume of the material with temperature exactly or at least partly compensates the temperature induced decreasing volume of the hydrogel.

[0056] Further, according to yet another aspect of the present invention, a method for detecting or determining the concentration of an analyte in a medium is disclosed, wherein a sensor system according to the present invention is used for detecting the presence or concentration of said analyte.

[0057] Further features and embodiments of the present invention shall be described with reference to the Figures, wherein

20 Fig. 1 a graphical illustration of the volume transition of a hydrogel in isobaric conditions;

Fig. 2 a schematic illustration of a sensor system according to the invention;

25 Fig. 3 a 2:1 crown ether complex (responsive compound for capturing metal ions, particularly K^+);

Fig. 4 different polymer structures of a hydrogel that can be used with the present invention: (A), (B), (C), (D), (E);

30 Fig. 5 shows the P-T-curve (e.g. measured pressure over temperature) of an isochoric hydrogel according to the present invention (without analyte);

35 Fig. 6 shows hydrogel specific parameters as derived from the p-T-curve of the isochoric hydrogel;

40 Fig. 7 shows hydrogel coefficient for a cross-linked hydrogel (squares) in comparison to linear hydrogels (diamonds);

45 Fig. 8 shows a preferred configuration of crown ether in a cross-linked hydrogel according to the present invention for capturing metal ions, particularly K^+ , and

50 Fig. 9 shows different possible structures ((A) to (E)) of a hydrogel according to the present invention.

[0058] Figure 2 shows a schematic representation of a sensor system 1 according to the present invention. As indicated in Fig. 2, the sensor system 1 comprises for detecting the presence or concentration of an analyte A in a medium M at least an (e.g. implantable) sensor 10

having an sensor head 100 enclosing a chamber 101, which sensor head 100 further comprises a permeable area 105 through which the analyte A can pass into the chamber 101 when the sensor head 100 contacts said medium M.

Further, the sensor 10 comprises a cross-linked hydrogel H filling said chamber 101, which hydrogel H is configured to undergo a change in volume when contacting the analyte A passed into the chamber 101 which leads to a change in pressure in said chamber (101), and a pressure sensor 102 configured to measure the pressure P in said chamber 101 for detecting the presence or concentration of said analyte A.

[0059] Particularly, as indicated in Fig. 1, the hydrogel H comprises a lower critical solution temperature LCST, which decreases (from LCSTb to LCSTa) when the analyte A, here K^+ , bonds to the hydrogel H and which increases when the analyte A is released from the hydrogel H.

[0060] However, due to the fact that the hydrogel H according to the present invention is kept in an isochoric state in said chamber 101, the pressure measured in said chamber by means of the pressure sensor 102 surprisingly depends essentially linearly (and/or mildly quadratically) on the temperature of the hydrogel H in said chamber 101 as indicated in Fig. 4, which leads to a constant sensitivity of the sensor 10 for the analyte A, here K^+ , over a broad range of temperatures. In contrast thereto, Fig. 3 shows the sigmoidal curves (swelling degree vs. temperature for free swelling which result in case of isobaric measurements that disadvantageously comprise a rather small measuring window of only a few degrees in temperature.

[0061] The hydrogel H according to the invention can be characterized by the following parameters that can be derived from the pressure-temperature curve (p-T curve) as shown in Fig. 6

[0062] The first of these parameters is the temperature coefficient $K_T = \Delta p / \Delta T$ which is the slope of the (isochoric) p-T-curve of the hydrogel H measured by pressure sensor 102 that preferably lies within the range of 200 mbar/K to 600 mbar/K.

[0063] A further characteristic parameter is the sensitivity for the respective analyte (here e.g. K^+) which is denoted as K_{K^+} . As indicated in Fig. 6, the latter quantity can also be easily derived from the p-T-curve. Due to the potassium-dependent shift of the p-T-curve, a pressure change occurs at each given temperature. For instance, in Fig. 6 a pressure reduction of about -400 mbar occurs in the chamber 101 at 28 °C due to adding of 20 mM KNO_3 to the hydrogel H. Thus, the corresponding sensitivity is $K_{K^+} = -400 \text{ mbar} / 20 \text{ mM} = -20 \text{ mbar/mM}$ for the hydrogel N residing in chamber 101.

[0064] A third parameter is the so called hydrogel coefficient, which is defined as $K_{HG} = K_{K^+} / K_T$ and described the concentration dependent (left) shift of the p-T-curve. Preferably $K_{HG} = K_{K^+} / K_T$ lies in the range from 0,001 to 0,2 and most preferably in the range from 0,004 to 0,1.

[0065] In contrast to a free (isobaric) swelling of the hydrogel (cf. Fig. 3), the sensitivity in the isochoric process according to the present invention is essentially constant over a broad range of temperatures as can be seen from Figs. 4 and 6.

[0066] Furthermore, so far, only linear 2:1 hydrogels have been discussed which comprise depending on potassium concentration a high sensitivity in a narrow temperature region. However, until now, for the cross-linked variants high sensitivities have not been observed. The high sensitivities of the linear polymers are possible due to a high fraction of crown ether in the respective polymer, particularly - depending on the crown ether fraction - the hydrogel coefficient K_{HG} can be increased by increasing the crown ether fraction in the linear hydrogel, i.e., in case of a fraction of 15 mol-% a coefficient K_{HG} being larger than 0,3 K/mM is observed (cf. Fig. 7), which corresponds to a shift of the swelling degree temperature curve of more than 6 K when adding 20 mM KNO_3 in water.

[0067] Surprisingly, the hydrogel coefficients K_{HG} for the linear and cross-linked hydrogels correlate very well (cf. Fig. 7), wherein the sole parameter seems to be the crown ether fraction. This is intriguing since the detection of potassium in a 2:1 complex two crown ethers are required. The original assumption according to which linear hydrogels can fold more easily than 3D cross-linked polymers so that the crown ethers can come more easily in close proximity can thus not be verified. However, in case of cross-linked hydrogels an increase of the crown ether fraction is not easily possible without further ado, since the relatively large crown ether monomers disturb the cross-linking sterically.

[0068] Adapting the synthesis accordingly, cross-linked hydrogels having a crown ether fraction of 10% could be generated.

[0069] Suitable hydrogels can be prepared according to the following exemplary procedure:

The monomers of N-isopropylacrylamid, acrylamide and 4-acrylamidobenzo-15-crown-5 (B15C5Am) as well as the crosslinking agent N, N'-methylenebis(acrylic acid amide) (MBAAM) were solved in KNO_3 solution in a vial equipped septum and magnetic stir bar, degassed for 15 minutes upon argon supply and cooled to 15 °C. Subsequently an enhancing agent tetramethylethylenediamine (TEMED) and an initiator (ammonium persulfate, APS) were added to the KNO_3 solution and the resulting solution is degassed for about 90 sec. Thereafter the reaction solution is transferred quickly to argon flooded reactor. After reacting for 24 hours at 15 °C the resulting hydrogel is washed with water (MilliQ water) up to 6 times.

[0070] In order to significantly increase the fraction of crown ether, the following strategies can be applied.

[0071] As indicated in Fig. 8, here in case of the analyte A in form of K^+ , the H of the sensor system 1 comprises

a plurality of a responsive component R, here crown ether, wherein the respective responsive component R is responsive to the analyte A such that the volume of the hydrogel H tends to decrease when the respective responsive component R binds to the analyte A such that the pressure P in said chamber 101 decreases, and wherein the volume of the hydrogel H tends to increase when an analyte A is released from the respective responsive component R. Particularly, as indicated in Fig. 8, said crown ether molecules R are attached such to the hydrogel H that they form pairs, particularly in a chelation configuration, wherein the two crown ethers of the respective pair are configured to bind the analyte A simultaneously, wherein particularly the analyte A is arranged between the crown ethers of the respective pair in its bound state, as shown in Fig. 8. Particularly, the two crown ethers can be generated in said chelation configuration before the actual hydrogel synthesis. The configuration of crown ethers shown in Fig. 8 is also denoted as pincer structure.

[0072] For this, the two crown ethers are spatially and covalently bonded in the form of said pincer by using suitable synthesis parameters. This allows an easy increase of the crown ether amount by a factor of two. Furthermore, the crown ethers are already positioned in close proximity for the potassium detection, so that a reaction due a changing potassium concentration can take place much faster than using a conventional approach, since polymer chains do not have to be re-oriented for finding potassium due to said crown ether sandwich.

[0073] Further, according to an embodiment indicated in Fig. 9 (A), the crown ether fraction may be enhanced by providing a hydrogel H that comprises a network N, e.g. a PNIPAAm network, comprising a plurality of cross-linked polymer chains wherein a plurality of non-cross linked polymer chains C is bound to said network N (and distributed over the network), wherein only said non-crosslinked (e.g. linear) polymer chains C comprise said responsive component R (e.g. said crown ether structure). The molar fraction of crown ether in the non-cross linked (e.g. linear) chains preferably lies in the range about 40 to 60 % and is preferably approx. 50%. Particularly, a free radical polymerization can be used for producing this special hydrogel, in which the individual components are distributed inhomogeneously in the network in a defined manner. Particularly a PNIPAAm network without crown ethers can be provided, in which linear (non-crosslinked) areas with high crown ether fraction are incorporated by polymerization.

[0074] Alternatively, the PNIPAAm network N may comprise a high fraction of crown ether. The linear (non-cross linked) areas can be configured as before.

[0075] The generation of said linear regions C with a high crown ether fraction in the hydrogel H can also be achieved by configuring the polymer of the hydrogel H as a star polymer (cf. Fig. 9(B)), as a dendrimer (cf. Fig. 9 (D)), as a blend (cf. Fig. 9 (C)), or as a copolymer, particularly as a graft polymer or toothbrush-like polymer

(cf. Fig. 9 (E)).

[0076] As an alternative to the free radical polymerization also strategies for a controlled radical polymerization can be used (e.g. NMP, ATRP, RAFT).

5 **[0077]** As also indicated in Fig. 2, the sensor system 1 may comprise further components.

[0078] Particularly, in an embodiment, the sensor system 1 further comprises an analyzing unit 2 that is configured to determine the presence or concentration of the respective analyte A using the measured pressure P.

10 **[0079]** Preferably, for transmitting the measured pressures P to the unit 2, preferably wirelessly, the sensor 10 may comprise a telemetry unit 104 that communicates with analyzing unit 2.

15 **[0080]** This allows to implant the sensor 10 into a patient, while the analyzing unit can be arranged outside the patient.

[0081] Furthermore, for reducing cross-sensitivity of the sensor 10, the sensor system 1 may comprise at least one reference sensor 11 that comprises a reference hydrogel H'. The reference hydrogel H' can be identical to the hydrogel H wherein now the analyte A is not allowed to enter the chamber 101 of the sensor head 100 of the reference sensor 11. For this the reference sensor 11 may comprise a suitable area 105 or no such area at all. Alternatively, the reference hydrogel H may be configured as the hydrogel H, but does not comprise the responsive component R so that a change in analyte concentration in the chamber 101 of the reference sensor 11 does not affect the reference pressure measured by means of the pressure sensor 102 of the reference sensor.

20 **[0082]** Preferably, the analyzing unit 2 is configured to offset the pressure measured in the chamber 101 of the sensor 10 against the reference pressure measured by means of the pressure sensor 102 of the reference sensor 11.

25 **[0083]** Particularly, in an embodiment, the sensor system 1 may comprise multiple sensors 10, which may be identically configured, with the exception that each sensor 10 comprises a hydrogel that is sensitive to a different analyte. Further, such a system 1 may comprise a corresponding number of reference sensors 11 to reduce/eliminate cross-sensitivities and other disturbances.

30 **[0084]** For further reduction of such cross-sensitivities and disturbances, the sensor system may further comprise a separate temperature sensor 20 and/or pressure sensor 30 that may also communicate with the analyzing unit 2.

35 **[0085]** Furthermore, the sensor system may comprise an external pressure sensor 40 for reducing the influence of external barometric pressure changes on the pressure measurements in the sensors 10 and/or 11.

40 **[0086]** Finally, as indicated in Fig. 2, each sensor 10, 11 or individual sensors of the system 1 may optionally comprise a component 103, O which mechanically decouples a change in the pressure of the hydrogel H, H'

in the respective chamber 101 resulting from a change in temperature of the hydrogel H from the concentration dependent pressure P measured by the respective pressure sensor 102. Particularly, the respective pressure sensor 102 may be arranged in compartment 101 a separated from said chamber 101 by a flexible pressure membrane (e.g. bellows, polymer film) 103 to which the hydrogel H residing in the respective chamber 101 is mechanically coupled (e.g. contacts said pressure membrane 103). Particularly, the respective compartment 101a may be filled with a material (e.g. a fluid), such as an oil O, which material O comprises a positive temperature coefficient and particularly functions as a compensation component. In contrast thereto, the hydrogel H (H') comprises a negative temperature coefficient. In case the hydrogel's H temperature increases, the hydrogel H shrinks, whereas the material O arranged in said compartment 101a expands. Particularly, the amount of said material O is selected such that the increase in volume of the material O with temperature exactly or at least partly compensates the temperature induced decreasing volume of the hydrogel.

Claims

1. A sensor system (1) for detecting the presence or concentration of an analyte (A) in a medium (M), comprising:

- a sensor (10) having a sensor head (100) comprising a chamber (101), which sensor head (100) comprises a permeable area (105) through which the analyte (A) can pass into the chamber (101) when the sensor head (100) contacts said medium (M),
- a cross-linked hydrogel (H) filling said chamber (101), said hydrogel (H) is configured to undergo a change in volume when contacting the analyte (A) passed into the chamber (101) which leads to a change in pressure in said chamber (101), and
- a pressure sensor (102) configured to measure the pressure (P) in said chamber (101) for detecting the presence or concentration of said analyte (A).

2. The sensor system (1) according to claim 1, **characterized in that** the hydrogel (H) comprises a plurality of a responsive component (R), wherein the respective responsive component (R) is responsive to the analyte (A) such that the volume of the hydrogel (H) tends to decrease when the respective responsive component (R) binds to an analyte (A) such that the pressure (P) in said chamber (101) decreases, and wherein the volume of the hydrogel (H) tends to increase when an analyte (A) is released from the respective responsive component (R) such that the

pressure (P) in said chamber (101) increases.

3. The sensor system (1) according to claim 2, **characterized in that** the responsive component (R) comprises one of:

- a crown ether, wherein particularly the analyte is K^+ ,
- a boronic acid moiety, wherein particularly the analyte is glucose.

4. The sensor system (1) according to claim 3, **characterized in that** said crown ether molecules (R) are attached such to the hydrogel (H) that they form pairs, wherein the two crown ethers of the respective pair are configured to bind an analyte (A) simultaneously, wherein particularly the analyte (A) is arranged between the crown ethers of the respective pair in its bound state.

5. The sensor system (1) according to one of the preceding claims, characterized in that the hydrogel (H) comprises at least one of:

- a temperature coefficient $K_T = \Delta p / \Delta T$ being the slope of the p-T-curve of the hydrogel (H), which temperature coefficient K_T preferably lies within the range of 200 mbar/K to 600 mbar/K,
- a hydrogel coefficient $K_{HG} = K_K + K_T$ in the range from 0,001 to 0,2.

6. The sensor system (1) according to one of the preceding claims, **characterized in that** the hydrogel (H) comprises a polymer of one of the following monomers: N-isopropylmethacrylamide, N-ethyl-N-methyl acrylamide, N,N-diethyl acrylamide, N,N-dimethylaminoethyl methacrylate and (ethylenglycol)methacrylate.

7. The sensor system (1) according to claim 3 or one of the claims 3 to 6 when referring to claim 3, **characterized in that** the hydrogel (H) comprises a network (N) comprising a plurality of cross-linked polymer chains wherein a plurality of non-cross linked polymer chains (C) are bound to said network (N), and wherein only said non-crosslinked polymer chains (C) comprise said responsive component (R).

8. The sensor system (1) according to one of the preceding claims, **characterized in that** the sensor (10) comprises an analyzing unit (2) that is configured to determine the presence or concentration of the analyte (A) using the measured pressure (P).

9. The sensor system according to claim 8, **characterized in that** the sensor (10) comprises a telemetry unit (104) that is configured to transfer the measured pressure (P) to the analyzing unit (2).

10. The sensor system (1) according to one of the preceding claims, **characterized in that**, for reducing cross-sensitivity of the sensor (10), the sensor system (1) comprises at least one reference sensor (11) which comprises a reference hydrogel (H'), wherein particularly the reference sensor (11) also comprise a sensor head (100) having a chamber (101) filled with said reference hydrogel (H) and a pressure sensor (102) for measuring a reference pressure in said chamber (101) of the reference sensor (11), wherein particularly the analyzing unit is configured to correct the pressure measured in the chamber (101) of the sensor (10) with or by subtracting the reference pressure.
11. The sensor system according to one of the preceding claims, **characterized in that** said sensor (10) comprises a component (103, O) which mechanically decouples a change in the pressure of the hydrogel (H) in said chamber (101) resulting from a change in temperature of the hydrogel (H) from the analyte concentration dependent pressure (P) measured by the pressure sensor (102) of said sensor (10).
12. Crosslinked hydrogel on basis of Poly(N-isopropylacrylamid) having at least 20 mol-% crown-ether chemically bonded to the polymer chains.
13. Hydrogel according to claim 12, wherein the crown ether is 15-crown-5.
14. Use of a hydrogel according to claim 12 or 13 for a sensor for analyzing concentrations.
15. Use according to claim 14, wherein the concentrations relate to analytes in body fluids.

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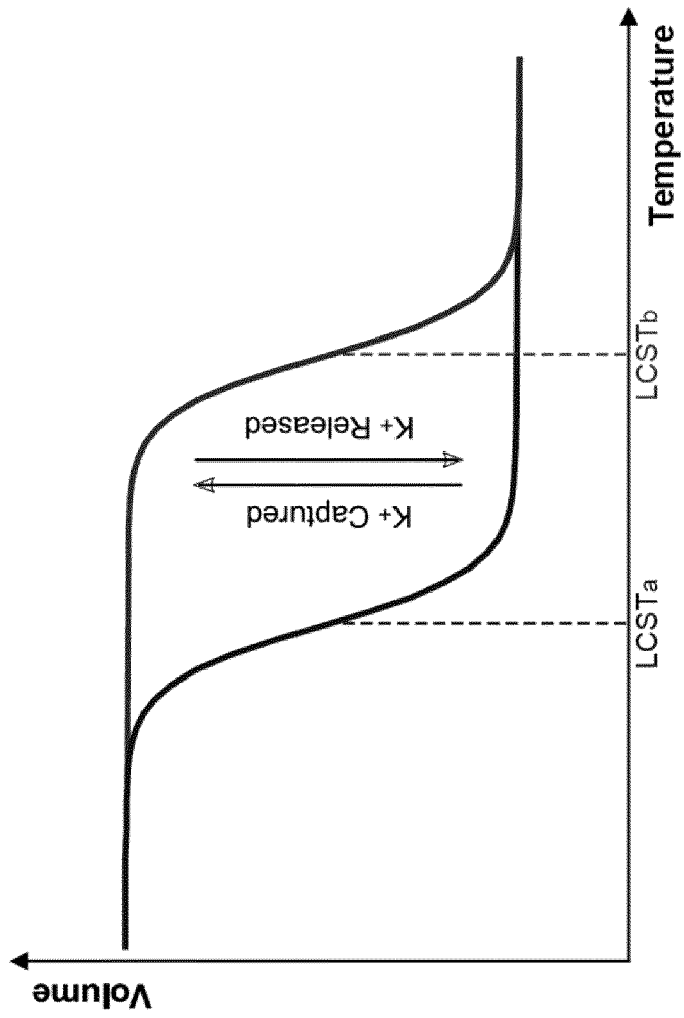


FIG. 1

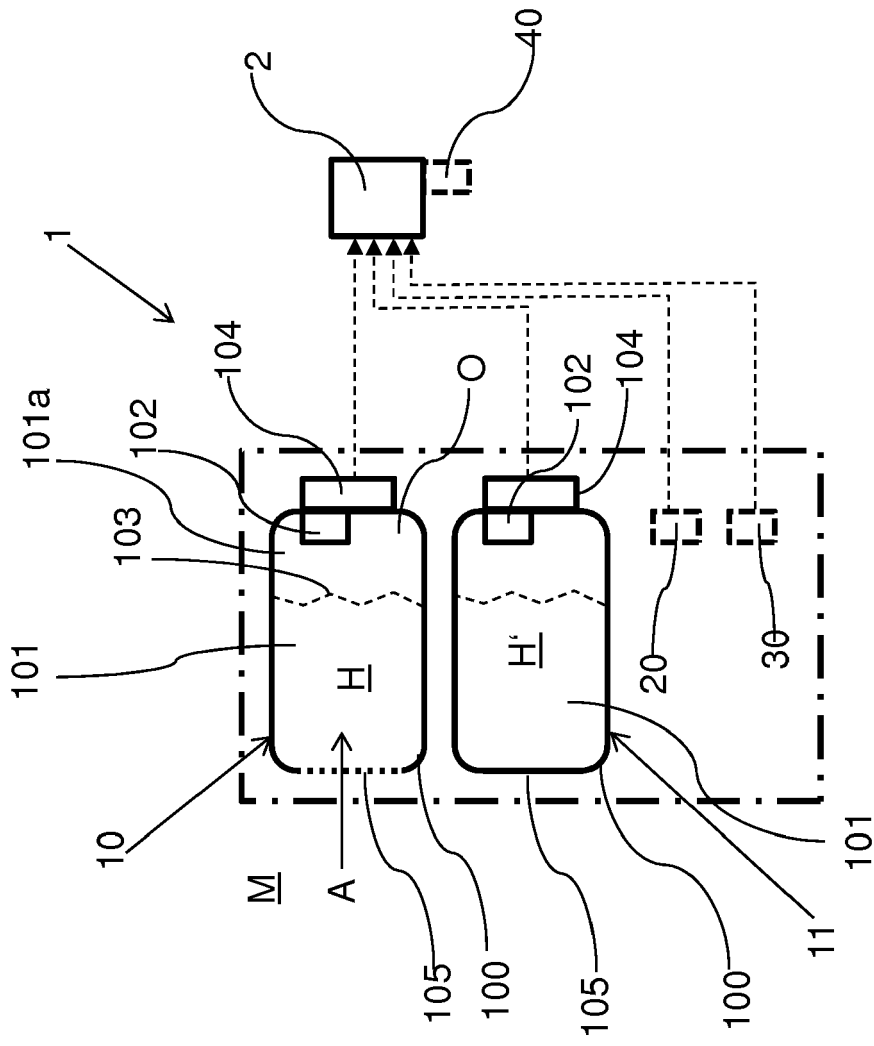


FIG. 2

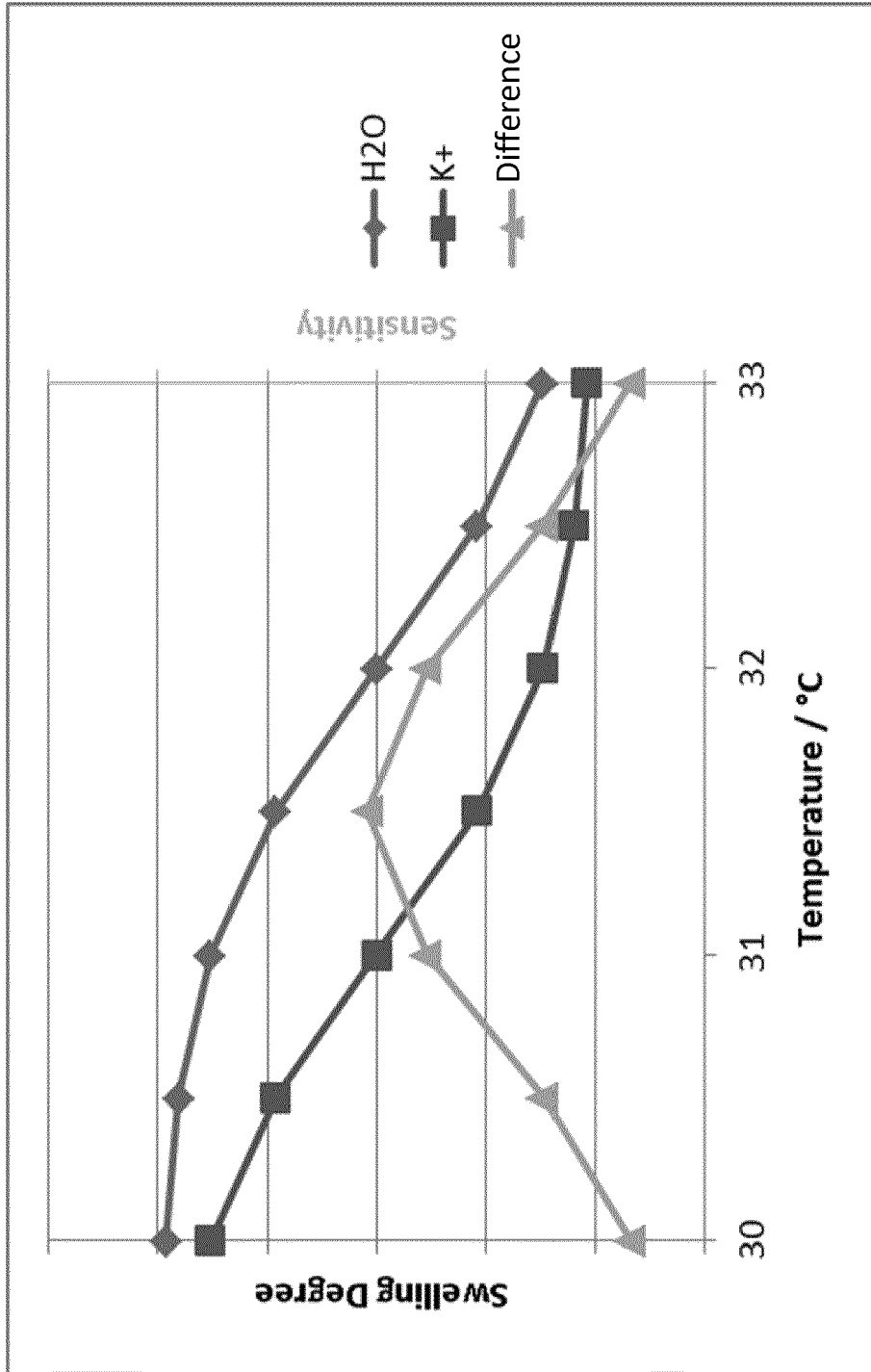


FIG. 3

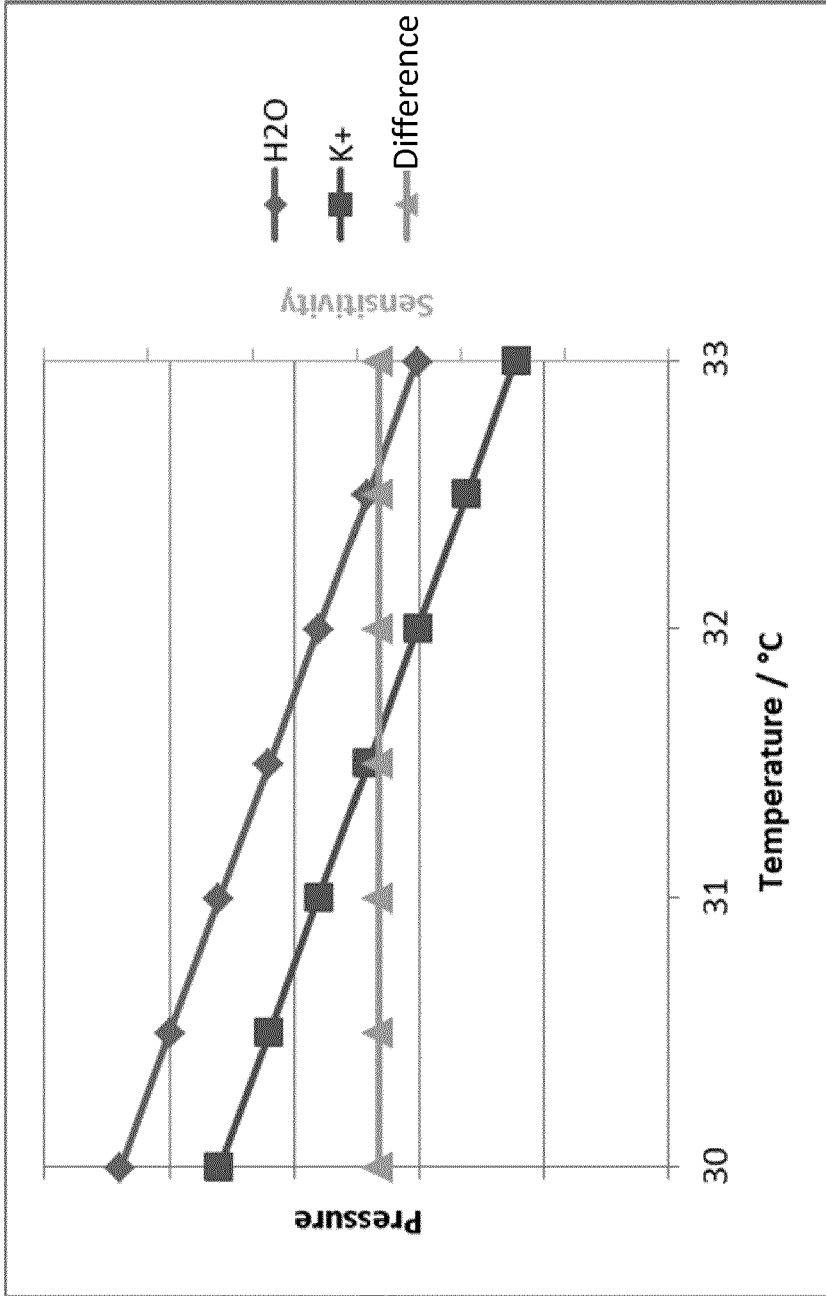


FIG. 4

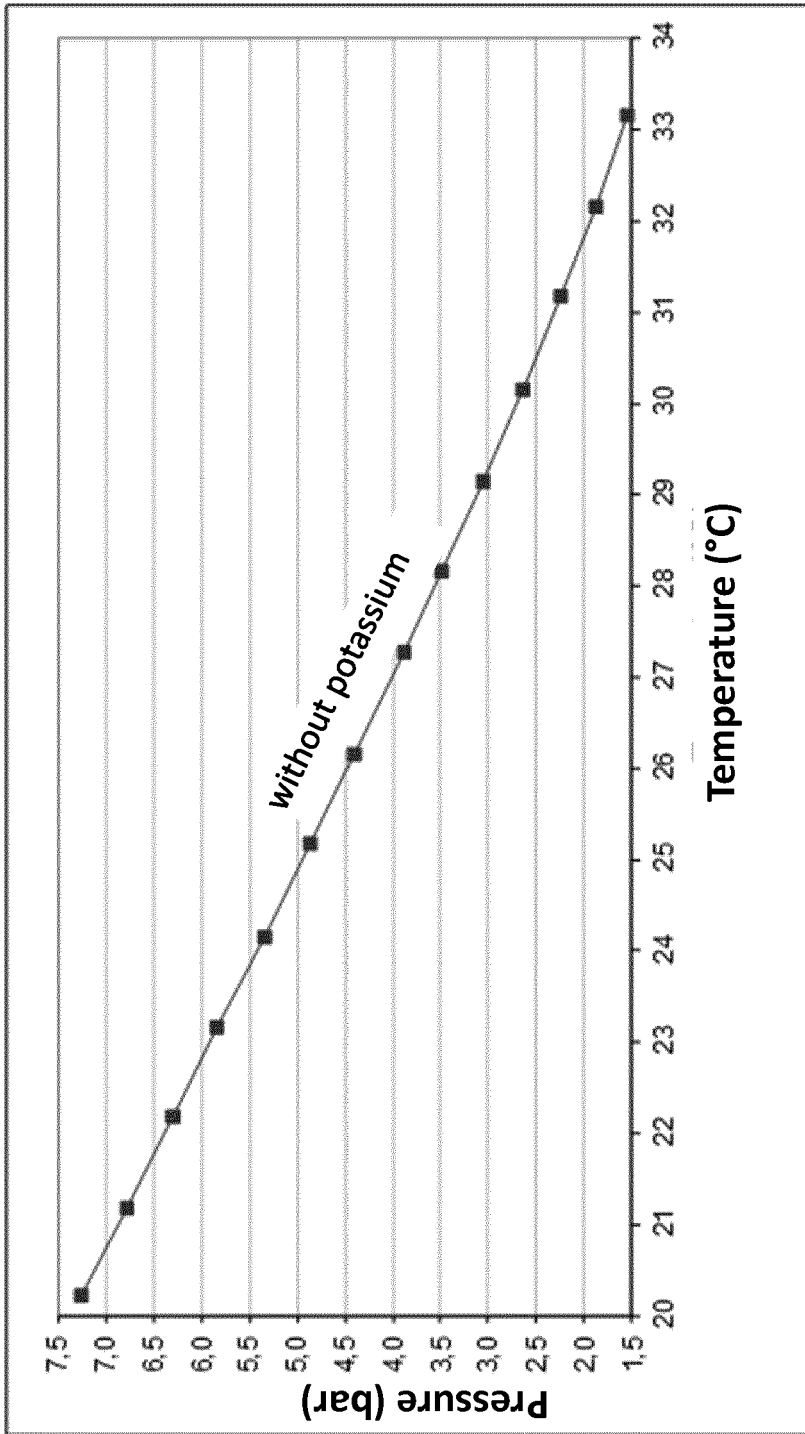


FIG. 5

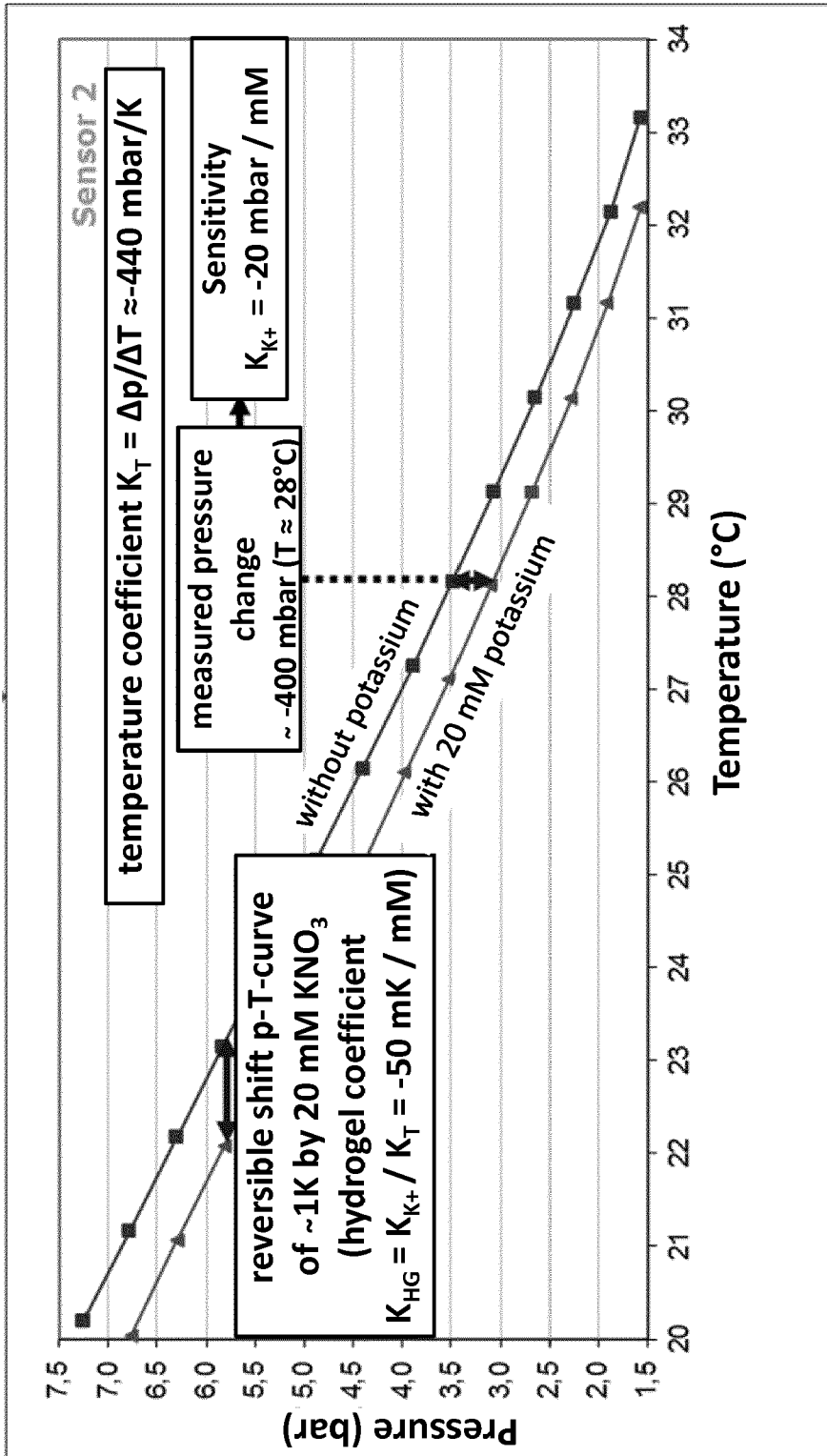


FIG. 6

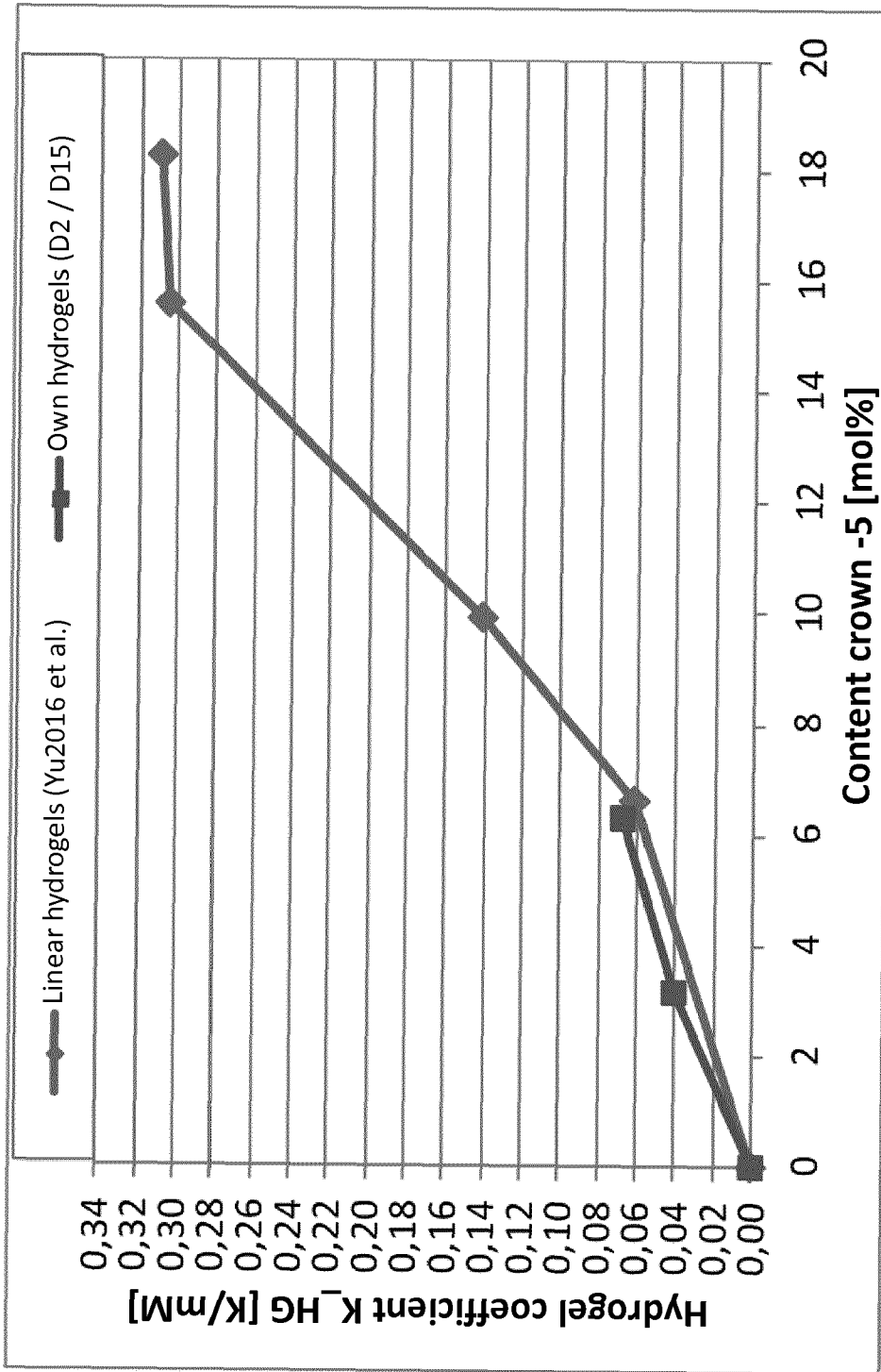


FIG. 7



FIG. 8

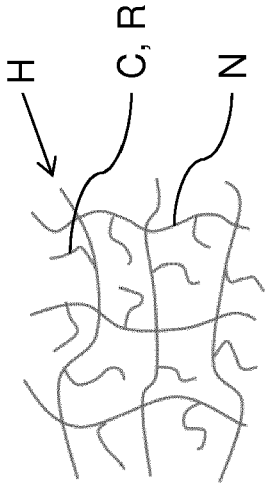


FIG. 9A

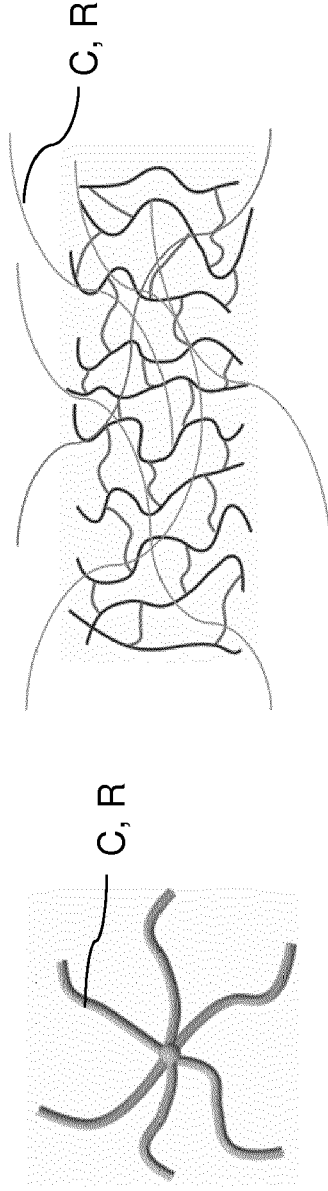


FIG. 9B



FIG. 9C

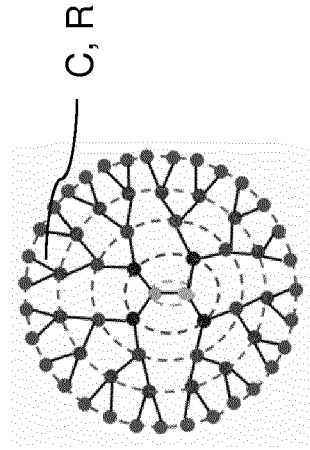


FIG. 9D

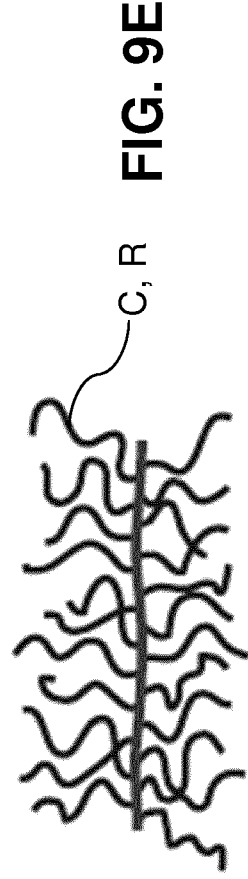


FIG. 9E



EUROPEAN SEARCH REPORT

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The present search report has been drawn up for all claims				
Place of search Munich		Date of completion of the search 11 September 2017	Examiner Behrens, Ralf	
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The present search report has been drawn up for all claims			
Place of search Munich		Date of completion of the search 11 September 2017	Examiner Behrens, Ralf
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CLAIMS INCURRING FEES

The present European patent application comprised at the time of filing claims for which payment was due.

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Only part of the claims have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due and for those claims for which claims fees have been paid, namely claim(s):

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No claims fees have been paid within the prescribed time limit. The present European search report has been drawn up for those claims for which no payment was due.

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LACK OF UNITY OF INVENTION

The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

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see sheet B

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All further search fees have been paid within the fixed time limit. The present European search report has been drawn up for all claims.

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As all searchable claims could be searched without effort justifying an additional fee, the Search Division did not invite payment of any additional fee.

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Only part of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the inventions in respect of which search fees have been paid, namely claims:

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None of the further search fees have been paid within the fixed time limit. The present European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims, namely claims:

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The present supplementary European search report has been drawn up for those parts of the European patent application which relate to the invention first mentioned in the claims (Rule 164 (1) EPC).

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**LACK OF UNITY OF INVENTION
SHEET B**Application Number
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The Search Division considers that the present European patent application does not comply with the requirements of unity of invention and relates to several inventions or groups of inventions, namely:

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1. claims: 1-11(partially)

Sensor of claim 1, characterised in that the responsive component (R) comprises a crown ether.

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2. claims: 1-11(partially)

Sensor of claim 1, characterised in that the responsive component (R) comprises a boronic acid moiety.

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3. claims: 12-15

Cross-linked hydrogel of claim 12 and its use.

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ANNEX TO THE EUROPEAN SEARCH REPORT
ON EUROPEAN PATENT APPLICATION NO.

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This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report. The members are as contained in the European Patent Office EDP file on
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11-09-2017

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