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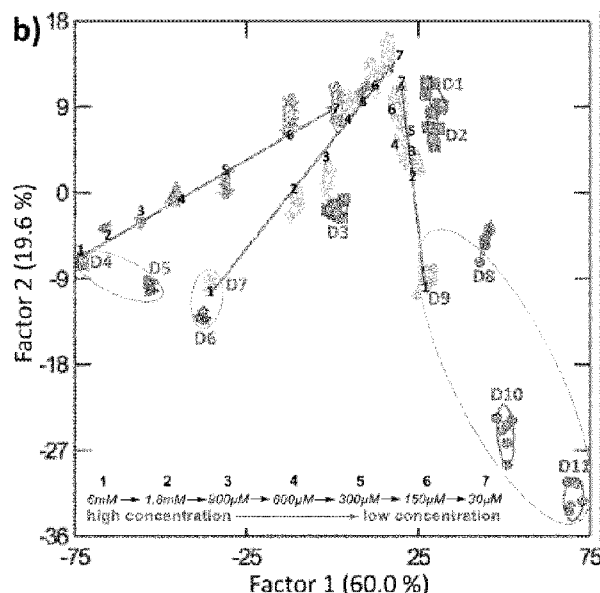
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(54) Title: METHOD AND SENSOR ARRAY FOR IDENTIFYING AN ANALYTE

Fig 5B



(57) **Abstract:** A method and sensor array for identification of an analyte is disclosed. The method comprises preparing a plurality of solutions at a plurality of pH values of at least one fluorescent poly(para-phenyleneethynylene) and its complex(es), exposing the complex analyte to the plurality of the solutions and measuring the fluorescence intensity of the exposed complex analyte. The fluorescence intensity is compared with a library and the complex analyte identified from the comparison.



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Description

Title: Method and Sensor Array for identifying an Analyte

5 **Cross-Relation to other applications**

[0001] This application claims benefit to and priority of UK Patent Application No. 1616038.4 filed on 21 September 2016.

Summary of the Invention

[0002] A method for the identification of an analyte using a sensor array using at least one
10 highly fluorescent, water-soluble polymer under different conditions is disclosed, as well
as a sensor array. The analytes identified include complex analytes, such as pharmaceutical
preparations and liquids, such as wine and fruit juice but also simple analytes, such as
carboxylic acids.

Background of the invention

15 [0003] Discrimination and identification of complex analytes such as, but not limited to,
fruit juices, alcoholic beverages, pharmaceutical preparations, and illegal drugs is an
important and interesting topic.

[0004] For example, falsified, stretched, filled or faked drugs are a serious health policy
20 problem that not only affects countries in the third world. Counterfeit antimalarials,
antibiotics, painkillers, life-style drugs, HIV drugs etc. are all known. Such counterfeit
drugs can cause drug-resistant bacterial and microbial strains to develop and spread. As a
result, quality control, identification and fingerprinting of the active compounds in drugs,

but also of the whole of the processed drug formulation (tablet, drops, capsules, suppositories) is an important task.

[0005] Similarly, quality control of food and other complex analytes is an important task. Many different types of analytical methods have been exploited for these tasks, including
5 mass spectrometry, electrochemical tongues and noses, as well as biological methods (antibodies, genetics).

[0006] One method known in the art is the use of chemo-optical tongues. These chemo-optical tongues can indicate the spoiling of fish, fingerprint coffees, whiskeys, beers, soft
10 drinks, red wines and white wines. The chemo-optical tongues react by color change or fluorescence intensity modulation. These chemo-optical tongues are comprised of sensor arrays of different chromophores or fluorophores or receptors that are bound to indicators or quenchers that are replaced by the analytes. The action principle of the chemo-optical
tongues is different from that of classic sensors but also of that of instrumental analytical
15 methods. Suslick²⁶ described some of the features that are presumably necessary to achieve successful discrimination for complex analytes and stressed that "...in general, an optimal sensor array for general sensing purposes will incorporate as much chemical diversity as possible...". This guided the development of colorimetric arrays, in which a wide variety
of different colorimetric indicator molecules are employed to identify analytes. Suslick's
20 printed libraries typically consist of 16-36 elements for successful identification of different classes of analytes.

[0007] A second accepted tenet of these chemo-optical tongues was formulated by Anslyn, and is a weakened variation of the lock and key-principle of Fischer as nicely shown in

Figure 1 of Ebeler, ref 27. In this picture, molecular keys fit into many locks with a varying degree of fit. Several of such partially fitting receptors identify and discriminate groups of analytes by the unique signal patterns of the sum of the sensor elements. Here the most practical approach is to offer small libraries of receptors that are “filled” with dyes to be
5 replaced by the analytes with different efficiency.ⁱ

[0008] These prior art approaches stress that cross-reactivity, structural differentiation and structural variation of the sensor elements are important, as expressed by the wish to obtain high dimensionality sensor arrays that differentiate a broad variety of similar, but complex
10 analytes, such as soft drinks, coffees, beers, whiskeys, etc.

[0009] Both described prior art approaches, i.e. the weakened lock and key principle but also the chemical diversity of the sensors are sufficient principles to guide the production of useful sensor arrays. These prior art approaches generate an arbitrary and large number
15 of working tongues and sensor elements, but neither predicts or defines the minimum structural variation in sensor elements necessary to discriminate complex analytes.

Summary of the Invention

[0010] An alternative approach is presented in this document. An array of charged fluorescent polymers in water at two different pH values is used as a four-element sensor,
20 which acts as an efficient chemical “tongue”. The sensor is able to discern different types of non-steroidal anti-inflammatory drugs (NSAID) and is also able to discriminate between different brands of ibuprofen and aspirin. The sensor could be formed from a microtitre plate, a microwell plate or a microfluidic array.

[0011] It has already been shown that different versions (including conjugated polymer-gold nanoparticle complexes,²⁰ conjugated polymer-green fluorescent protein complexes,²¹ and conjugated polymer-conjugated polymer complexes²²⁻²⁴) of this concept successfully discriminated anions, white wines, proteins, cells, cancer states in mammalian cells etc.

5 This document demonstrates the discrimination of 11 nonsteroidal anti-inflammatory drugs (NSAIDs, as shown in Figure 1), red wines, green, black and oolong teas and fruit juices. These complex analytes are sufficiently narrow in scope, yet have significant differences.

[0012] It would also be possible to use the sensor for identifying other dissolved complex
10 powder analytes.

Description of the Drawings

[0013] Fig. 1 shows structure and pKa value of the NSAIDs.

15 [0014] Fig. 2 shows **a)** Water solubility of NSAIDs **D1-D11** (6 mM) at different pH values. **b)** Structures of positively charged **P1** and negatively charged **P2**, used for analgesics sensing (ϕ = quantum yield). **c)** Final selected four sensing factors by using single **P1** and its electrostatic complex **C** (**P1** + **P2**).

20 [0015] Fig. 3A shows the synthesis of **P2**.

[0016] Fig. 3B shows the synthesis of **P4**.

[0017] Fig. 3C shows the synthesis of **P9**.

[0018] Figure 4A shows a fluorescence response pattern obtained with an array of **P1**, **C1-2** (each at pH10 and 13, buffered) treated with **D1-D11**.

[0019] Fig. 4B shows a 2D canonical score plot obtained with an array of **P1**, **C1-2** (each at pH10 and 13, buffered) treated with analgesics **D1-D11** (6 mM) and **D4**, **D7** and **D9** (from 30 μ M to 1.8 mM). Each point represents the response pattern for a single concentration of analgesics to the array.

[0020] Fig 5A shows the concentration dependencies of **D4**, **D7** and **D9** in an LDA plot.

[0021] Fig 5B shows the concentration dependencies of **D4**, **D7** and **D9** together with the remaining NSAIDs.

[0022] Figure 6A shows the fluorescence response pattern ($I - I_0 / I_0$) obtained by **P1** (500 nM, at pH10 and 13, buffered) and complex **C1-2** (**P1-P2** at 500 nM-250 nM, at pH10 and 13, buffered) treated with NSAIDs **D2** (aspirin, 6 mM, control) and **D8** (ibuprofen, 6 mM, control) and commercial available OTC tablet aspirin (**ASS1-ASS5**, 6 mM), ibuprofen (**IBU1-IBU5**, 6 mM).

[0023] Fig. 6B shows a 2D canonical score plot for the first two factors of simplified fluorescence response patterns obtained with an array of **P1**, **C1-2** (each at pH10 and 13, buffered) with 95% confidence ellipses. Each point represents the response pattern for a single analgesic to the sensor array.

[0024] Figure 7A shows a 2D canonical score plot and Figure 7B shows a 3D canonical score plot obtained with an array of **P1**, **C1-2** (each at pH10 and 13, buffered) treated with

NSAIDs **D1-D11** and commercial available OTC tablet aspirin (**ASS1-ASS5**), ibuprofen (**IBU1-IBU5**).

[0025] Figure 8A shows the results of a systematic evaluation and selection of the successful tongue elements of the sensor array for the juice sensing and Figure 8B shows the chemical structures and quantum yields (ϕ) of P1 and P2.

[0026] Figures 9A and 9B shows the fluorescence response patterns combined with LDA of all of the fruit juices.

[0027] Figures 10A-10C combines the response results from all juices after LDA.

[0028] Figure 11 shows the fluorescence response patterns of the self-made juices compared to commercial juices (black currant, green grapes, red grapes) and mixtures of red and green grape juices.

[0029] Fig. 12 shows the response pattern for three different types of teas.

[0030] Fig. 13 shows the PPEs 1-3 used for the preparation for complexes with C8 (also shown in Fig. 13).

[0031] Fig. 14 shows the PPEs for the discrimination of red wines.

[0032] Fig. 15 shows a microwell plate, similar to those used in this application as the sensor array.

Detailed Description of the Invention

[0033] Figure 1 shows the structure of eleven different types of NSAIDs chosen as a test bed in a first aspect of the concept. The structural similarity of the different NSAIDs

suggests their separation into four groups, viz. salicylates, fenamic acids, profens and arylacetic acids.

[0034] Figure 2A shows the water solubility of the NSAIDs at 6 mM concentration at different pH values and the selected four-member array.

5 [0035] It is known that a sensor comprising poly(*para*-aryleneethynylene)s (PAE) and their polyelectrolyte complexes discriminates 21 aromatic acids in aqueous solution (see Han, J.; Wang, B.; Bender, M.; Seehafer, K.; Bunz, U. H. F. Water-Soluble Poly(p-aryleneethynylene)s: A Sensor Array Discriminates Aromatic Carboxylic Acids. *ACS Appl. Mater. Interfaces* **2016**, 8, 20415-20421. The structures of the tested aromatic acids
10 are similar to that of the NSAIDs. Therefore, the optimal array for the aromatic acids known from the Han et al 2016 paper was selected as a starting point for discrimination of the NSAIDs.

[0036] Two types of elements work typically well within a sensor, particularly for sensors
15 comprised of the complexes shown in refs.22-24 and for PAE/Protein conjugates as reported previously in Ref 21: (1) individual highly fluorescent PAEs and (2) complexes composed of a fluorophore and a quencher-PAE. A number of PAEs were synthesized. The synthesis of **P1**, **P6**, **P10**, **P12** and **P13** have been reported in ref. 23. The synthesis of **P3** and **P7** have been reported in ref. 21. The synthesis of **P5** was reported in ref 28. The
20 synthesis of **P8** was reported in ref. 29. The synthesis of **P11** was reported in ref. 30.

[0037] The synthesis of **P2** is shown in Fig. 3A and was carried out as follows. Compound **1** (100 mg, 0.083 mmol) was dissolved in degassed CH₃CN/CHCl₃ (5 mL/2 mL). 1-Methyl-imidazole (1 mL) was added slowly and refluxed for 8 days under N₂ atmosphere.

After evaporation of the solvents, the mixture was re-dissolved in distilled water and then dialyzed against DI water for 7 days. Freeze-drying gave **P2** as yellow solid (99 mg, 86%). The M_n and PDI was determined on the precursor **1**. ^1H NMR (300 MHz, MeOD) δ = 8.32-8.93 (d, 2 H), 7.41-7.60 (m, 4 H), 7.31-7.10 (m, 4 H), 4.58-4.34 (m, 6H), 4.01-4.28 (m, 4 H), 3.76-3.84 (m, 6 H), 3.32-3.75 (m, 56 H), 3.11-3.25 (m, 12 H), 2.31-2.46 (m, 4 H) ppm. Due to low solubility, ^{13}C NMR spectrum could not be obtained. IR (cm^{-1}): ν 3410, 2871, 2359, 1647, 1575, 1508, 1490, 1470, 1420, 1350, 1272, 1200, 1088, 1042, 949, 849, 623. Quantum yields (Φ = 0.29).

[0038] Fig 3B shows the synthesis of **P4**, which was carried out as follows. Compound **1** (275 mg, 0.228 mmol) was dissolved in degassed $\text{CH}_3\text{CN}/\text{CHCl}_3$ (8 mL/8 mL). Diethylamine (8 mL) was added slowly and reacted for 7 days under N_2 atmosphere at room temperature. After evaporation of the solvents, the mixture was re-dissolved in distilled water and then dialyzed against DI water for 7 days. Freeze-drying gave **P4** as yellow solid (220 mg, 82%). The M_n and PDI was determined on the precursor **1**. ^1H NMR (300 MHz, CDCl_3) δ = 7.17-7.08 (m, 2 H), 7.03-6.88 (m, 2 H), 4.56-4.32 (m, 2H), 4.18-3.92 (m, 4 H), 3.87-3.38 (m, 56 H), 3.36-3.17 (m, 12 H), 2.91-2.32 (m, 12 H), 2.12-1.74 (m, 4 H), 1.17-0.86 (m, 4 H) ppm. Due to low solubility, ^{13}C NMR spectrum could not be obtained. IR (cm^{-1}): ν 2870, 2817, 2361, 1508, 1489, 1469, 1420, 1380, 1350, 1272, 1200, 1101, 1041, 953, 850, 718. Quantum yield (Φ = 0.21).

[0039] Fig. 3C shows the synthesis of compound **4** and **P9**. Compounds **2** and **3** were synthesized according to the literature: Kim, I.-B.; Phillips, R.; Bunz, U. H. F. Carboxylate Group Side-chain Density Modulates the pH-dependent Optical Properties of PPEs.

Macromolecules **2007**, *40*, 5290-5293 and Bender, M.; Seehafer, K.; Findt, M.; Bunz, U. H. F. Pyridine-based Poly(aryleneethynylene)s: a Study on Anionic Side Chain Density and Their Influence on Optical Properties and Metallochromicity. *RSC Adv.* **2015**, *5*, 96189-96193.

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[0040] Under a nitrogen atmosphere, compound **2** (193 mg, 400 μ mol, 1.0 eq) and compound **3** (356 mg, 400 μ mol, 1.0 eq) were solved in degassed toluene (3.9 mL) and TEA (2.6 mL). Then CuI (4 mg, 20 μ mol, 0.05 eq) and Pd(PPh₃)₂Cl₂ (23 mg, 20 μ mol, 0.05 eq) were added, before the reaction was heated to 60 °C in a closed flask. After stirring for 24 h, the solution was allowed to reach ambient temperature. The gelatinous solution was solved in chloroform and THF (1:1, 50 mL), before it was washed with NH₄Cl_{aq} (50 mL). The two layers were separated, the aqueous layer was extracted with DCM (3 x 50 mL) and the combined organic layers were dried over MgSO₄ and filtered before the solvent was removed under reduced pressure. The resulting residue was dissolved in chloroform (5 mL) and precipitated in pentane (400 mL) and stirred for one hour. The suspension was filtered and the precipitate was dried in vacuum to give compound **4** as a brown solid (348 mg, 72%). The M_n was estimated to be 2.4×10^3 with a PDI of 14. ¹H NMR (600 MHz, CDCl₃): δ = 7.12-7.62 (m, 3 H), 4.97 (br. s, 2 H), 4.50-4.54 (m, 2 H), 4.06-4.30 (m, 8 H), 3.46-3.75 (m, 56 H), 3.29 (br. s, 12 H), 1.18 (br. s, 6 H) ppm. Due to low solubility, ¹³C NMR spectrum could not be obtained. IR (cm⁻¹): ν 2871, 1743, 1684, 1498, 1455, 1398, 1350, 1259, 1193, 1024, 850, 804, 697, 611, 541, 500, 418 cm⁻¹. Compound **4** (148 mg, 122 μ mol, 1.0 eq) was suspended in 2.5 N NaOH (1.5 mL, 50 eq) and refluxed at 50 °C for 24 h. After cooling down to room temperature, the pH-value was adjusted to 7.0 (HCl). The solution was filled into a membrane and was dialyzed

for three days, before the water was removed by freeze-drying to give **P9** as a rubber-like yellow solid (131 mg, 89%). ^1H NMR (600 MHz, D_2O): δ = 8.28-8.37 (m, 1 H), 7.74-7.79 (m, 1 H), 5.04-5.06 (m, 2 H), 3.93-3.96 (m, 4 H), 3.46-3.84 (m, 60 H), 3.25 (br. s, 12 H) ppm. Due to low solubility, ^{13}C NMR spectrum could not be obtained. IR (cm^{-1}): ν 3382, 2872, 2362, 1597, 1499, 1453, 1397, 1198, 1094, 1031, 934, 845, 718, 539, 427, 416 cm^{-1} . Quantum yield (Φ = 0.16).

[0041] The positively charged **P1** and **P3** (not shown), neutral **P4** (not shown) and negatively charged **P5** (not shown) and four fluorophore-quencher type complexes (**C1-2**, **C1-7** (not shown), **C1-8** (not shown) and **C1-9**(not shown)) were screened at different pH values. It was found that a sensor array comprising a cationic poly(*para*-phenyleneethynylene) (PPE) **P1** and its electrostatic complex **C** (**C1-2**) with the weakly fluorescent high-density charged **P2** at pH10 and pH13 works well to discriminate between the different ones of the NSAIDs (Figure 2b).

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[0042] The 11 NSAIDs of Fig. 1 show varied responses towards this sensor array, as can be seen in Figure 4. Processing these data by linear discriminant analysis (LDA, sextuplet data sets), one discriminates all of the NSAIDs according to their Mahalanobis distances, employing two dimensionless factors. The LDA converts the training matrix (4 factors x 11 NSAIDs x 6 replicates) into canonical scores. The first two canonical factors shown in Fig. 4B represent 80% of the total variation. The canonical scores are clustered into eleven different groups. The jack-knifed classification matrix with cross-validation reveals 100% accuracy and the sensor system successfully discriminates the different NSAIDs.

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[0043] Figure 4A shows a fluorescence response pattern ($I - I_0 / I_0$) obtained by **P1** (500 nM, at pH10 and 13, buffered) and its complex **C** (**P1-P2** at 500 nM-250 nM, at pH10 and 13, buffered) treated with NSAIDs **D1-D11** (6 mM). Each value is the average of six independent measurements; each error bar shows the standard deviation of these measurements. Fig. 4B shows a 2D canonical score plot for the first two factors of simplified fluorescence response patterns obtained with the sensor array comprising **P1**, **C** (each at pH10 and 13, buffered) with 95% confidence ellipses. Each point represents the response pattern for a single analgesic to the array. The canonical scores are clustered into eleven different groups. The jack-knifed classification matrix with cross-validation reveals 100% accuracy.

[0044] To validate its efficiency, tests were performed with randomly chosen ones of the NSAID samples of the training set. The new cases are classified into groups, generated through the training set, based on their shortest Mahalanobis distance to the respective group. All of the 44 tested unknown NSAID samples were correctly identified using the sensor array and the training set. In the 2D LDA plot (Figure 4B), results from eleven NSAIDs clustered independently in accordance to their structural similarity. Super groups form, i.e. all salicylates cluster differently than the profens, the fenamic acids and the arylacetic acids.

[0045] A Concentration Dependent Discrimination of 'Fenamic Acid' – NSAID D4 was now investigated.

[0046] The fluorescence modulation data for the NSAID **D4** was recorded at concentrations from 0 to 1.8 mM. The LDA converts the training matrix (4 factors x **D4**, nine concentrations x 6 replicates) into nine canonical scores. The first two canonical

factors represent 94% of the total variation. The jack-knifed classification matrix with cross-validation reveals 100% accuracy. Eight different concentrations (without control, 0 mM) of the NSAID **D4** from the training set were randomly chosen for blind testing. The new cases are classified into groups, generated through the training matrix, based on their shortest Mahalanobis distance to the respective group. Among 32 unknown concentration samples, all were classified correctly.

[0047] The concentration is linearly mapped in the LDA plot, with the zero-point in the upper right-hand corner (Figure 5A). The same experiment was performed with the NSAIDs **D7** and **D9**, and here also the concentration is linearly correlated with the response. This suggests that for every NSAID we have a slice of exclusion where one can identify NSAIDs at unknown concentrations without interference from other NSAIDs. There is a corollary to this: if two or more NSAIDs are on the same vector connecting to the origin, then their concentration dependent profiles cannot be discerned. However, in the other cases one should be able to obtain both structure and concentration from an unknown sample, even though at low concentrations this would become increasingly difficult. Figure 5B depicts the concentration dependent data in the context of all of the other NSAIDs. The cases that cannot be discerned when different concentrations are allowed are the NSAIDs **D6**, **D7** or **D1**, **D2**, **D8**, **D10**, **D11**. In an ideal case, the concentration dependent slope would be significantly different for each and any NSAID.

[0048] **Table 2** Detailed Information of the ten Over-the-Counter (OTC) NSAIDs used in this study

Abbr	Brand name (Company)	Main/total (mg) ^a	Side ingredients
ASS1	ASS-Ratiopharm® (Ratiopharm)	500/620	corn starch, cellulose powder
ASS2	Aspirin® (Bayer)	500/670	Na ₂ CO ₃ , highly dispersed SiO ₂ , carnauba wax, hydroxypropylmethylcellulose (HPMC), Zn-stearate
ASS3	ASS 500mg HEXAL® (Hexal AG)	500/620	micro-crystalline cellulose, corn starch
ASS4	ASS 500-1A Pharma® (1A Pharma)	500/620	micro-crystalline cellulose, corn starch
ASS5	ASS STADA® (STADA pharm)	500/650	micro-crystalline cellulose, corn starch
IBU1	Ibuflam® akut (Winthrop)	400/590	microcrystalline cellulose, corn starch, lactose monohydrate, E468, highly dispersed SiO ₂ , Mg-stearate, polyvinylalcohol, Macrogel 3350, talcum powder
IBU2	IbuHEXAL® akut (Hexal AG)	400/480	microcrystalline cellulose, E468, HPMC, Macrogel 400, Mg-stearate, highly dispersed SiO ₂ , talcum powder, TiO ₂
IBU3	Ibu 400 akut-1A Pharma® (1A Pharma)	400/480	microcrystalline cellulose, E468, HPMC, Macrogel 400, Mg-stearate, highly dispersed SiO ₂ , talcum powder, TiO ₂
IBU4	Ibuprofen AL 400 (ALIUD PHARMA)	400/680	Mg-stearate, corn starch, Macrogel 400, 6000, carboxymethyl starch sodium, HPMC
IBU5	Dolormin® (McNeil)	400/820	Microcrystalline celluloses, povidon, Mg-stearate, TiO ₂ , hydroxypropyl cellulose, HPMC

^a The weight of the main ingredient and the total weight of each tablet.

- 5 [0049] Sensing of OTC Samples (Aspirin and Ibuprofen) was now carried out. The test was the identification and discrimination between different, commercially available NSAIDs. Different fillers, super-disintegrants etc. are present in varying concentrations. Five commercially available samples of aspirin and five samples of ibuprofen were

selected. Table 2 shows the composition and the weight of all of the ingredients according to the package insert. Figures 6A and 6B show the fluorescence responses of the different ibuprofen and aspirin samples. For aspirin, the sample **ASS2** is the least fitting in this series, probably due to the presence of colored carnauba wax, (yellow/brown) in the sample. The other ASS-samples cluster closely. In the case of the ibuprofens, the two samples **IBU2, 3** cluster and are away from the data point for the NSAID **D8**. **IBU2, 3** contain titanium dioxide, another ingredient that will interfere with the fluorescence modulation of the chemical tongue by ibuprofen. The other **IBU** samples cluster more closely. The super cluster of the **IBUs** does not overlap with the super cluster of the ASS-species.

[0050] Figure 6A shows the fluorescence response pattern ($I - I_0 / I_0$) obtained by **P1** (500 nM, at pH10 and 13, buffered) and complex **C1-2** (**P1-P2** at 500 nM-250 nM, at pH10 and 13, buffered) treated with NSAIDs **D2** (aspirin, 6 mM, control) and **D8** (ibuprofen, 6 mM, control) and commercial available OTC tablet aspirin (**ASS1-ASS5**, 6 mM), ibuprofen (**IBU1-IBU5**, 6 mM). Fig. 6B shows a 2D canonical score plot for the first two factors of simplified fluorescence response patterns obtained with an array of **P1, C1-2** (each at pH10 and 13, buffered) with 95% confidence ellipses. Each point represents the response pattern for a single analgesic to the sensor array.

[0051] Once we co-process the data employed for Figure 6B with all of the data obtained for the other NSAIDs, we find that the **IBU** and the **ASS** samples form super clusters that do not overlap with any of the other NSAIDs (here shown in grey, Figure 7). The response to the sensor field in the sensor array, while modulated by the additives and formulations, is fundamentally determined by the active drug component. The selected sensor field - in combination with LDA - easily handles these discriminative tasks.

[0052] Figure 7 shows in 7A a 2D canonical score plot and in 7B a 3D canonical score plot obtained with an array of **P1**, **C1-2** (each at pH10 and 13, buffered) treated with NSAIDs **D1-D11** and commercial available OTC tablet aspirin (**ASS1-ASS5**), ibuprofen (**IBU1-IBU5**). Each point represents the response pattern for a single analgesic to the array. The
5 grey/black colours represent pure analgesics, the colorful shape represent the OTC aspirin and ibuprofen.

[0053] The four-element sensor array comprising a highly fluorescent cationic PPE and its complex with a weakly fluorescent anionic PAE is merely exemplary. In this example,
10 both highly fluorescent cationic PPE and its complex with a weakly fluorescent anionic PAE (at pH10 and pH13) discern 11 different NSAIDs, even at different concentrations. The sensor array is able to identify and discriminate commercial NSAIDs (over-the-counter ibuprofen and aspirin). The different ibuprofens and aspirins cluster together. It is still possible to identify a tablet from a specific drug maker. This successful discrimination
15 demonstrates the power of these sensor arrays composed of weakly selective elements.

[0054] The sensor arrays work by a combination of hydrophobic and electrostatic interaction of the analytes with the conjugated polymer(s) or with their formed complex(es). These hydrophobic and electrostatic interactions are magnified due to
20 fluorescence based detection. The excited state of the fluorophores is far more responsive towards external stimuli than the ground state.

[0055] The array sensor described in this document works well and surpasses in its flexibility and discriminatory power prior art specific sensors. Such specific sensors often

do not exist (at any rate) for discrimination of even fairly simple or complex analytes we are interested in.^{15,25} This suggests that the sensor array described in this document has an enormous potential fundamentally but also for application, particularly if transparent and easily applicable rules are developed that connect analyte class to an appropriate fluorophore and quencher type.

[0056] It is also suggested that the sensor array of this document enables discrimination of fake and/or adulterated drug formulations.

[0057] In a second aspect of the concept, a sensor array for the detection of fruit juices was investigated.

[0058] **Sample Preparation.** 14 apple juices (**AJ1-AJ14**), 5 black currant juices (**BJ1-BJ5**) and 6 red grape juices (**GJ1-GJ6** (detailed information see Table 2) were purchased from local supermarkets and used directly in our discrimination experiments with **P1** and **P2** (Figure 8B). The pH values were measured immediately after opening with a pH-meter. Chemicals, solvents and buffers (pH 3, citric acid/NaOH/NaCl; pH 7, $\text{KH}_2\text{PO}_4/\text{Na}_2\text{HPO}_4$; pH 13, glycine/NaOH/NaCl) were purchased from commercial laboratory suppliers. Reagents were used without further purification unless otherwise noted.

[0059] **Table 2.** Detailed Information of the Investigated Juices (14 Apple Juices **AJ1-AJ14**, 5 Black Currant Juices **BJ1-BJ5** and 6 Red Grape Juices **GJ1-GJ6**)

Abbr.	Commercial Juice Name	pH ^a	Conc.	Fat/Fatty acids ^b	Carbohydrate ^s /Sugar ^b	Proteins ^b	Salts ^b
AJ1	^{Bio} Apple Juice	3.47	100%	<0.5g /0.5g	11.0g/10.0g	<0.5g	<0.01g
AJ2	Apple Juice	3.40	100%	0.1g/0.02g	11.0g/10.5g	0.1g	0.005g
AJ3	Riod'oro Apple Juice	3.49	100%	<0.1g/0.1g	10.3g/9.9g	0.1g	<0.01g
AJ4	Riod'oro Premium Apple Juice	3.41	100%	0g/0g	11.0g/11.0g	0g	0g
AJ5	REWE Apple Juice	3.50	100%	0g/0g	11.2g/10.7g	0g	0g
AJ6	Albi Apple Juice	3.60	100%	<0.5g/<0.1g	11.0g/10.0g	<0.5g	<0.01g
AJ7	^{Bio} Solevita Bio Apple Juice	3.56	100%	0.1g/<0.1g	11.0g/10.5g	0.1g	<0.01g
AJ8	VITAFIT Apple Juice	3.60	100%	0.1g/0.02g	10.5g/10.0g	0.1g	<0.01g
AJ9	VITAFIT Premium Apple Juice	3.63	100%	0.1g/0.02g	11.0g/10.5g	0.1g	<0.01g
AJ10	Amecke Apple Juice	3.65	100%	0.1g/<0.1g	11.1g/10.6g	0.5g	0.01g
AJ11	Ja Apple Juice	3.56	100%	0g/0g	10.2g/9.8g	0g	0.01g
AJ12	EDEKA Apple Juice	3.73	100%	0.1g/0.02g	10.5g/10.0g	0.1g	0.008g
AJ13	Lift Apple spritzer	3.53	55%	0g/0g	6.0g/5.8g	0g	0g
AJ14 ^c	Hessischer Apple Wine	3.76	5.5% Alcohol	-	-	-	-
BJ1	^{Bio} Cassis Black Currant juice	3.10	30%	0g/0g	82g/82g	0g	0g
BJ2	^{Bio} Nektar Black Currant juice	3.60	25%	0.1g/0.02g	13g/13g	0.1g	0.001g
BJ3	Heimische Black Currant juice	3.60	25%	<0.5g/<0.1g	12g/12g	0.1g	<0.01g
BJ4	REWE Black Currant juice	3.54	25%	0g/0g	12.9g/12.9g	0.3g	0.01g
BJ5	Jacoby Black Currant juice	3.57	25%	<0.5g/<0.1g	8.4g/8.4g	<0.5g	<0.01g
GJ1	^{Bio} Grape juice	4.06	100%	0.01g/0.002g	17g/17g	0.2g	0.003g
GJ2	^{Bio} REWE Red Grape juice	4.07	100%	0g/0g	16.6g/16.6g	0g	0g
GJ3	REWE Grape juice	3.92	100%	0g/0g	16.9g/16.9g	0g	0g
GJ4	Riod'oro Premium Grape juice	3.68	100%	0g/0g	16.6g/16.6g	0g	0g
GJ5	Jacoby Grape juice	3.77	100%	<0.5g/<0.1g	16g/16g	<0.5g	<0.01g
GJ6	REWE Merlot Grape juice	3.63	100%	0g/0g	17g/17g	0.3g	0.01g

^a Measured immediately after opening. ^b Contents per 100ml, information obtained from the label. ^c Apple Wine. ^{Bio} with BIO label of the Europe union.

[0060] **Preparation of red and green grape juice:** Seedless green grapes, Sugraone, Spain, 500g, and red grapes Summer Royal, Italy, 500g, were purchased from local supermarkets. Grapes were removed from their stems and washed with cold water, drained off and mashed with a potato masher. The resulting grape sludge was centrifuged with an ultracentrifuge Beckman L7-55, 20000 rpm, 0.5 h, 20 °C to isolate clear grape juice as supernatant.

[0061] **Black currant juice:** black currants, Germany, 500g, were purchased from local supermarkets. The black currants were washed and de-stemmed. 250 mL of water were added, the mixture was mashed with a potato masher and heated for 10 min to gentle boil to furnish 550 mL of a thick solution. Ultracentrifugation (20000 rpm, 0.5 h, 20 °C) furnished a clear dark black currant juice, which was diluted to 40% of its original concentration by distilled water.

[0062] **Fluorescence response patterns.** Emission spectra were recorded and analyzed on a CLARIOstar (firmware version 1.13) Platereader (BMG Labtech, built in software, version 5.20 R5). Data were analyzed by CLARIOstar MARS Data Analysis Software (version 3.10 R5) from BMG Labtech. The specific response for each analyte was measured six times, the peak values acquired. These were used as the observables for the subsequent linear discriminant analysis (LDA).

[0063] **LDA.** The acquired data were evaluated by LDA in SYSTAT (version 13.0). In LDA, all variables were used in the model (complete mode). The tolerance was set as

0.001. The fluorescence response patterns were transformed to canonical patterns. The Mahalanobis distances of each individual pattern to the centroid of each group in a multidimensional space were calculated and the assignment of the case was based on the shortest Mahalanobis distance.

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[0064] The structure of P1 and P2 is shown in Fig. 8B and their preparation described in ref.22. Fluorescence response pattern and linear discriminant analysis was carried out on the two polymers P1 and P2 to identify their suitability for use in the sensor array. Preliminary screening was done recording fluorescence patterns with different PAEs in
10 presence of the analyte. Using the response patterns with the highest distinction yielded a workable tongue showing six elements, consisting of P1 and P2 at different pH values (pH 3, pH 7 and pH 13). P1 is anionic (Figure 8B) while P2 is positively charged; both are highly fluorescent in water (Figure 8A). Figure 8A shows the results of a systematic evaluation and selection of the successful tongue elements of the sensor array for the juice
15 sensing and Figure 8B shows the chemical structures and quantum yields (ϕ) of **P1** and **P2**.

[0065] Table 2 shows the different apple, grape and black currant juices in this study. Juices, complex mixtures of different compounds, the number of which probably ranges in the hundreds, are 8-17% aqueous solutions of sugar at a pH between pH 3.1- 4.1. Their low
20 pH prevents fast microbial spoiling. As a first juice experiment, all of the juices were exposed towards PPE **P1** and **P2**.

[0066] Figures 9A and 9B show the quenching results of the PPEs when the juices are added at different pH values. Figures 9Aa and 9Ba show fluorescence response patterns (I

- I_0 / I_0) obtained by **P1** (2 μ M, at pH3, pH7 and 13, buffered) and by **P2** (2 μ M, at pH3, pH7 and pH13, buffered) treated with commercial apple juice (1), black currant juice (2) and red grape juice (3) samples (50 μ L per 300 μ L for **P1** and 1 μ L per 300 μ L for **P2**). Each value is the average of six independent measurements; each error bar shows the standard deviation of these measurements. Figures 9A and 9B show the canonical score plots for the first two factors of simplified fluorescence response patterns obtained with an array of **P1** and **P2** with 95% confidence ellipses. Each point represents the response pattern for a single juice sample to the array.

[0067] Fig. 10A shows a combined 2D canonical score plot obtained with an array of **P1** (2 μ M, at pH3, 7, 13, buffered) treated with apple, black currant and red grape juices (50 μ L). Fig 10B shows a combined 2D canonical score plot obtained with an array of **P2** (2 μ M, at pH3, 7, 13, buffered) under the same conditions using 1 μ L of juice. Fig. 10C shows a combined 2D LDA plot for the first two factors of simplified fluorescence response patterns from six sensing elements obtained from **P1** (pH 3, pH 7 and pH 13) and **P2** (pH3, 7, 13, buffered) using the same selection of 25 juices.

[0068] The fluorescence quenching of the cationic polymer **P2** is much more effectively quenched (1 μ L analyte vs. 50 μ L analyte per 300 μ L buffer/PPE solution) than that of the anionic **P2**. This behavior suggests that electrostatic effects play a role in the discrimination of the fruit juices. The major fluorescence quenching “interactome” of the fruit juices with the PPEs is negatively charged, allowing a strong interaction with the positively charged PPE **P2**. All of the fruit juices are discriminated either by **P1** or by **P2**, when working at the three different pH values used in the example. Discrimination is

possible when inspecting the raw data but it is much better visualized after linear discriminant analysis (LDA) of the data. Figure 10 combines the response results from all juices after LDA. Both **P1** as well as **P2** discriminate all of the fruit juices. **P2** does a better job at the discrimination, as all of the red grape juice and the black currant juices are discriminated. For unknowns, **P2** is not perfect for apple juice, while **P1** is not optimal for grape juice. The black currant juices are discriminated by both **P1** and **P2**. LDA of the combination of data extracted from **P1** and **P2** (Figure 10C, totally six sensing elements), results in improved discrimination. The jackknifed classification matrix with cross-validation reveals a 100% accuracy, the randomly chosen 100 unknown juice samples using combined six elements were calculated with the training matrix. The accuracy increased to 100%. A more detailed fingerprint is conferred on each juice with the increase of sensing elements (Table 3).

[0069] It is seen that the sensor array is more discriminating for single juice elements, the inter group differentiation between apple juice and red grape juice is less pronounced than for **P2** alone (Table 3).

[0070] **Table 3.** Jackknifed Classification Matrix Obtained From LDA **P1** and **P2** at Three Different pH-Values.^a

Sensing elements		P1 (pH3, pH7 and pH13)			P2 (pH3, pH7 and pH13)			Combined
Juice types		AJ	BJ	GJ	AJ	BJ	GJ	All types
Jackknifed classification matrix	Number of samples	84	30	36	84	30	36	150
	Correctly classified	83	30	35	84	30	36	150
	Accuracy (%)	98.8	100	97.2	100	100	100	100
Blind test	Unknown samples	56	20	24	56	20	24	100
	Correctly identified	56	20	22	54	20	24	100
	Accuracy (%)	100	100	92	96	100	100	100

- 5 [0071] An important question arises, if some of the claimed grape juices are not pure grape juices. They might be mixtures of red grape juice with blackcurrant juice. Would it be possible to distinguish such mixtures of red grape juice with blackcurrant juice? Admixing black currant juice deepens the colour of red grape juice if that is desired. To test this hypothesis, the sensor array comprising the **P2** tongue at pH 3, 7 and 13 was selected under
- 10 standard conditions (1µL juice /300µL matrix). We added black currant juice **B4** or **B5** to either **G6** or **G1**. If one does this, **B4** can substitute up to 50% of **G6** or **G1** and the mixture is still identified as red grape juice. The alternative does not work, i.e. if one adds grape juice towards black currant juice, **P2** indicates leaving the area that is assigned by LDA to the black currant juice. To obtain more insight we tested fruit juices we prepared in our
- 15 laboratory from commercially available green and red grapes, and black currants.

[0072] Figure 11 shows the fluorescence response of the self-made juices (blackcurrant, green grapes, red grapes) and mixtures of red and green grape juices. After LDA from the data obtained for the self-prepared juices, it is found that the admixing of the red and green grape juices is an additive process with respect to their properties expressed by LDA. The hot extracted blackcurrant juice does not group with the commercial blackcurrant juices, suggesting that commercial black currant juice is processed differently. The main discriminating factor in Figure 11 (x-axis, Factor 1) expresses color and the quenching ability of the juices. The green grape juice, the least coloured juice is placed on the left-hand side, while blackcurrant juice samples are placed on the right hand side. The red grape juices locate in the middle. The same applies for Figure 10, where the response of all of the fruit juices are displayed. The x-axis approximates the color depth of the juices, just mirror-symmetrical from the ordering seen in Figure 11. The y-axis is currently not ascribed to a simple physicochemical property and can neither be correlated with sugar content nor with acidity. It must represent a complex property or properties; it could be a combination of fruit acids (mandelic acid, citric acid, tartaric acid etc.) and / or sugar plus other complex coloured species present in these fruit juices.

[0073] It is therefore found that a single positively charged, water soluble conjugated polymer, **P2**, discriminates apple juices, black currant juices and grape juices. It was established that red grape juice can be mixed with black currant juice into a zone where the LDA-processed responses of a significant number of commercially available (pure) grape juices are located. The result poses several questions. a) Some of the commercial red grape juices might contain small to moderate amounts of black currant juice or b) the variation of the response of grape juices is -due to the multiple dozens of different grape varieties- to be

expected, or c) our tongue is not sufficiently developed to discriminate mixtures, or all of the above.

[0074] The minimalist nature of the sensor array is surprising, as the discriminative power of **P2** is brought out by its employ at different pH-values, i.e. only change of the sensing conditions. This one polymer acts therefore as an efficient three-element-tongue, where the change of the analytes with the pH-value must significantly contribute towards the successful recognition strategy. Why are **P1** and **P2** successful in discriminating complex analytes such as fruit juices? **P1** and **P2** display a fairly rigid backbone, and - depending upon their conformation - could either be viewed as a “sticky” molecular board (phenyl rings parallel to each other) or a “sticky” molecular rod (phenyl rings twisted with respect to each other). The stickiness or non-specific affinity towards arbitrary analytes comes from hydrophobic interactions, hydrogen bonding, and electrostatic interactions. All of these interactions must be promiscuous and non-specific, as our sticky boards/rods have no inbuilt shape recognition elements and neither do they show great variations in their chemical structure, not even upon protonation. These results shed a different light on both the lock-and-key principle discussed in the introduction, but also on the professed need to employ chemically different tongue elements (Suslick) to reach recognition. Neither of these constraints are active in our boards or rods, just the presence of a molecular surface with varying “stickiness” or non-specific affinity for interactions with complex analytes.

[0075] Sticky linear molecular surfaces such as in our PPEs are powerful as they allow the sensing and the discrimination of almost all and any conceivable analytes because of the complete lack of shape requirements for either analytes or tongue elements.

[0076] If one looks into the identification of counterfeit products, drugs, or consumer goods, the absence of a clearly identifiable signal molecule means that counterfeit and adulterated products are more easily recognized as the signal generation and identification process is complex and unknown to both the counterfeiter but also the legal producer of the analyzed product, making potential protection stronger. The results show a minimalist chemical tongue made from **P2** for use in the sensor array that is able to discriminate fruit juices at different pH-values without any problem.

[0077] The sensor array of the document has been further used to identify different types of teas. 8 black teas, 6 green teas and 8 oolong teas were investigated as shown in the table below.

[0078] Table 4 Detailed information on the investigated teas (8 black teas B1–B8, 6 green teas G1–G6 and 8 oolong teas O1–O8) used in this study.

Name abbreviation	Category	Fermentation degree	brand	Geographical origin
B1 ^a	Black tea	Fermented	Teekanne	ASSAM
B2	Black tea	Fermented	Teekanne	ASSAM
B3	Black tea	Fermented	Teekanne	Ostfriesen
B4	Black tea	Fermented	Teekanne	Darjeeling
B5	Black tea	Fermented	Meßmer	Darjeeling
B6	Black tea	Fermented	Tee Gschwendner	Nepal
B7	Black tea	Fermented	Tee Gschwendner	Nordindien
B8	Black tea	Fermented	Tee Gschwendner	Darjeeling
G1 ^b	Green tea	Non-fermented	Longjing	Hangzhou Xihu
G2 ^b	Green tea	Non-fermented	Longjing	Hangzhou Xihu
G3	Green tea	Non-fermented	Teekanne	China
G4	Green tea	Non-fermented	Meßmer	China
G5	Green tea	Non-fermented	Linglong	Hunan Guidong
G6	Green tea	Non-fermented	Biluochun	Jiangsu Dongting
O1 ^b	Oolong tea	Semi-fermented	Tieguanyin	Fujian Anxi
O2 ^b	Oolong tea	Semi-fermented	Tieguanyin	Fujian Anxi
O3	Oolong tea	Semi-fermented	Huangguanyin	Fujian Wuyishan
O4	Oolong tea	Semi-fermented	Tieluohan	Fujian Wuyishan
O5 ^b	Oolong tea	Semi-fermented	Rougui	Fujian Wuyishan
O6 ^b	Oolong tea	Semi-	Rougui	Fujian Wuyishan

		fermented		
O7	Oolong tea	Semi-fermented	Shuixian	Fujian Wuyishan
O8 ^b	Oolong tea	Semi-fermented	Rougui	Fujian Wuyishan

^a Earl grey. ^b Tea samples (G1, G2; O1, O2 and O5, O6, O8) were obtained from different manufacturers in China.

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[0079] Figure 12 shows the identification of different black teas (Fig. 12a), green teas (Fig. 12b), and oolong teas (Fig. 12c) using a 6-element tongue of PPEs complexed with curcubit[8]uril (C8). In this case, C8 forms a complex with two PPE-molecules and this complex is reduced in its fluorescence. The addition of C8 makes the system more sensitive and increases the discriminative power of the PPE-tongue. The structure of C8 is shown in Fig. 13.

[0080] It was that, after exposure to the six element tongue, in which we combined three different PPEs shown in Fig. 13 with curcubit[8]uril (C8) at two different pH values (pH 3 and pH13), it was possible to obtain a data set which upon linear discriminant analysis discriminated all of the different teas according to character, type and brand. We could (see Table 5) identify 84% of all tested unknown samples of black teas and 100% of green and oolong teas.

Table 5. Jackknifed classification matrix and unknown sample identification obtained from LDA

Tea samples		black tea	green tea	oolong tea
Jackknifed classification matrix	Number of samples	48	36	48
	Correctly classified	45	36	48
	Accuracy (%)	94	100	100
Unknown samples identification	Number of samples	32	24	32
	Correctly classified	27	24	32
	Accuracy (%)	84	100	100

- 5 [0081] A further application is the discrimination of red wines which can be analyzed through a sensor array that is very similar to that employed for the identification of the white wines. Fourteen different red wines were tested and discriminated using the tongue already employed for the white wine discrimination, as shown in the table 6 below. First experiments also suggest that we can discriminate Amarone wines from Ripasso wines and
- 10 Barolo types using the PPEs shown in Fig 14

Ref.	Vintage	Varietal	%	Brand	Sugar -29-	Origin Sonnenberg Forlmann	Acids (g/l)	Resid. sugar (g/l)	Grape 2 (%)	vol. (%)
Blau	2013	<u>Blaufränkisch</u> (<u>Lemberger</u>)	100	Weingut Heinz Pfaffmann Lemberger feinherb		Germany	6.1	15.5		11.5
Bordeaux	2014	<u>Bordeaux</u> grape 1: <u>Merlot</u>	80	Chai de Bordes Rouge - Cheval Quancard Bordeaux		France			<u>Cabernet</u> <u>Sauvignon</u>	13
CaSa	2015	<u>Cabernet</u> <u>Sauvignon</u>	100	Grand Verdier Cabernet Sauvignon		France				13
Dorn1	2015	<u>Dornfelder</u>	100	Adam Müller - Dornfelder QbA	Dry	Germany	5.2	2.8		12.5

[illegible]

Samtrot	2014	<u>Samtrot</u> (mutation of <u>Pinot Meunier</u> / <u>Schwarzriesling</u>)	10	Lauffener Katzenbeißer Samtrot Kabinett	Germany	4.3	16.1			10.5
Syrah	2014	<u>Syrah (Shiraz)</u>	10 0	Kangaroo Ridge - Shiraz	Australia					13.5
Zinfandel del	2015	<u>Zinfandel</u> (<u>Primitivo</u>)	10 0	Farnese Farneto Primitivo Puglia	Italy					13.5

[0082] In a further aspect, the method and sensor array can be used to verify the authenticity of products. This involves extracting a small portion of the product and dissolving this small portion in, for example, water or alcohol to form a solution. The solution will be applied to the sensor array and the fluorescence response measured. The fluorescence response can be compared against the expected fluorescence response for genuine materials.

[0083] This method enables a manufacturer of a product to send, for example, a vial of a reference solution of the product to a testing laboratory so that the testing laboratory has a standard against which products can be tested. In a further aspect, the reference solution is dried on a substrate before being sent from the manufacturer to the testing laboratory. The dry product can then be made up again into a reference solution at the testing laboratory

[0084] In a further aspect, the method and sensor array can be used as a marker for genuine articles. The solution is placed on part of the article at a known position and allowed to dry. The position is known and the testing laboratory can at a later stage remove part of the dried solution for testing. Examples of marked products include, but are not limited to, high value watches.

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Claims

1. A method for identification of an analyte comprising:
preparing a plurality of solutions at a plurality of pH values of at least one
5 fluorescent poly(*para*-phenyleneethynylene) and its complex;
exposing the complex analyte to the plurality of the solutions;
measuring the fluorescence intensity of the exposed complex analyte;
comparing the fluorescence intensity with a library; and
identifying the complex analyte from the comparison.
10
2. The method of claim 1, wherein the analyte is at least one of a fruit juice or an active pharmaceutical preparation.
3. The method of claim 1, wherein the poly(*para*-phenyleneethynylene) is selected
15 from one of the poly(*para*-phenyleneethynylene)s shown in Fig. 2, 3 or Fig. 8.
4. A sensor array for the identification of an analyte comprising:
a plurality of wells having solutions at a plurality of pH values of at least one
fluorescent poly(*para*-phenyleneethynylene) and its complex;
20 an excitation light source;
a fluorescent light detector; and
a storage device for recording a plurality of fluorescent light patterns.
5. The sensor array of claim 4, further comprising a processor adapted to accept
25 measured fluorescent light intensity from the fluorescent light detector and compare the measured fluorescent light intensity with the plurality of stored fluorescent light patterns.
6. The sensor array of claim 4 or claim 5, wherein the poly(*para*-
30 phenyleneethynylene) is selected from one of the poly(*para*-phenyleneethynylene) shown in Fig. 2, 3 or Fig. 8.

Fig. 1

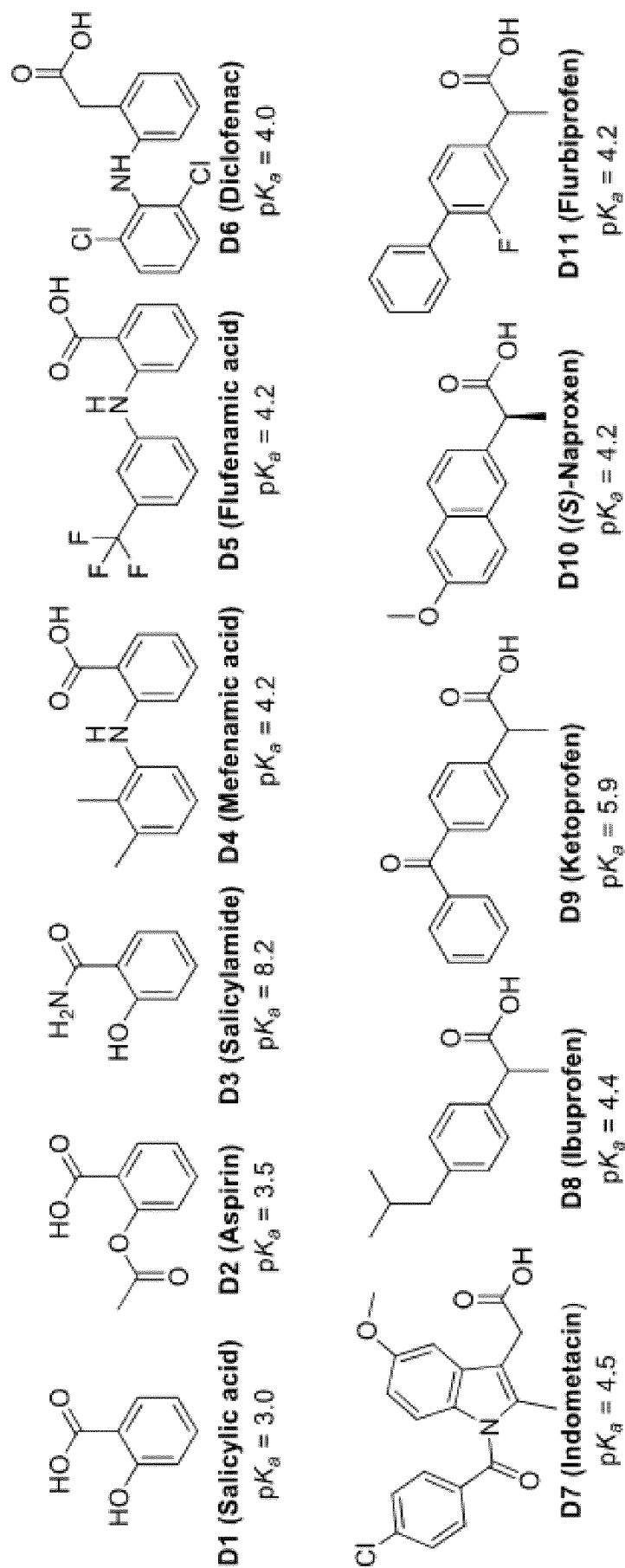
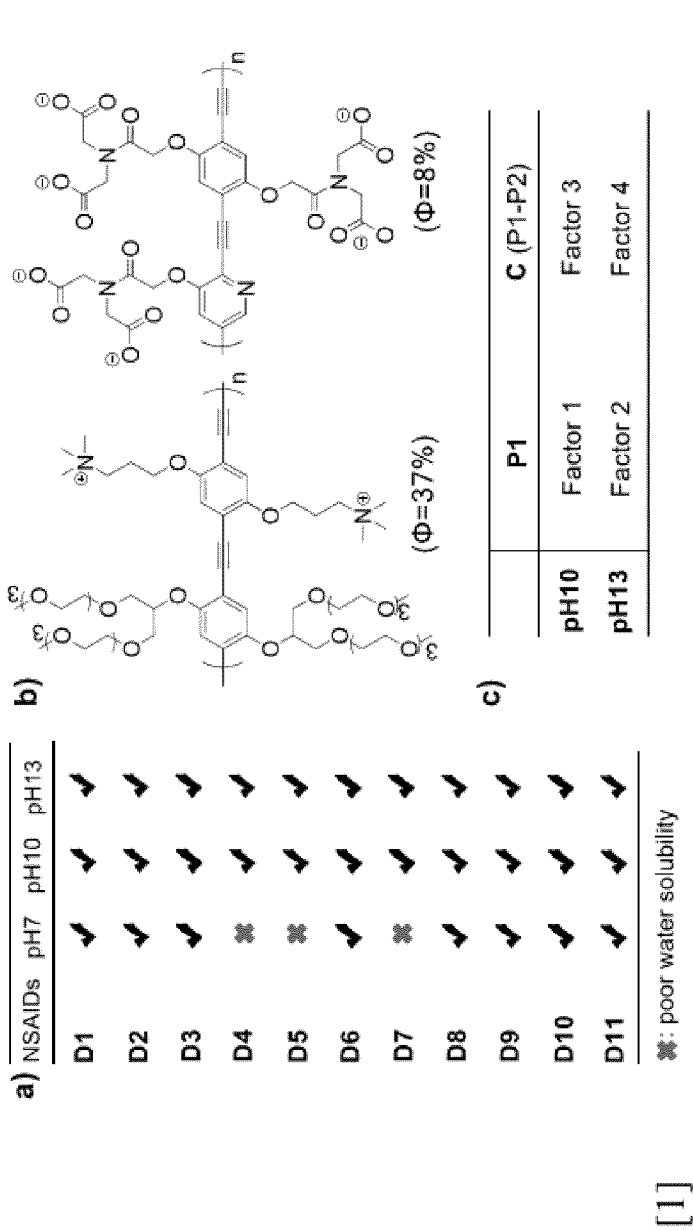


Fig. 2



[1]

a) Water solubility of NSAIDs **D1-D11** (6 mM) at different pH values. **b)** Structures of positively charged **P1** and negatively charged **P2**, used for analgesics sensing (ϕ = quantum yield). **c)** Final selected four sensing factors by using single **P1** and its electrostatic complex **C (P1 + P2)**.

Fig. 3A

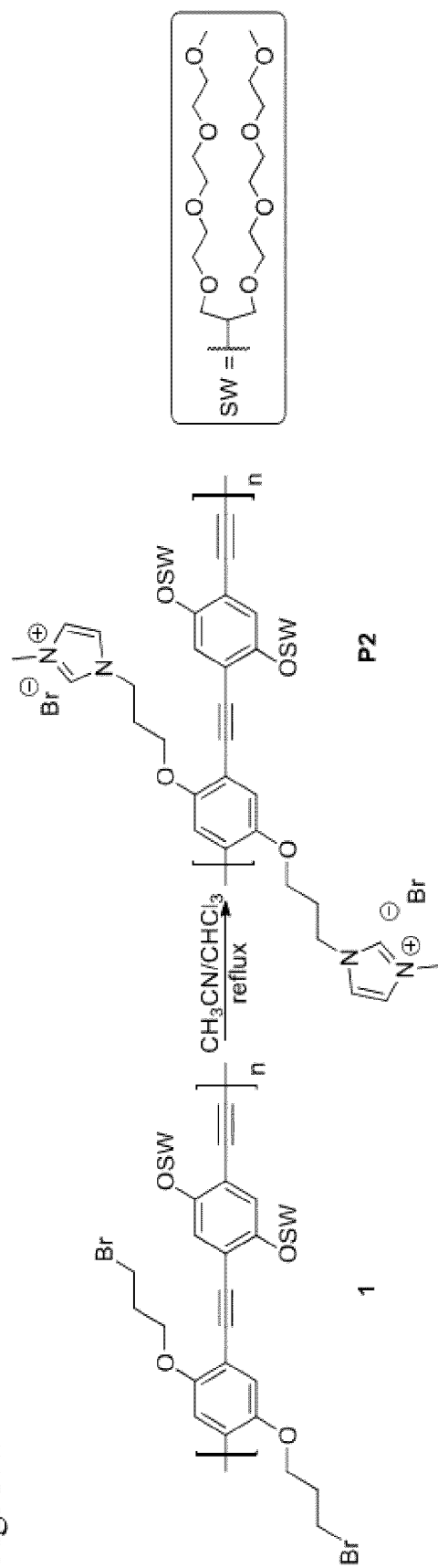


Fig. 3B

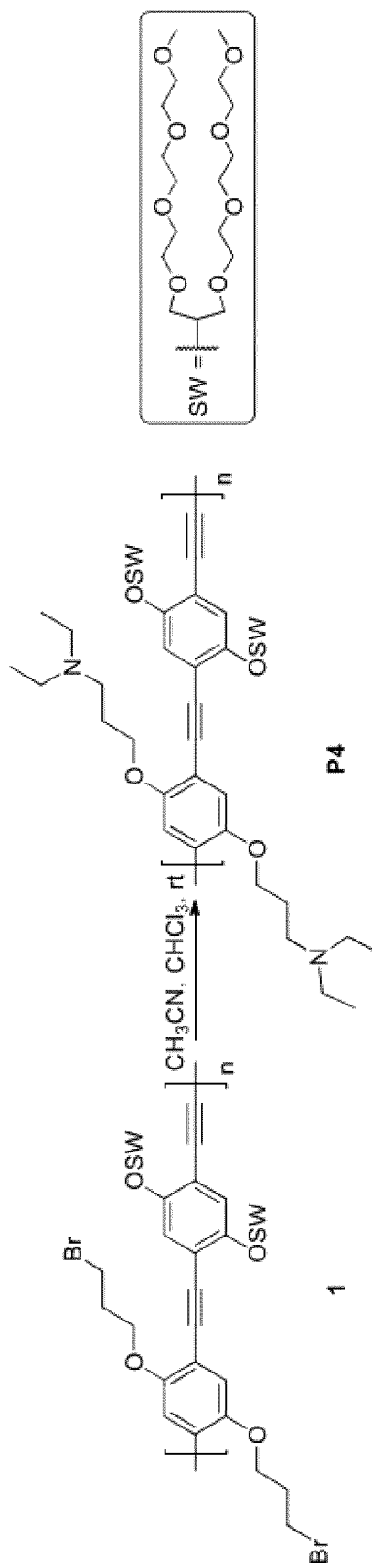


Fig. 3C

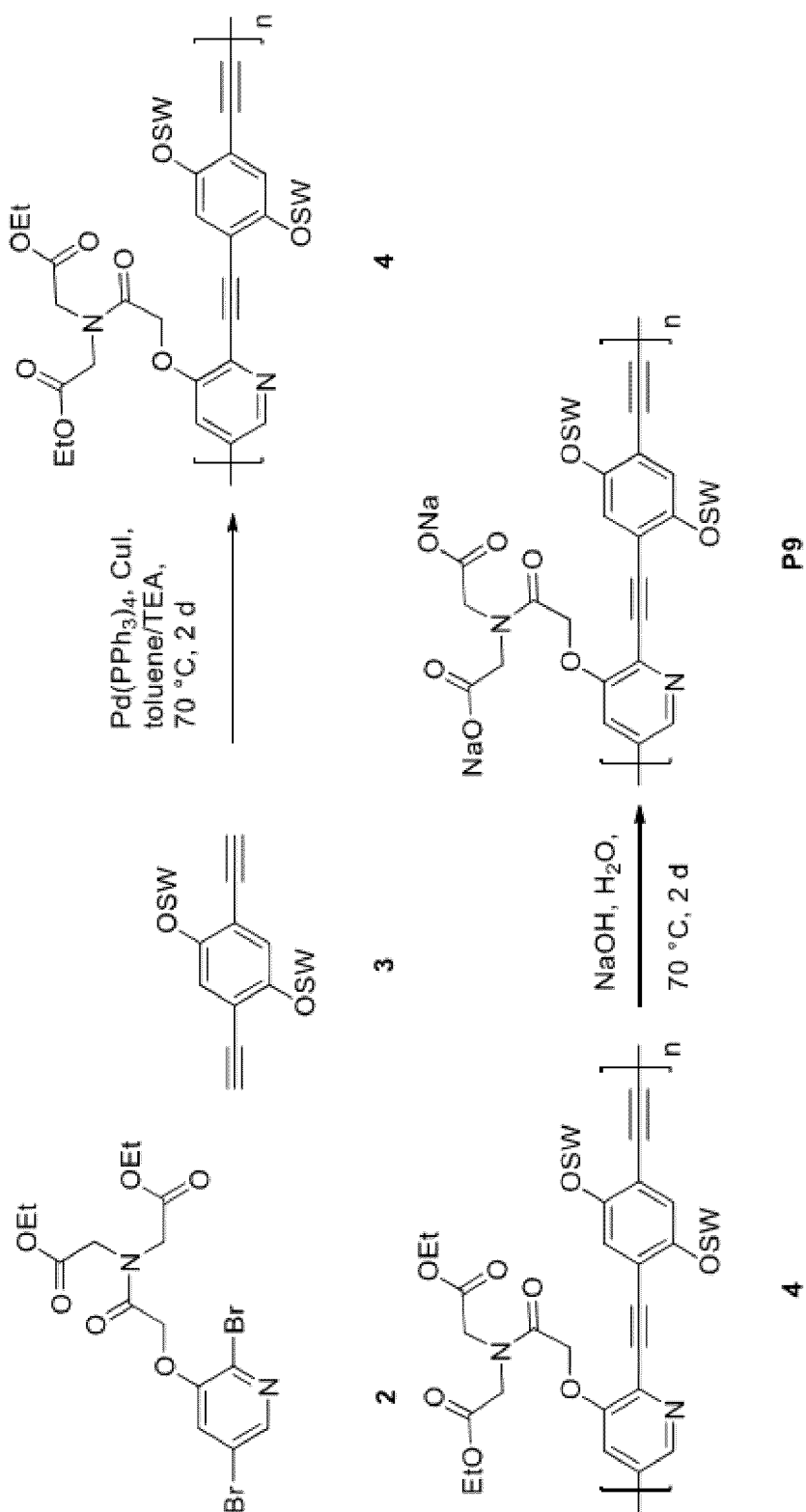
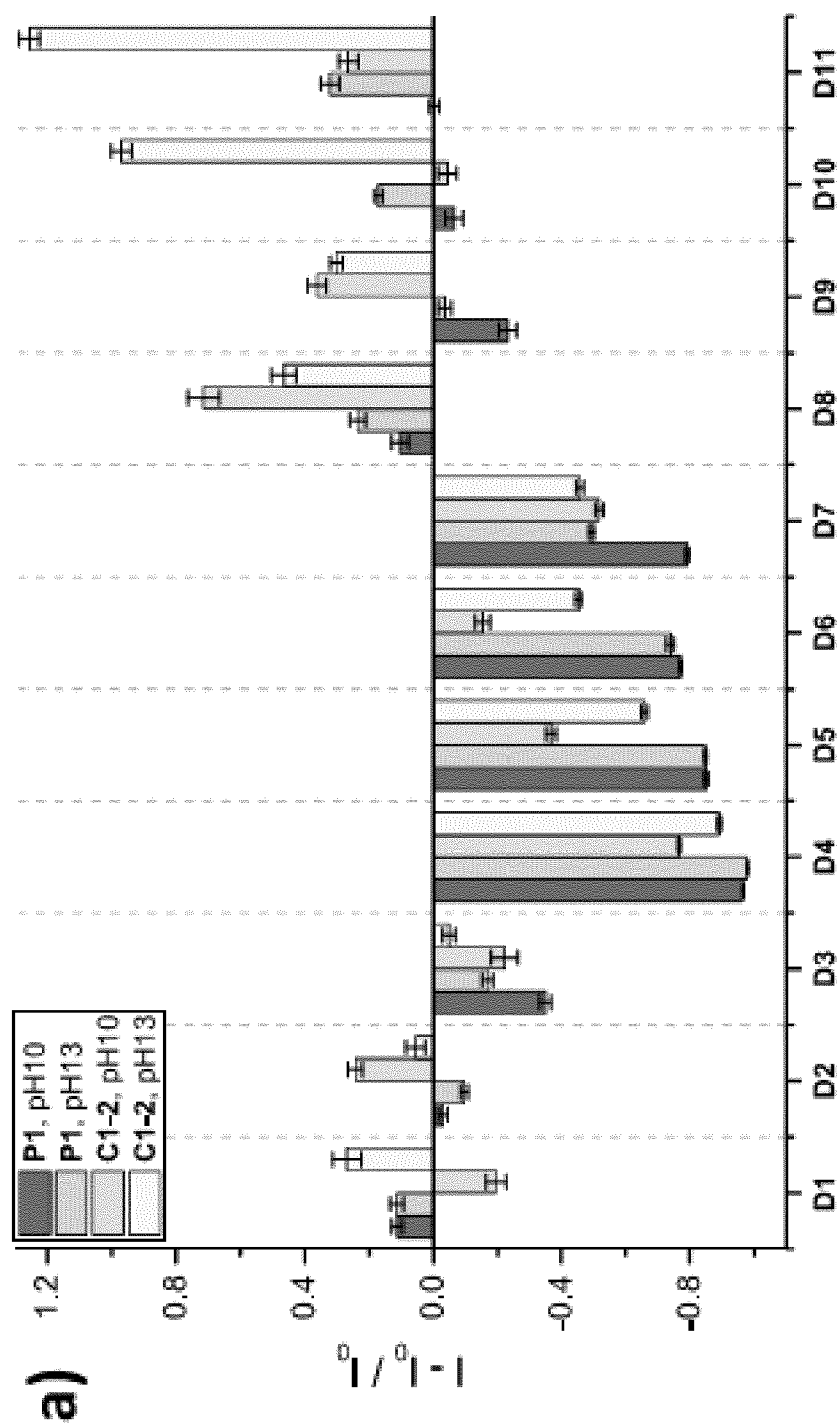


Fig. 4A



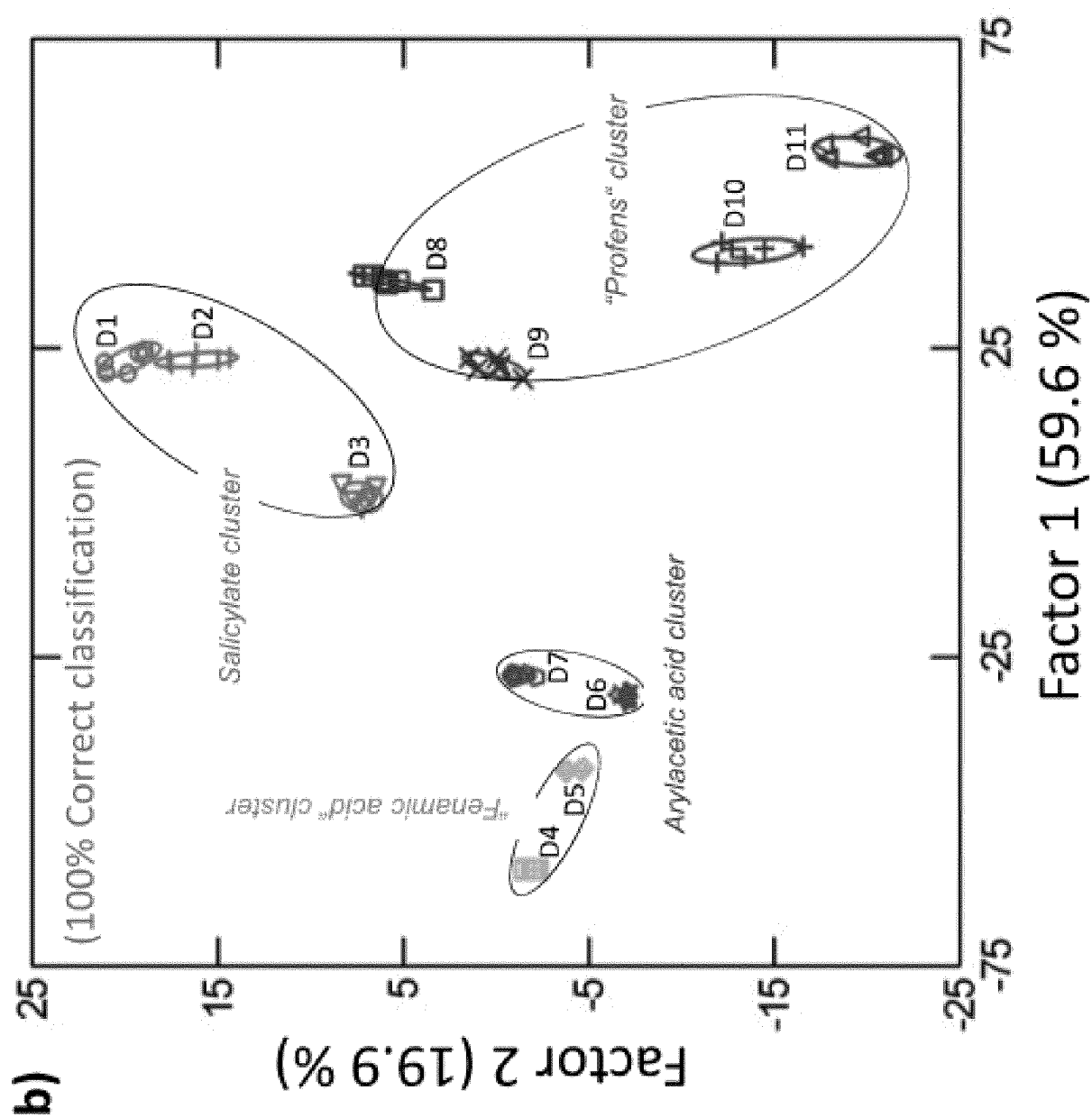


Fig 5A

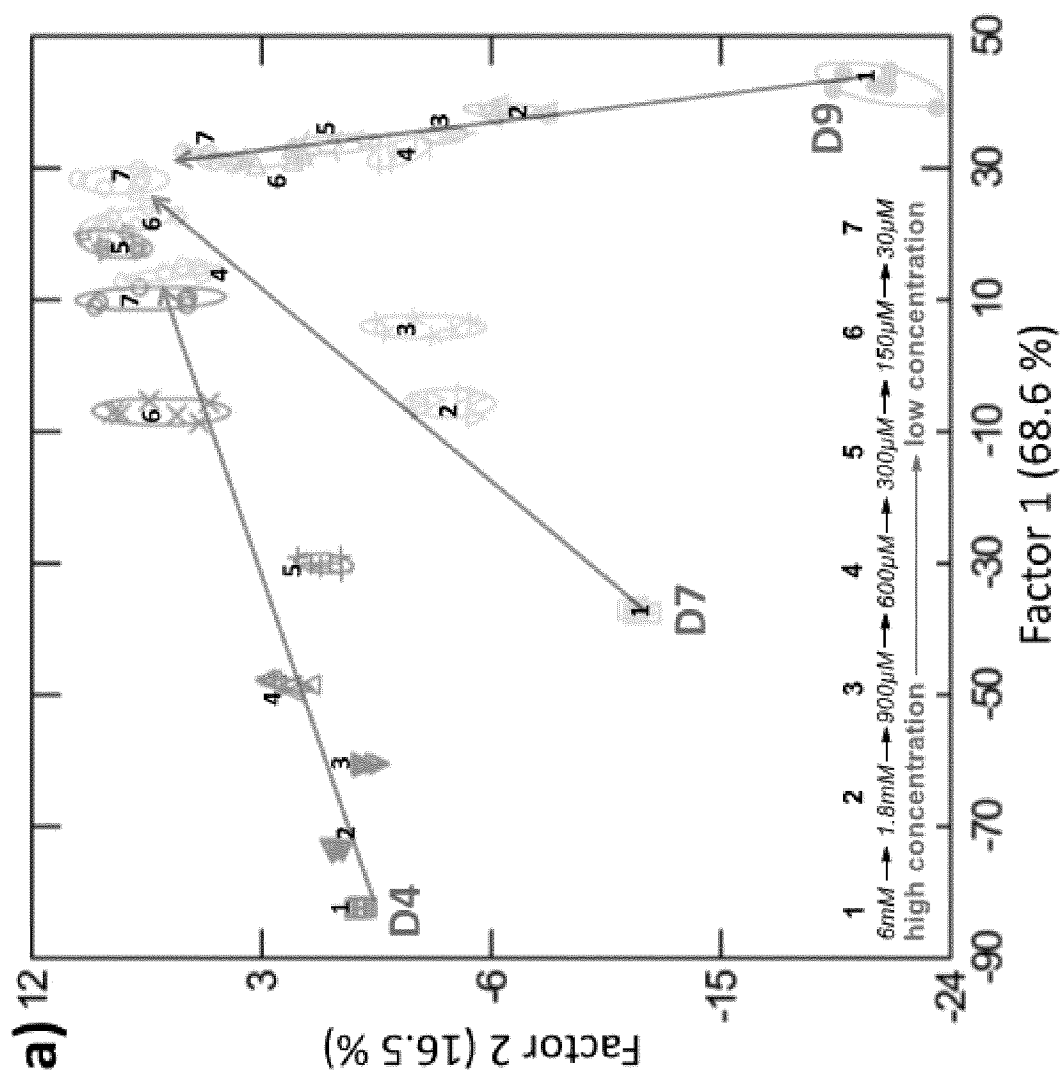


Fig 5B

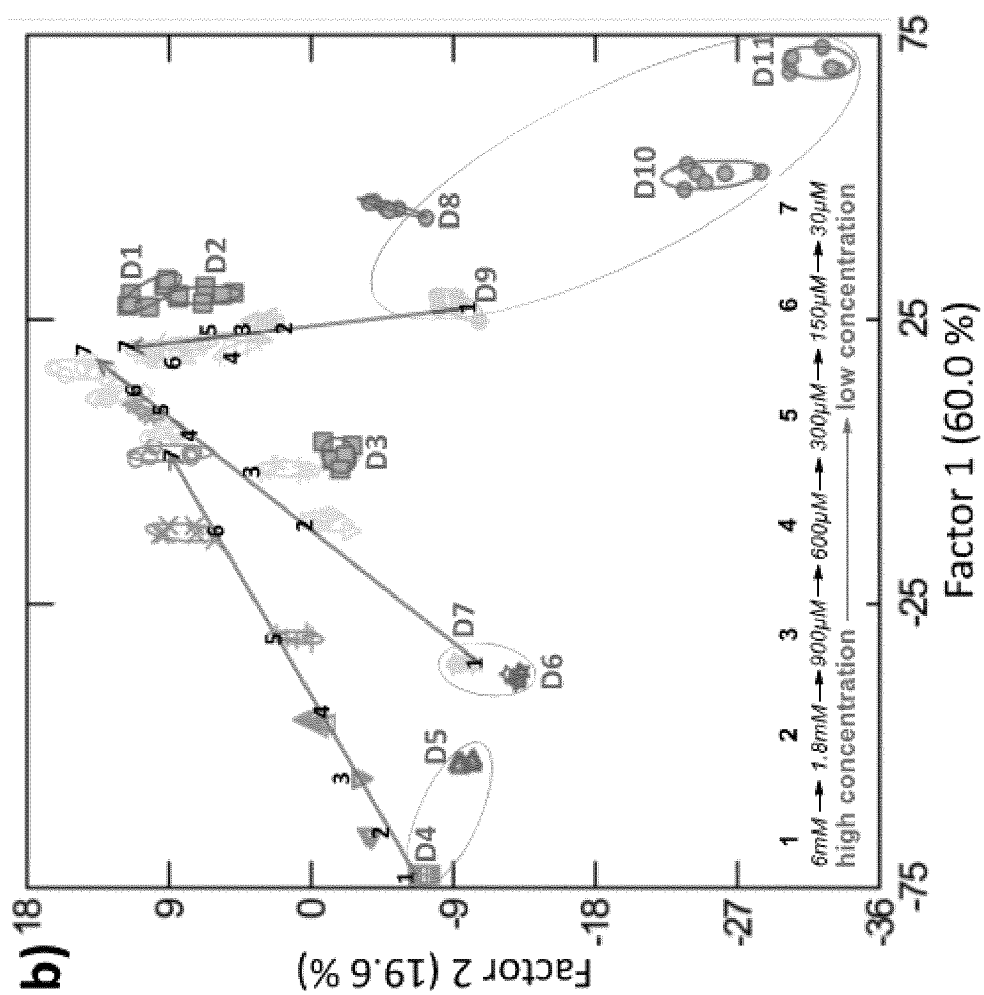


Fig 6A

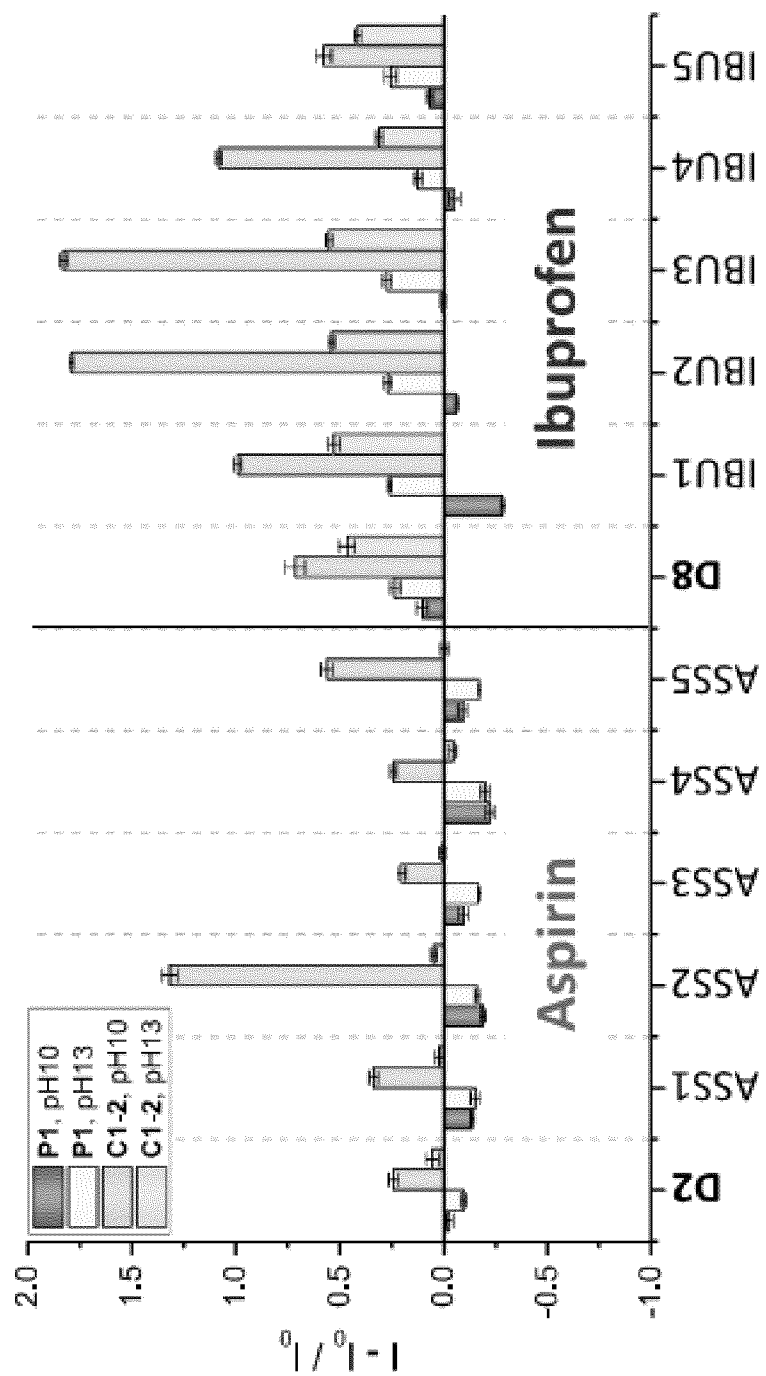


Fig 6B

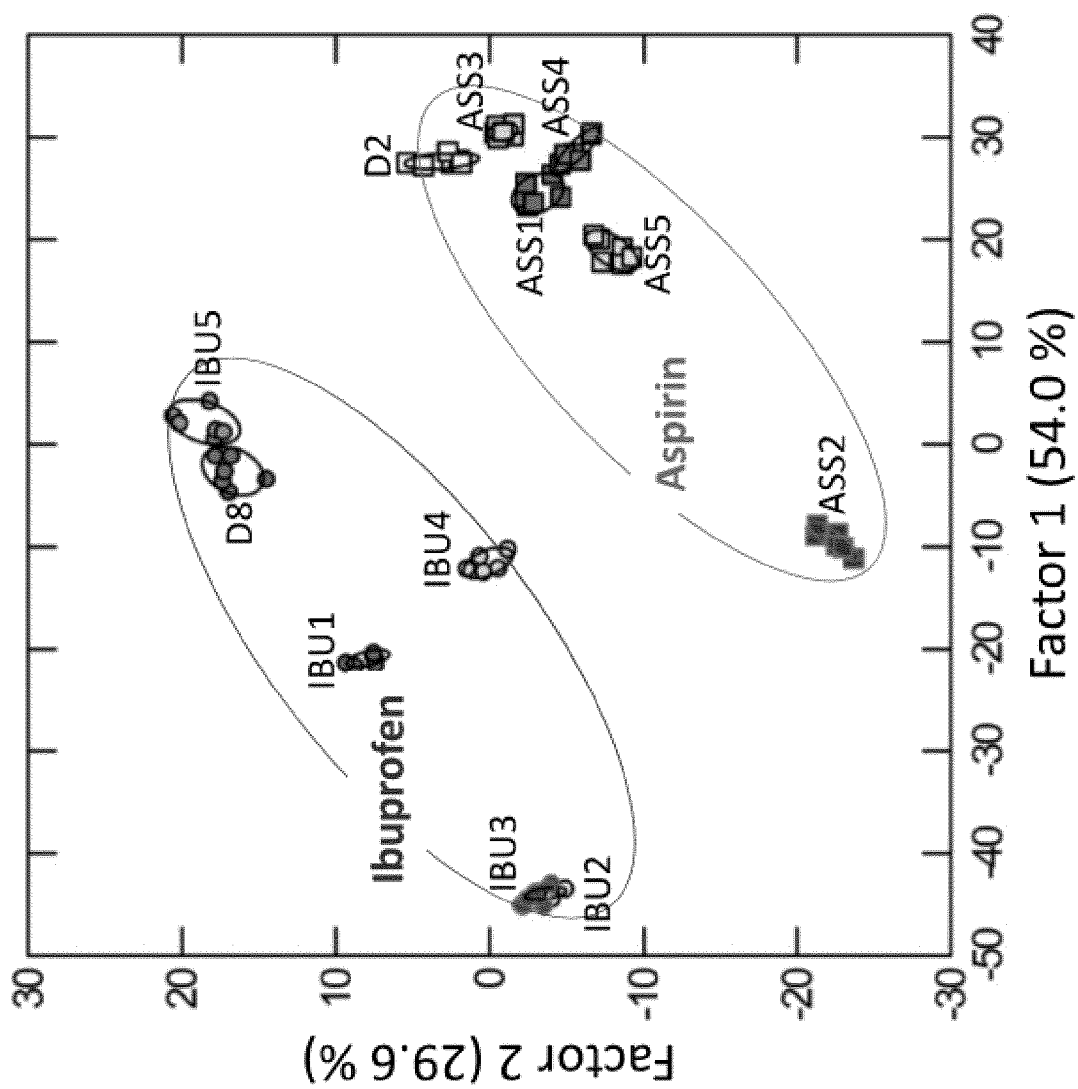


Fig 7A

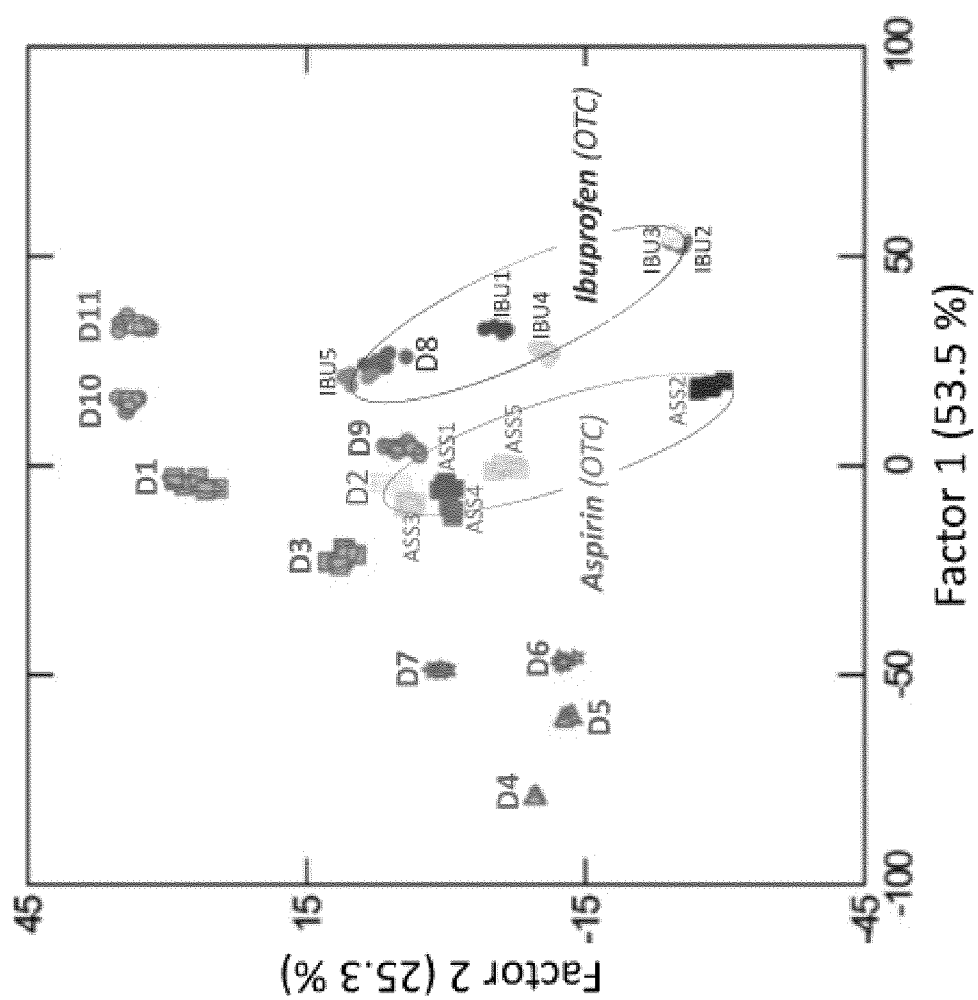
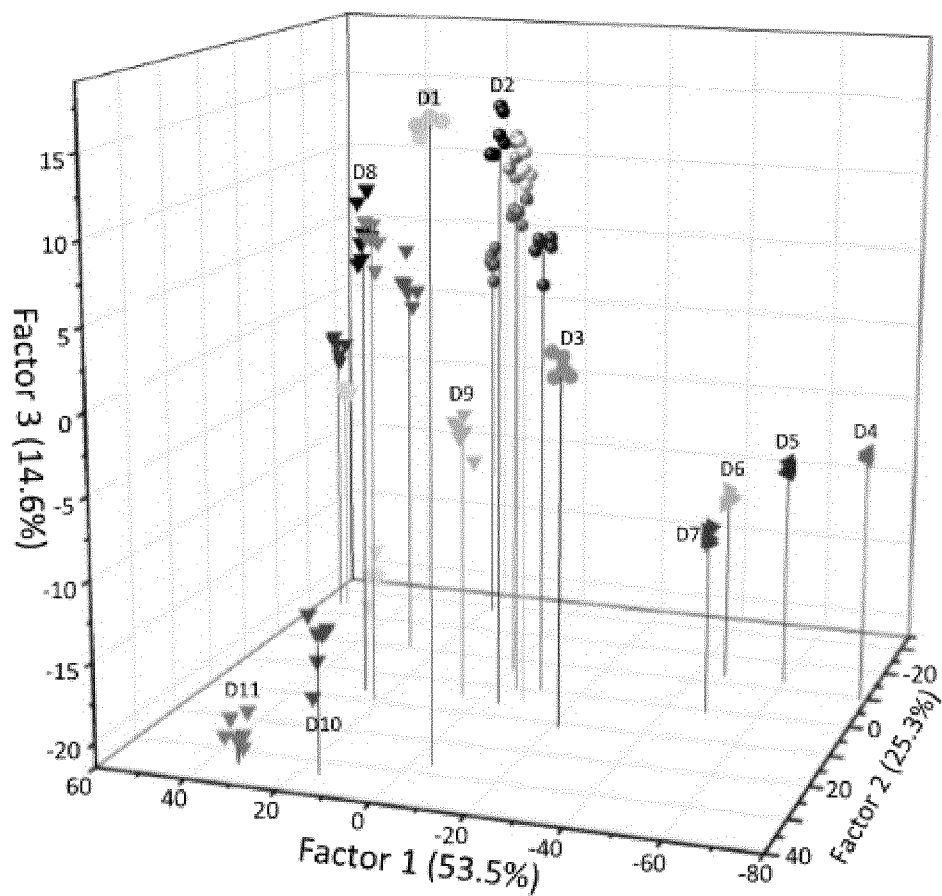


Fig 7B



<i>Ibuprofen (OTC)</i>	<i>Aspirin (OTC)</i>	<i>NSAIDs samples</i>	
▽ IBU1	● ASS1	● D1	▶ D6
▽ IBU2	● ASS2	● D2	▶ D7
▼ IBU3	○ ASS3	● D3	▼ D8
▼ IBU4	● ASS4	◀ D4	▼ D9
▼ IBU5	● ASS5	◀ D5	▼ D10
			▼ D11

Figs 8A and 8B

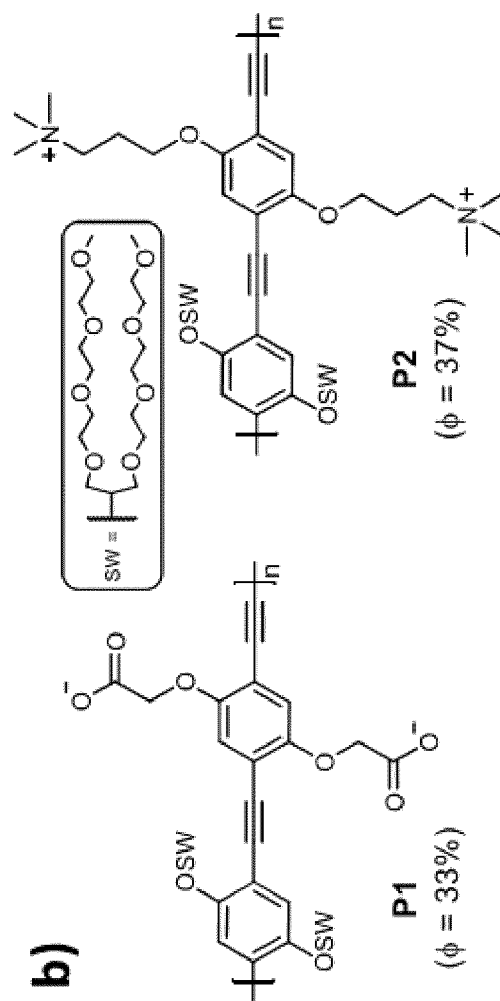
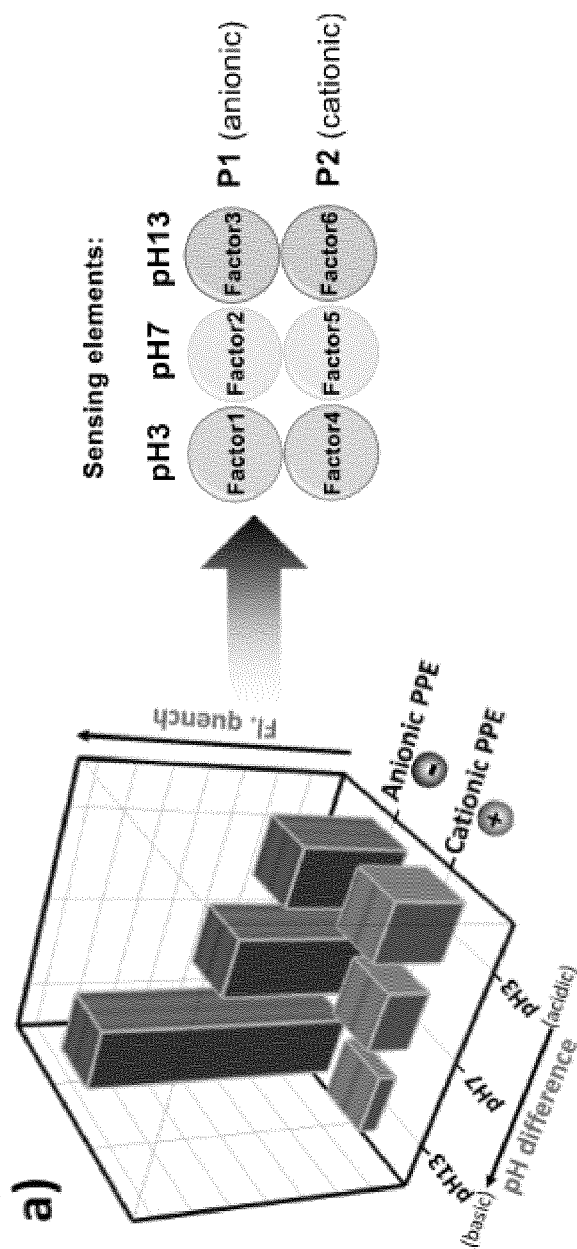


Fig 9A

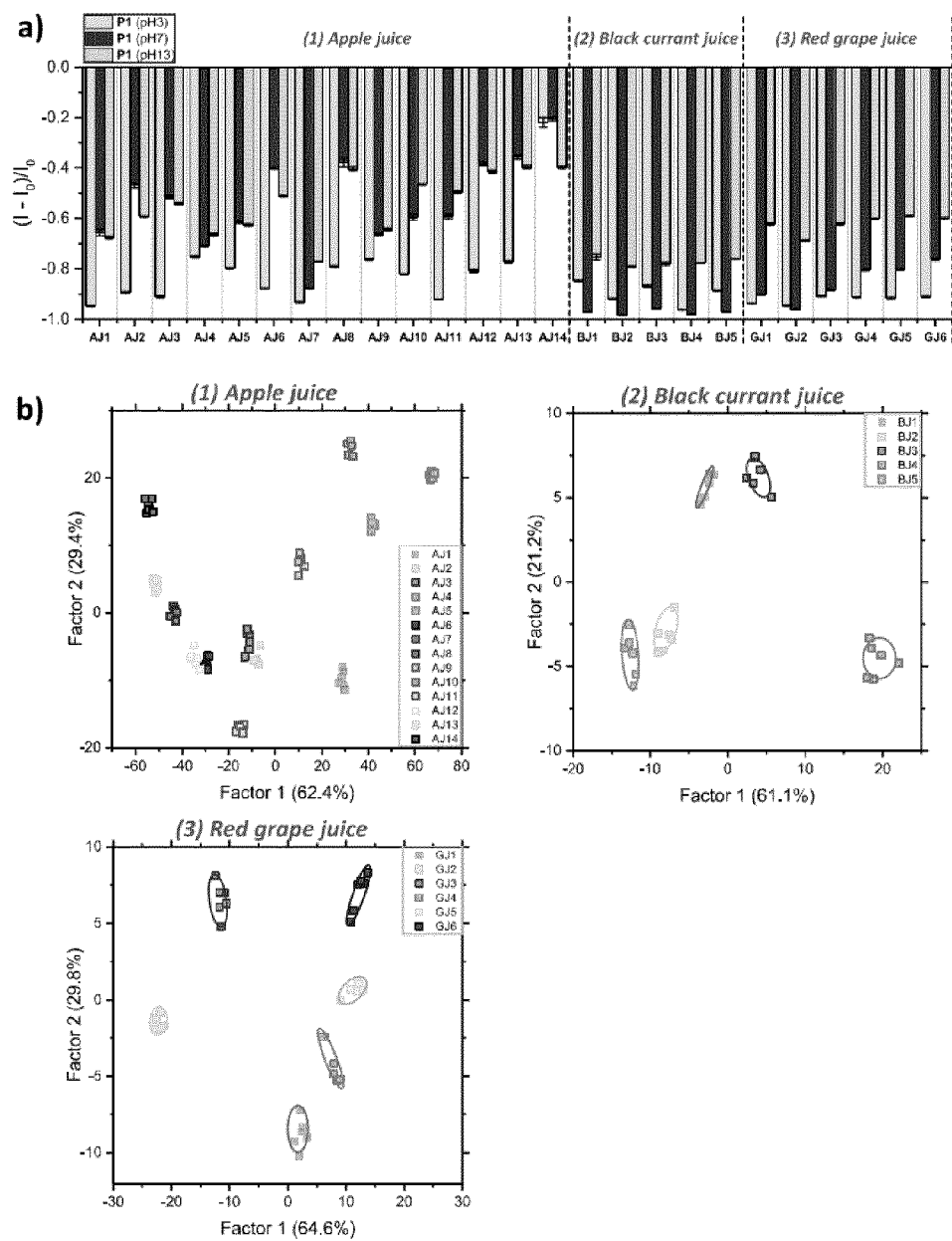
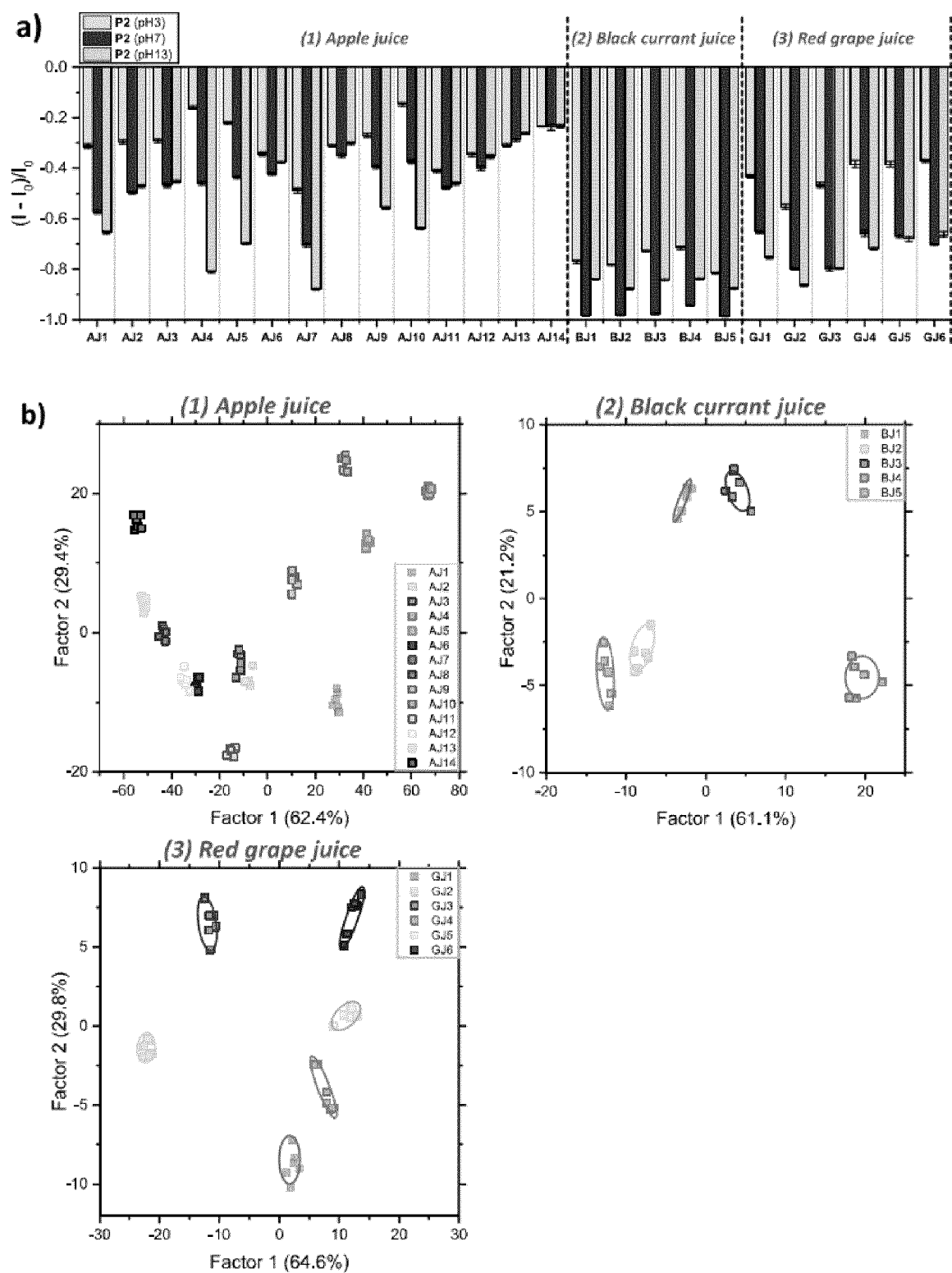
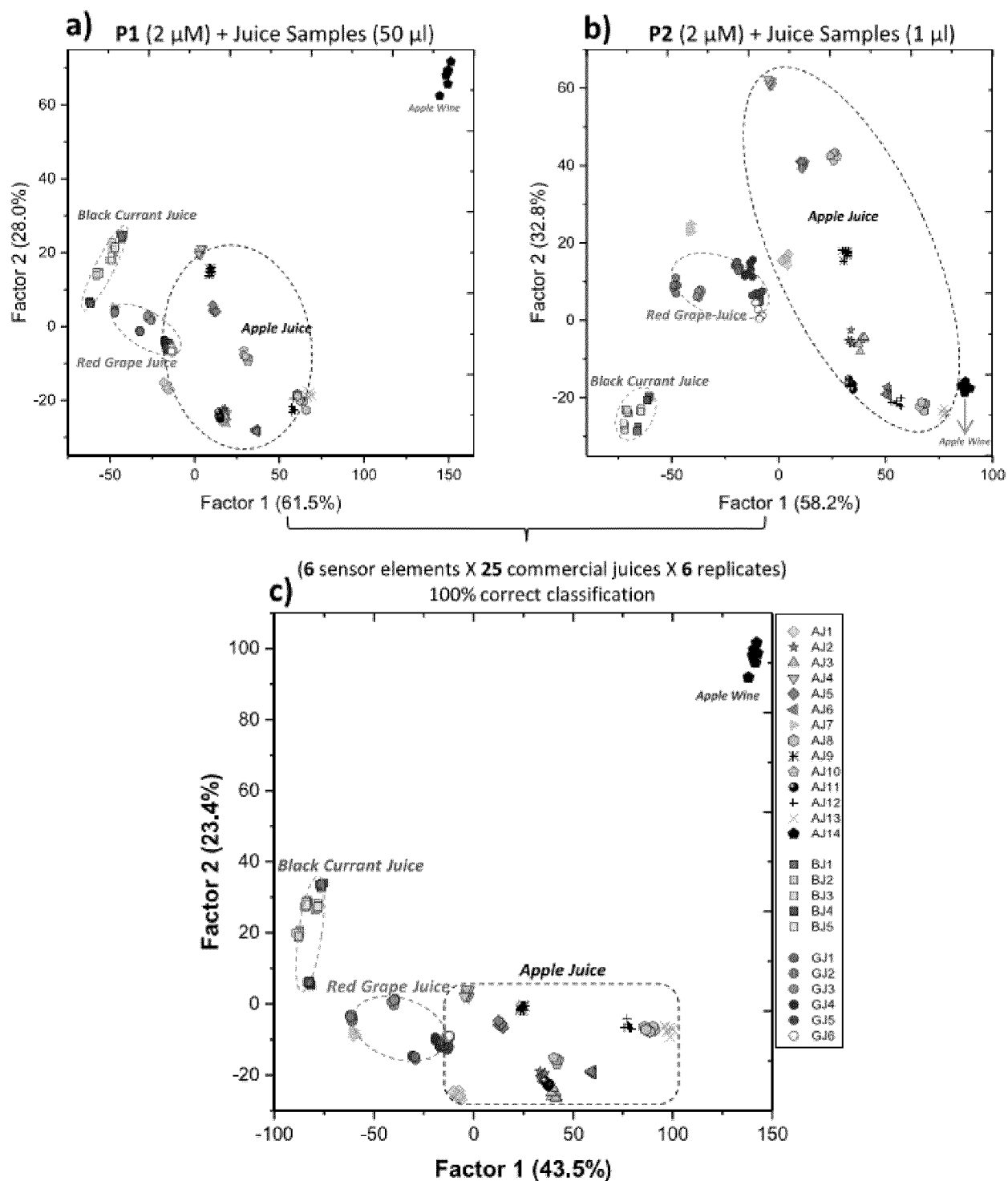


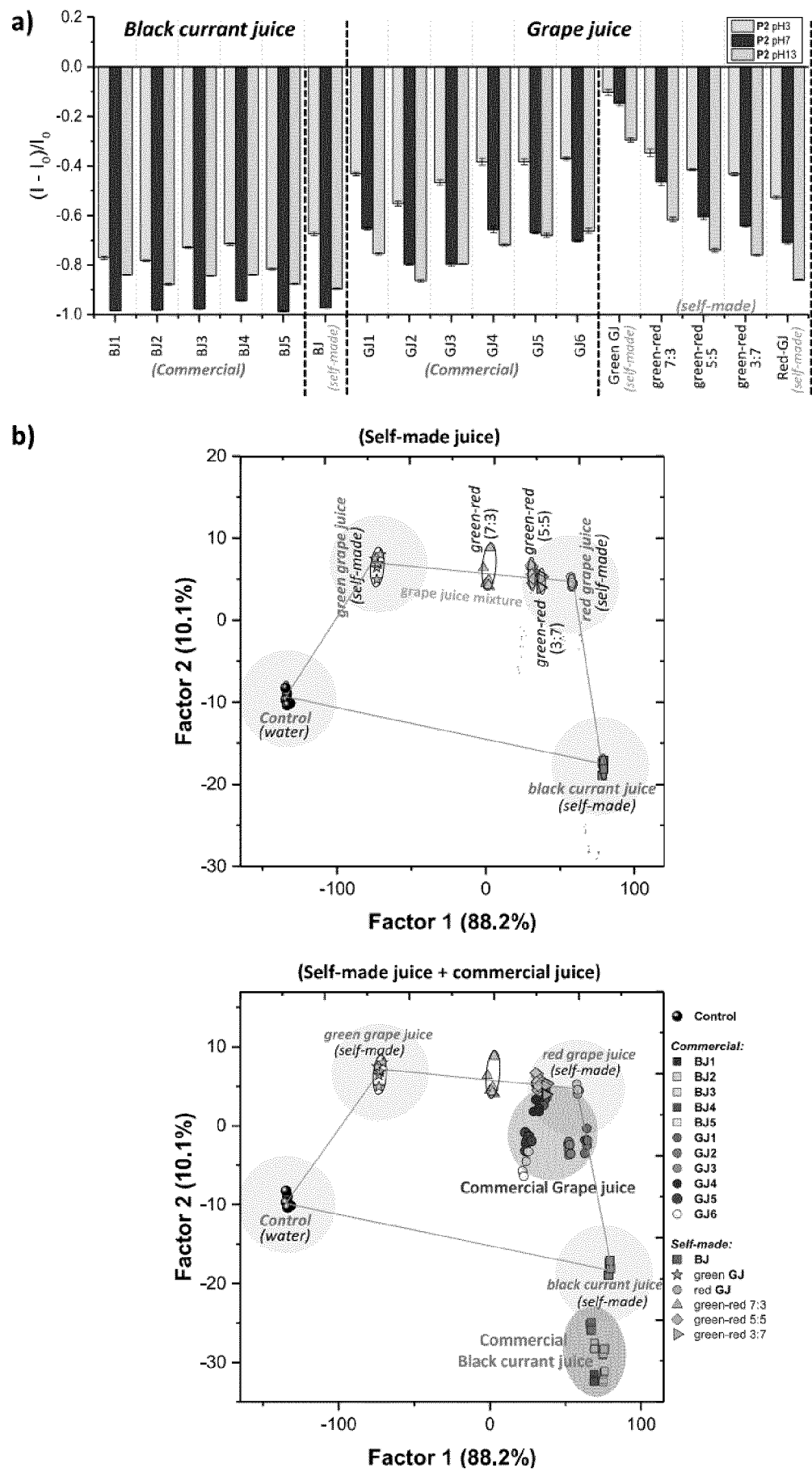
Fig. 9B



Figs 10A-10C



Figs. 11



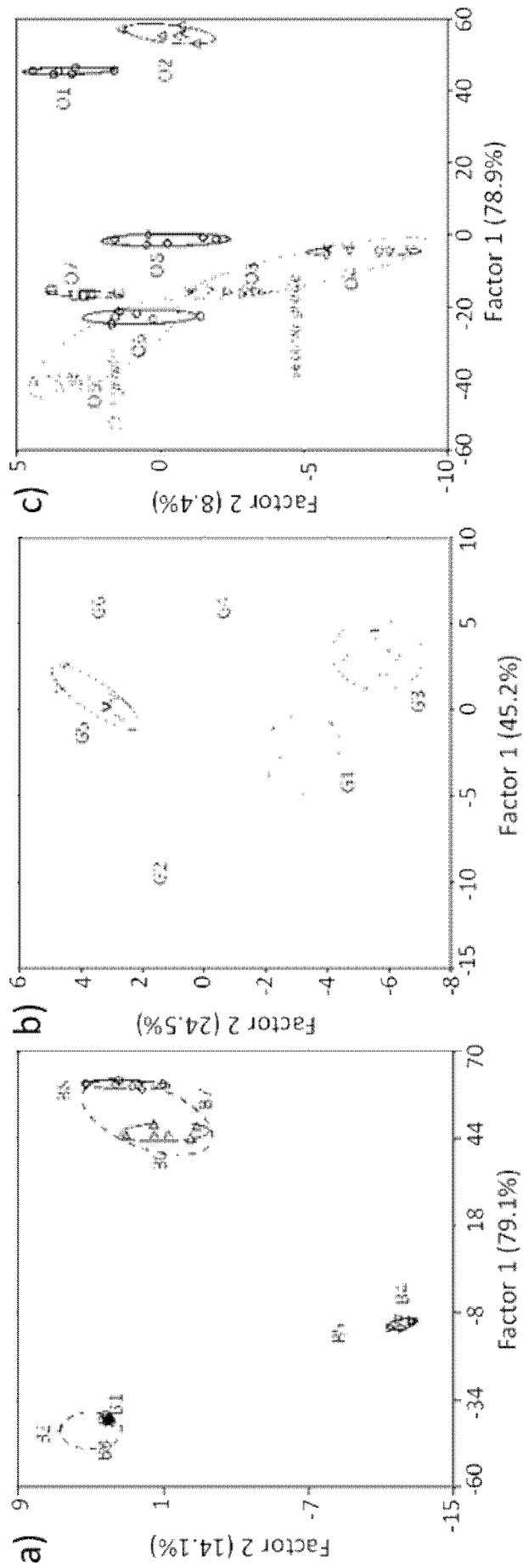


Fig. 12

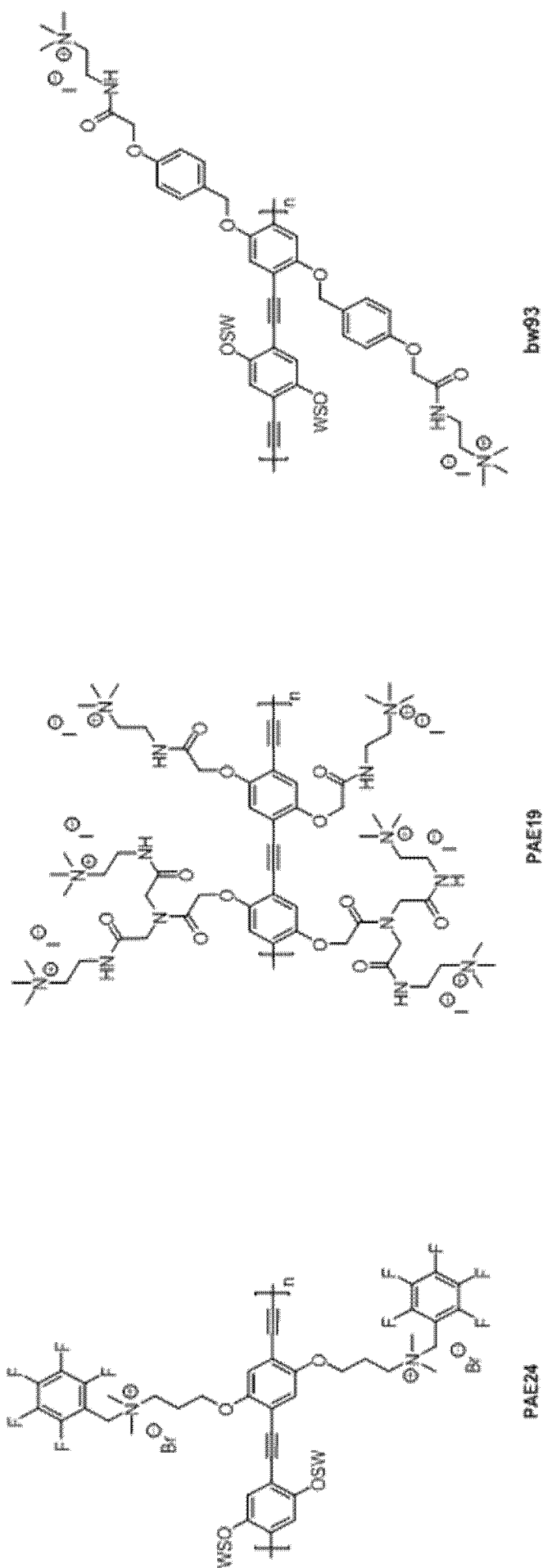


Fig. 13

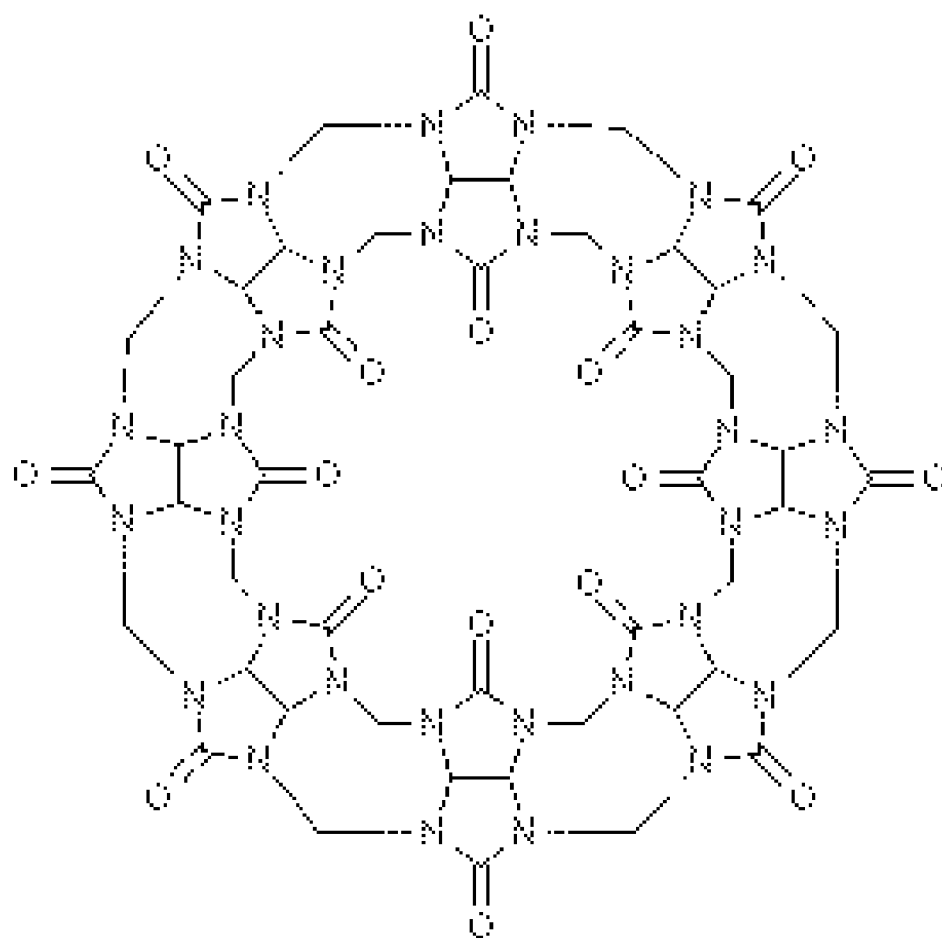


Fig. 14

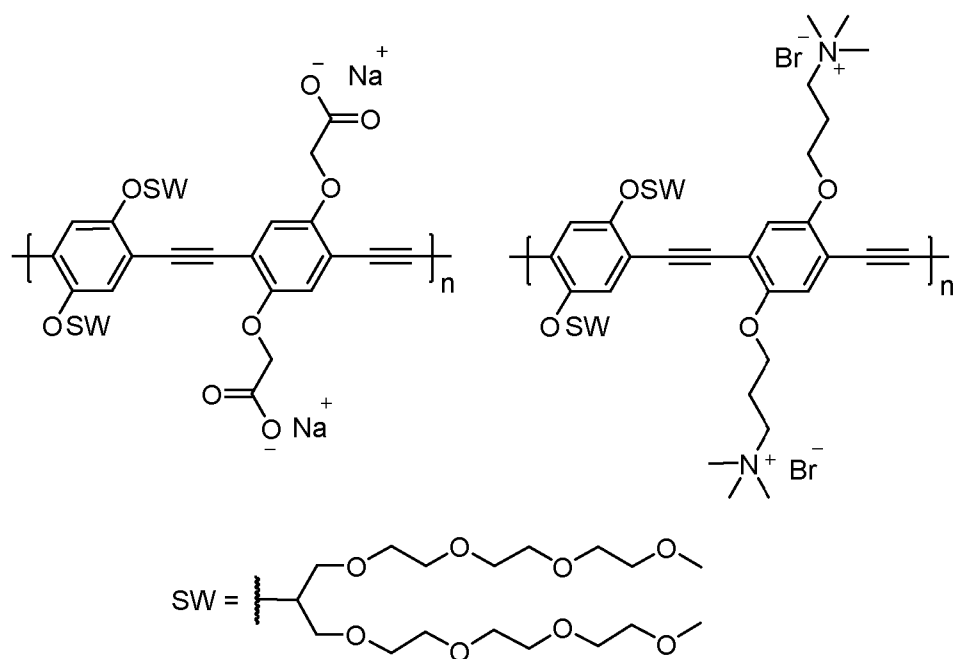
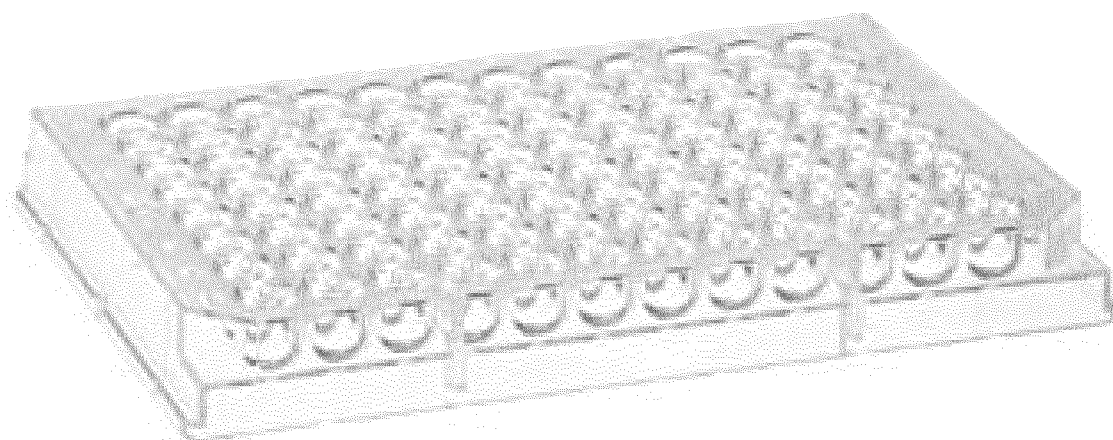


Fig. 15



INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2017/073949

A. CLASSIFICATION OF SUBJECT MATTER

INV. G01N21/64 C09K11/06 C08G61/12 G01N21/77
ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

G01N C09K C08G

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	JINSONG HAN ET AL: "Water-Soluble Poly(p-aryleneethynylene)s: A Sensor Array Discriminates Aromatic Carboxylic Acids", ACS APPLIED MATERIALS & INTERFACES, vol. 8, no. 31, 29 July 2016 (2016-07-29), pages 20415-20421, XP055438648, US ISSN: 1944-8244, DOI: 10.1021/acsami.6b06462 cited in the application the whole document ----- -/--	1-6



Further documents are listed in the continuation of Box C.



See patent family annex.

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

9 January 2018

Date of mailing of the international search report

15/01/2018

Name and mailing address of the ISA/

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Authorized officer

Riblet, Philippe

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2017/073949

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	JINSONG HAN ET AL: "Identification of White Wines by using Two Oppositely Charged Poly(p -phenyleneethynylene)s Individually and in Complex", ANGEWANDTE CHEMIE INTERNATIONAL EDITION, vol. 55, no. 27, 3 June 2016 (2016-06-03), pages 7689-7692, XP055438658, ISSN: 1433-7851, DOI: 10.1002/anie.201602385 cited in the application the whole document -----	1-6