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Neurochemistry: Microelectrodes for listening to the brain

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Neurochemistry: Microelectrodes for listening to the brain

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Abstract

Electrochemical sensors are of increasing interest, especially those developed for direct application in biological areas such as pharmacology and neuroscience. This divulgation article evidences the results obtained by nanotechnology researchers using modified nanotube-microelectrodes, which can be used for neurotransmitter measurements in the brain.

Resumen

Los sensores electroquímicos son de interés creciente, en especial los elaborados para su aplicación directa en las áreas biológicas, como la neurociencia y la farmacología. Este artículo de divulgación estudiantil evidencia los resultados obtenidos por un grupo de investigadores en nanotecnología usando microelectrodos modificados con nanotubos, que pueden ser utilizados para mediciones de neurotransmisores en el cerebro.

Keywords: nanotube-microelectrodes • neurosciences • brain • neurotransmitters • neurochemistry

Neurochemistry: microelectrodes for listening to the brain

Upon neural stimulation, there will be rapid changes in extracellular concentration of several species, and the complete understanding of regulatory and signalling processes requires the real time monitoring of such species. In the past, the means for studying bioactive species release and extracellular concentration of signalling agents (i.e. neurotransmitters and modulators) have been directly limited in its temporal and spatial resolution relative to the dynamics of chemical signalling and the structures of interest in the brain [1, 2]. The monitoring of bioactive species in the brain are fundamentally based in vitro and/or in vivo measurements and these will allow increasing the advantages in experimental and in clinic areas. Likewise, these allow understanding the neurochemistry of brain function and use this knowledge to develop better treatment protocols [2, 3].

During the last years, two emerging areas in the science field have attracted the interest of the neuroscientists: nanotechnology and electrochemical microelectrodes which are leading to many promising application in neurosciences. Nanotechnologies exploit materials and devices with a functional organization that has been engineered at the nanometre scale. Nanomaterials have showed to interact with biological systems at fundamental molecular levels with a high degree of specificity [1]. By taking advantage of this unique molecular specificity, these nanotechnologies can stimulate, respond to and interact with target cells and tissues in controlled ways to induce desired physiological responses, while minimizing undesirable effects. Applications of nanotechnology in basic and clinical neuroscience are only in the early stages of development, partly because the complexities associated with interacting with nervous system. Despite this, an impressive body of research is emerging that hints at the potential contributions these technologies could make to neuroscience research [1].

On the other hand, electrochemical microelectrodes have shown to be an attractive methodological approach able to measure the concentration of chemical agents (in vivo/ in vitro) which play key roles in the regulation of cellular events and bioactive species which requires constant monitoring, i.e. glucose, uric acid, and some drugs [2]. These microelectrodes are of increasing relevance in many areas of biological monitoring, including the study of brain functions, and clinical monitoring of brain health during intensive care. In recent years considerable progress has been made in the application of the electrochemical microelectrodes to neurophysiology and neuropharmacology measurements, where neurotransmitters release and uptake have been investigated in single cells, brain slices, and the intact brain [2, 3]. Recently, the electrochemistry has allowed developing new electrochemical techniques for detecting several species, using different electrode materials and/or modified surfaces, minimizing the changes in the analyte signal. Moreover, the research work has focused on: i) the efficiency in detecting various bioactive species at different electrochemical microelectrodes; ii) the improvement of the electrocatalytic activity and electrochemical stability of the electrode materials; iii) the investigation of factors affecting the process performance, iv) and the exploration of neurochemical mechanisms and kinetic detections.

Using microelectrodes, researchers have able to measure neurotransmitters concentrations directly in situ. Any compound can oxidize on an electrode if the voltage is sufficiently great; however, some compounds, notably the biogenic amines such as

dopamine, histamine, serotonin and noradrenaline, may oxidize at more modest voltages (Figure 1) [3]. Electrochemical techniques such as the fast-scanning cyclic voltammetry can be used to obtain a characteristic signature for these compounds even in vivo. The current recorded by the microelectrode is then plotted against voltage and with some signal processing characteristic oxidation and reduction peaks occurring at specific voltages can be seen on the cyclic voltammogram, which enable identification of the chemical species. These techniques can also potentially be applied more widely (e.g. to the determination of purines) and related techniques have been used to measure nitric oxide levels in the brain [3]. For all these reasons, the combination of both areas will allow significant results in neurosciences.

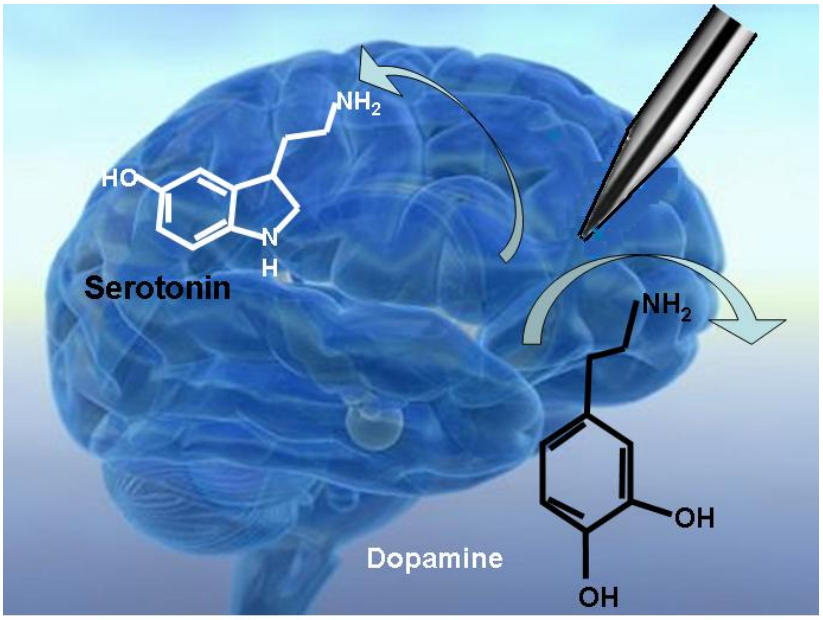

Figure 1. Representation of the electrochemical measurements of the dopamine and serotonin in the brain using a microelectrode.

Recently, the group of Prof. Venton showed new evidence about the electrochemical detection of neuro-transmitters using a singular kind of microelectrodes [4]. This relevant study demonstrated that the combination of two sciences, nanotechnology and electrochemistry, have significantly modified the scenery in neuroscience. This detailed study about the carbon-fiber microelectrodes modified with single-walled carbon nanotubes5 describes the co-detection of dopamine and serotonin in vivo measurements for the first time, using fast-scan cyclic voltammetry (electro-chemical technique).

Dopamine and serotonin are important neurotransmitters that interact in the brain. Serotonin is known to regulate sleep and is a common target for the treatment of depression. Dopamine has been linked to locomotion, reward and motivation and is a common target for illicit drugs. The loss of dopamine in the nigrostriatal pathway is the cause of Parkinson's disease. However, two are inherently linked; cocaine is known to act

on both dopamine and serotonin transporters, for example. Consequently, the simultaneous, rapid, in vivo detection of these compounds is a requirement if their interactions in the brain are to be understood. While dopamine is easily detected with electrochemical sensors, the detection of serotonin is more difficult because reactive species formed after oxidation can adsorb to the electrode, reducing sensitivity [4]. Microelectrodes prepared by Venton and co-workers were used to monitor stimulated dopamine and serotonin changes simultaneously in the striatum of anesthetized rat after administration of a serotonin synthetic precursor. These studies showed that nanotube-coated microelectrodes have a greater sensitivity and resistance to the fouling (effect of block the electrode surface with the organic compounds).

The simultaneous detection of dopamine and serotonin using other materials as microelectrodes has been already reported by other authors [6, 7]. In this study, different oxidation potentials were observed for dopamine and serotonin with slow-scan cyclic voltammetry and differential pulse voltammetry techniques. However, these methods are not suitable for in vivo use because the electrodes are large and the electrochemical methods cannot measure fast changes. For this reason, the experiments reported by Venton [4] were carried out using fast-scan cyclic voltammetry technique, in which the peaks are shifted to more positive potentials and serotonin and dopamine can be distinguished. For in vivo experiments, the animals were anesthetized and each rat was placed in a stereotaxic frame and holes drilled in the skull for the placement of the electrodes. A synthetic precursor of serotonin, 5-hydroxytryptophan (5-HTP) was administered to increase the amount of serotonin in the brain. The complete protocol used during in vivo experiments has been described by the authors where they took into consideration several chemical mechanisms to cause the release of serotonin from dopamine neurons.

Figure 2 reported in [4] provides a visualization of all the data collected in vivo during an electrical stimulation. According to described by Venton, it shows increased dopamine release, after administration of carbidopa, but before administration of the serotonin precursor 5-HTP. Oxidation of dopamine and reduction were achieved by zones plot, while graphics as function of the current vs. time showed the reductive currents for dopamine and serotonin around the time of the stimulation. Before administration of a serotonin precursor, no change was observed in the current at the expected reduction peak potential for serotonin. Twelve minutes after 5-HTP administration, two reduction peaks were observed. Twenty-four minutes after 5-HTP, larger amounts of serotonin were observed and reduction peaks for serotonin and dopamine had similar peak heights. Concentrations at 24 min were estimated from pre-calibration curves about 250 nM and 130 nM, for dopamine and serotonin respectively [4].

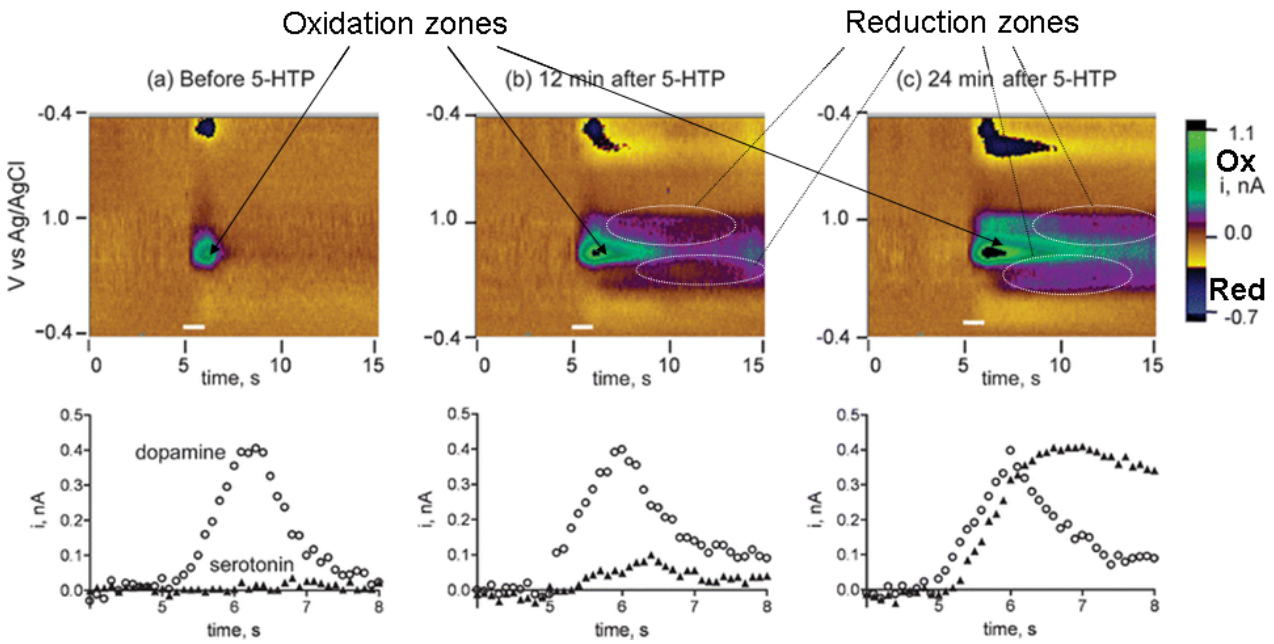

Figure 2. In vivo detection of dopamine and serotonin by CNTs. Superior plots show the data in three dimensions, with oxidation and reduction currents. The duration of the stimulation is marked as a white bar on the bottom of the color plot. Graphics below are current vs. time traces for the potentials where reduction peaks for dopamine (○) and serotonin (▲) would be detected. Reduction currents are shown in the positive direction for visual clarity, and only the time around the stimulation is depicted. (a) Before the serotonin precursor 5-HTP was administered, 30 min after carbidopa (25 mg kg–1), the superior plot is characteristic of dopamine release. The current vs. time traces show only changes in dopamine. (b) Twelve minutes after administering 5-HTP, reduction peaks for both dopamine and serotonin can be seen in the superior plot and the oxidation peak lasts longer. The current vs. time traces indicate that both dopamine and serotonin are released. (c) Twenty-four minutes after 5-HTP, there is significant release of serotonin, with nearly equal reduction peak currents. However, the time course of dopamine and 5-HT release are different, and serotonin appears to lag behind the dopamine release. Adapted from Venton et al.

Other remarks obtained from this study is that the appearance of serotonin reduction peak delays dopamine reduction peak in the time. This phenomenon was illustrated by the authors through cyclic voltammograms at different time points. As can be observed from Figure 3, 12 min after 5-HTP administration, the cyclic voltammogram at some seconds looks like dopamine and has no evidence of a serotonin reduction peak. After few seconds, an additional reduction peak was starting to appear. Finally, the reduction peak for dopamine was disappeared and this cyclic voltammetric profile was characteristic of serotonin. While electrode response to serotonin is slower, both dopamine and serotonin showed an increase in reductive currents directly after a mixture was exposed to the electrode. According to results reported by Venton, the delay might indicate a different biological mechanism for release. Serotonin may not be released by exocytosis but by a different mechanism such as reverse transport, which might account for the delay [4]. However, authors proposed new experiments to better explain these findings.

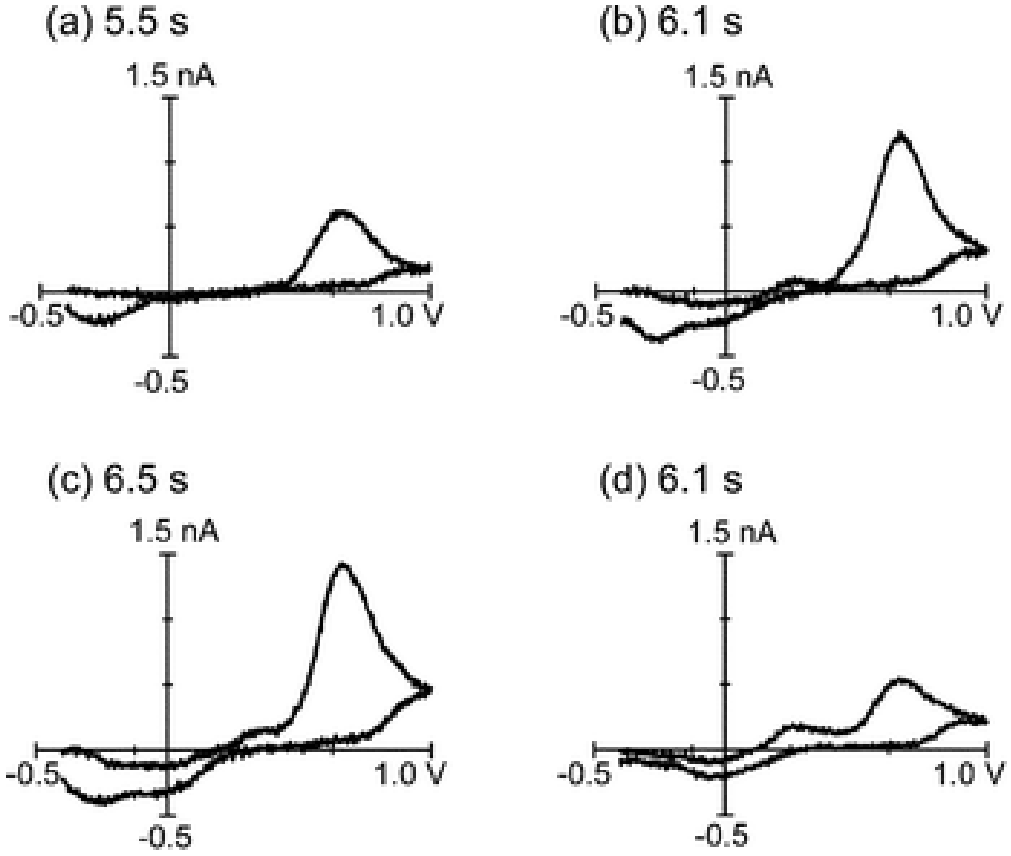

Figure 3. Individual cyclic voltammograms 12 min after 5-HTP administration: (a) at 5.5 s, during the stimulation, the cyclic voltammogram is characteristic of dopamine; (b) at 6.1 s, the reduction peak for serotonin begins to appear; (c) at 6.5 s, both dopamine and serotonin are present; and (d) by 7.5 s, the reduction peak for dopamine has disappeared and the cyclic voltammogram is characteristic of serotonin.

The recent advances obtained with carbon nanotubes modified microelectrodes suggest that their application to biological studies should be rapidly developed because of their better performance respect to other anode materials [6-8]. The fast neurotransmitters monitoring and good identification achieved during in vivo measurements are important profit determinants with the adoption of carbon nanotubes microelectrodes. Imagine, for example, the use of such practical commercial technology for detecting several analytes at once, novel biosensors for new neurochemicals of interest and also clinical applications.
This topic represents an important advance in the application of new technologies to monitor several bioactive species and it also is a challenger for the scientific community because future developments will rely upon the close collaboration of analytical chemists, engineers, neuroscientits and other scientists to ensure effective application and exploitation of new methods for monitoring neurotransmitters in vivo and answering important biological questions.

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