Anal. Chem. **2000,** *72,* 1632−1638

Electrochemical Oxidation of Histamine and Serotonin at Highly Boron-Doped Diamond Electrodes

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The electrochemistry of histamine and serotonin in neutral aqueous media (pH 7.2) was investigated using polycrystalline, boron-doped diamond thin-film electrodes. Cyclic voltammetry, hydrodynamic voltammetry, and flow injection analysis (FIA) with amperometric detection were used to study the oxidation reactions. Comparison experiments were carried out using polished glassy carbon (GC) electrodes. At diamond electrodes, highly reproducible and well-defined cyclic voltammograms were obtained for histamine with a peak potential at 1.40 V vs SCE. The voltammetric signal-to-background ratios obtained at diamond were 1 order of magnitude higher than those obtained for GC electrodes at and above 100 μM analyte concentrations. A linear dynamic range of 3−4 orders of magnitude and a detection limit of 1 μM were observed in the voltammetric measurements. Well-defined sweep rate-dependent voltammograms were also obtained for 5-hydroxytryptamine (5-HT). The characteristics of the voltammogram indicated lack of adsorption of its oxidation products on the surface. No fouling or deactivation of the electrode was observed within the experimental time of several hours. A detection limit of 0.5 μM (signal-to-noise ratio 13.8) for histamine was obtained by use of the FIA technique with a diamond electrode. A remarkably low detection limit (10 nM) was obtained for 5-HT on diamond by the same method. Diamond electrodes exhibited a linear dynamic range from 10 nM to 100 μM for 5-HT determination and a range of 0.5−100 μM for histamine determination. The FIA response was very reproducible from film to film, and the response variability was below 7% at the actual detection limits.

Highly boron-doped diamond thin films are emerging as unique electrode materials for several applications, including electroanalysis,[1−7] energy storage devices,[8] and electrosynthesis.[9] The attractive features of diamond include a wide electrochemical potential window in aqueous solutions,[10,11] very low voltammetric background current (∼1 order of magnitude lower than that of glassy carbon (GC)),[12,13] high resistance to deactivation via fouling,[1] extreme electrochemical stability,[13] and relative insensitivity to dissolved oxygen.[14] All of these properties make diamond a promising material, especially for electroanalysis.

There have been several reports in the past few years describing the outstanding performance of diamond electrodes for the electroanalysis of NADH,[1] dopamine,[3] polyamines,[4] and azide.[5] In most of these studies, diamond was found to outperform GC in terms of stability and sensitivity. Fundamental studies on boron-doped diamond (BDD) films have revealed that the lack of oxygen functional groups[5] on the as-deposited diamond surface and the very low tendency for adsorption of most chemical species[15] on the inert surface of diamond are mainly responsible for the superior performance of diamond electrodes. Our recent report on NADH oxidation is a notable example, in which we have demonstrated high sensitivity, high reproducibility, and long-term stability of diamond electrodes in air-saturated aqueous solution.[1] The voltammetric response of the diamond electrode was found to be reproducible for three months without any specific pretreatment, unlike the case of GC, which was deactivated in a few hours, with a significant anodic shift in the oxidation potential and subsequent fouling. The deactivation and fouling of GC electrodes is well known.[16,17]

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10.1021/ac9908748 CCC: $19.00
Published on Web 03/07/2000

Histamine and serotonin, also known as 5-hydroxytryptamine (5-HT), are important biogenic amines present in many food products and act as chemical messengers in biological systems.[18−20] Histamine is produced by the bacterial decarboxylation of histidine, the corresponding amino acid, which is present particularly in fish tissues.[18] Histamine is naturally present in many vegetables and fruits in amounts below toxic levels. When beer, wine, cheese, and fish in oil are not processed and packed using hygienic methods, the amounts of histamine can increase to toxic levels, causing food poisoning.[18] Therefore, the detection of histamine is very important as a monitor for hygienic manufacturing processes. Histamine is also a neurohormone synthesized in mast cells, which originate from bone marrow and can cause a variety of allergic reactions.[20] Serotonin is a neurotransmitter that is co-released along with histamine in mast cells. It is also present in meat and meat products along with other amines. Its role is very significant in gastrointestinal disorders[21] and psychiatric disorders such as depression.[19]

Traditionally, histamine (HI) has been determined by use of liquid chromatographic techniques with precolumn or postcolumn derivatization and spectrofluorometric detection.[18,22−24] Although electrochemical detection methods are inexpensive and provide high sensitivity, not much work has been reported on this type of detection, probably due to the high oxidation potential (∼1.2 V vs SCE) of HI, at which conventional electrodes such as GC exhibit high background currents due to oxidation of the solvent water or of the electrode material itself. One report on the direct electrochemical detection involves integrated pulsed amperometric detection (IPAD) at a gold electrode following liquid chromatographic separation.[25] Determination of serotonin along with histamine has been carried out mainly in mast cells by use of fast-sweep voltammetry,[20] laser vaporization-mass spectrometry,[26] and capillary electrophoresis with fluorescence detection.[21] Carbon fiber electrodes have most often been used in electrochemical detection by fast-sweep voltammetry. At slow and moderate sweep rates, carbon fiber electrodes and highly oriented pyrolytic graphite electrodes undergo deactivation due to strong adsorption of oxidation products of 5-HT, forming an insulating layer on the electrode surface.[19] Diamond, due to the inert nature of the surface, is a possible electrode material for simple amperometric determination of these amines. Recently, Swain and co-workers demonstrated the use of diamond electrodes in the flow injection analysis (FIA) determination of polyamines.[4]

In the present study, we report the use of high-quality, heavily boron-doped diamond electrodes for the electrochemical oxidation of histamine and serotonin by use of linear and cyclic voltammetry, hydrodynamic voltammetry, and FIA with electrochemical detection. Boron-doped diamond electrodes are shown to be superior to GC electrodes due to diamond's higher sensitivity, stability, and reproducibility. Diamond is demonstrated to be the best electrode material for the detection of histamine, possessing high sensitivity, even at high oxidation potentials, and for the detection of serotonin, which fouls other electrodes such as GC after oxidation. High stability and remarkable detection limits for the determination of histamine and serotonin by simple amperometry are notable points of this work.

EXPERIMENTAL SECTION

The boron-doped diamond thin films were grown with a microwave plasma chemical vapor deposition (MPCVD) system (ASTeX Corp., Woburn, MA). Films were grown on Si (100) substrates. The details of the preparation have been described previously.[14] A mixture of acetone and methanol in the ratio of 9:1 (v/v) was used as the carbon source. B_2O_3, the boron source, was dissolved in the acetone−methanol at a B/C molar ratio of 10^4 ppm. High-purity hydrogen was used as the carrier gas. The bubbling of the acetone−methanol−B_2O_3 solution was carried out at ∼25 °C. The deposition of the film was carried out at a microwave power of 5 kW. A film thickness of ∼40 μm was achieved after 10 h of deposition. The film quality was confirmed by Raman spectroscopy.

Electrochemical measurements were carried out in a single-compartment cell with a saturated calomel electrode (SCE) as the reference and platinum foil as the counter electrode. Cyclic voltammograms were recorded with a combined potentiostat−function generator (Toho Technical Research, PS-07) and X−Y recorder. The GC electrode (GC-20 plate or GC-20 rod, Tokai Carbon Co., Ltd.) was pretreated by polishing with alumina (1 μm), followed by ultrasonication in high-purity water before the experiment. The electrolyte was a mixture of 0.05 M KH_2PO_4 + 0.05 M K_2HPO_4 (KH_2PO_4/K_2HPO_4, pH 7.1 ± 0.1).

The FIA system used in the present study, consisted of a micro-LC pump (Bioanalytical Systems, LC-100), an injector (Rheodyne) with a 20-μL injection loop, a thin-layer flow cell (Bioanalytical Systems), an amperometric detector (Bioanalytical Systems LC-4C), and an X−Y recorder (Graphtec, WX4000). The upper limit of the flow rate set for the pump was 1 mL min^{-1}. The wall-jet-type flow cell consisted of the Ag/AgCl reference electrode and a stainless steel tube counter electrode, which also served as the tube for the solution outlet. A 0.5-mm-thick silicon rubber gasket was used as a spacer in the cell. The geometric area of the diamond electrode in the cell was estimated to be 0.64 cm^2. The cell volume was estimated to be 24 μL by assuming a 25% compression of the gasket. Phosphate buffer (KH_2PO_4/K_2HPO_4, pH 7.1 ± 0.1) was used as the mobile phase. The sample solutions were prepared with the same buffer. The flow rate was set at 1 mL min^{-1} and was confirmed before every experiment by measuring the volume of the buffer collected at the outlet at 10-min intervals.

Hydrodynamic voltammograms were obtained for each compound before the amperometric determination was performed. The data were obtained by recording the background current at a fixed potential, after a delay of 15 min to obtain a stable response, followed by the injection of the 20-μL analyte aliquot. The peak current after each injection was recorded, together with the

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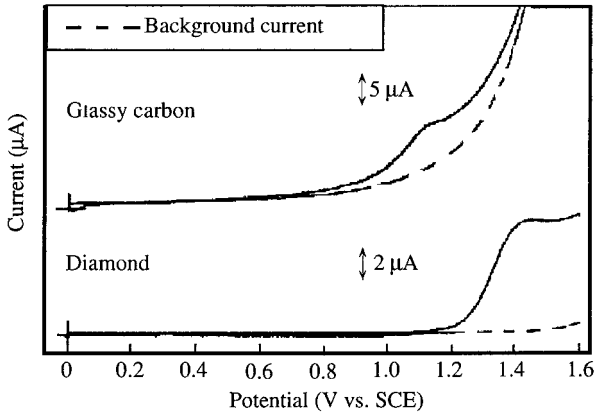

Figure 1. Linear sweep voltammograms for 100 μM HI in 0.1 M phosphate buffer (pH 7). (1) GC electrode (0.196 cm^2); (2) diamond electrode (0.189 cm^2). The potential sweep rate was 100 mV s^{-1}.

corresponding background current. These data were plotted as a function of applied potential to obtain hydrodynamic voltammograms. The amperometric measurements were carried out at an applied potential where the signal-to-background (S/B) ratio was found to be at a maximum in the hydrodynamic voltammogram, or in the limiting current range.

Histamine, serotonin hydrochloride, 5-hydroxy-3-indole acetic acid, KH_2PO_4, and K_2HPO_4 were obtained from Wako Chemical Co. and were used without further purification. All of the solutions were prepared using ultrapure (18 MΩ) water.

RESULTS AND DISCUSSION

Voltammetric Studies. Figure 1 shows linear sweep voltammograms for 100 μM HI together with the corresponding background voltammograms in 0.1 M phosphate buffer (pH 7) at GC and BDD electrodes. At the GC electrode, the oxidation of HI occurred at ~1.2 V vs SCE. A rapid increase in the current at this potential was also observed in the background voltammogram due to oxygen evolution and carbon oxidation. For this reason, the voltammogram was ill-defined. In the case of the diamond electrode, a very well-defined oxidation peak at 1.40 V vs SCE was observed. It is pertinent to note that very low background current (3.18 μA cm^{-2}) was observed at the potential limit of 1.6 V vs SCE. A rise in the background current at ~1.5 V was observed for relatively low quality films (based on Raman spectra), which were also characterized by relatively small grains (1−5 μm). The HI oxidation peak was clearly seen even on such films. The oxidation peak current decreased after the first cycle but reverted back to the original current after a time delay of 3 min without stirring, or 1 min with stirring, indicating the absence of electrode fouling. The background-corrected voltammetric peak current response was found to be very linear ($r = 0.98$) in the concentration range examined (0−100 μM). The peak current varied linearly with the square root of sweep rate, $v^{1/2}$, indicating semi-infinite linear diffusion of reactant to the electrode surface. This curve was also highly linear ($r = 0.99$) up to a sweep rate of 500 mV s^{-1} and passed through the origin.

Table 1 presents a comparison of the voltammetric data obtained for a GC electrode and two diamond film electrodes obtained from different batches. Both of the films were of high quality, as confirmed by Raman. The voltammetric data obtained at two different concentrations, 100 and 500 μM, are summarized in the table. The background current at 1.2 V vs SCE for GC was ~7 times higher than that for the diamond electrode. For these two concentrations, the S/B ratios were 1 order of magnitude higher for the diamond electrode than that for the GC electrode. At 10 μM and below, no discernible voltammetric peak could be obtained at the GC electrode. For the diamond electrodes, even at a concentration as low as 1 μM, a peak could be obtained with a S/B ratio of ~3. It is evident from the oxidation peak potentials and current responses given in the table that the oxidation of histamine is reproducible from film to film. The low background current and high S/B ratios associated with the diamond electrode lead to high sensitivities for histamine determination, even though the peak potential is relatively high.

Table 1. Comparison of S/B Ratios Obtained from the Cyclic Voltammetric Data for the Oxidation of Histamine in 0.1 M Phosphate Buffer (pH 7), for Diamond and GC Electrodes[a]

electrode	histamine (μM)	E_p^{ox} (mV)	I_p^{ox} (μA)	S/B ratio
D31298	500	1440	31	26
D71298	500	1440	31	25
GC	500	1200	28	3.5
D31298	100	1380	6	5
D71298	100	1400	5.8	4.8
GC	100	1130	6	0.75

[a] Sweep rates, 100 mV/s.

Because 5-HT is also an important neurotransmitter, which coexists with histamine in mast cells and is present in several meat products, cyclic voltammetric studies of this compound were carried out under identical conditions. It is well known that 5-HT adsorbs strongly on pyrolytic graphite[27] and carbon fiber electrodes,[20] thus providing good sensitivity. However, the oxidation products form insulating layers on the electrode surface, resulting in fouling.[19] Diamond is expected to overcome this problem due to the relative inertness of the surface. Figure 2a shows cyclic voltammograms for 10 μM 5-HT at a diamond electrode. Cyclic voltammograms were also obtained for GC for comparison (Figure 2b). Both of the electrodes yielded voltammograms with nearly identical features. However, the voltammograms obtained at the diamond electrode were somewhat better defined, with an oxidation peak at 0.42 V vs SCE. This value is slightly higher than that for the GC electrode, as in the case of HI. Indeed, diamond exhibits relatively high overpotentials for a number of irreversible oxidation reactions.[1,5] The amplitude of the peak current calculated for the irreversible oxidation wave determined at 100 mV s^{-1}, assuming 4-electron oxidation, is 0.53 μA. This value is in agreement with the observed peak current, 0.72 μA, considering the roughness of diamond surface. In the case of the GC electrode, the higher peak current is known to be due to serotonin adsorption on the surface.[19] The peak current for diamond was found to be linearly dependent on $v^{1/2}$ ($r = 0.993$), indicating a diffusion-controlled electrochemical process. A positive shift in the peak potential was observed with increasing sweep rate, i.e., a 100-mV shift by increasing the sweep rate from 10 to 500 mV s^{-1}. The most interesting observation is that the voltammograms for

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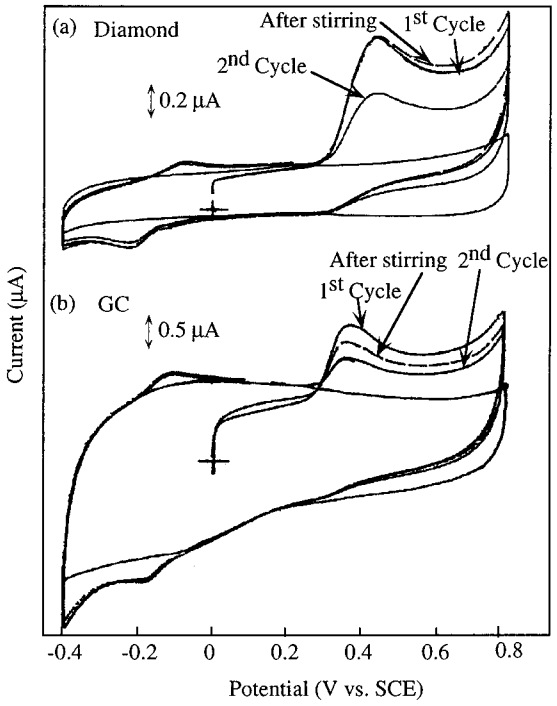

Figure 2. Cyclic voltammograms for 10 μM serotonin in 0.1 M phosphate buffer (pH 7): (a) diamond electrode (0.07 cm^2); (b) GC electrode (0.07 cm^2). The background current is also shown. The potential sweep rate was 100 mV s^{-1}.

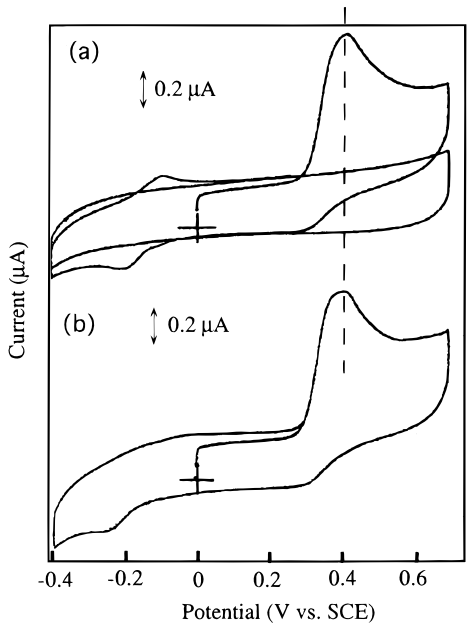

Figure 3. Cyclic voltammograms for 10 μM serotonin in 0.1 M phosphate buffer (pH 7): (a) as-deposited diamond electrode; (b) diamond electrode after oxidizing the surface at 1.8 V vs SCE for 10 min in the same electrolyte.

serotonin at the diamond electrode could be recovered by stirring the solution after the second cycle, in contrast to GC, where the recovery of the voltammogram was only partial, even after vigorous stirring. This result indicates the absence of electrode fouling. Similar results were obtained for 5-hydroxyindoleacetic acid (5-HIAA), which is a metabolite of 5-HT and an anion at physiological pH. The oxidation of this compound occurred with a peak at 0.49 V vs SCE, the value being slightly higher than that for 5-HT.

A redox couple observed in the more negative potential region (centered at −0.15 V, Figure 2) is due to an adsorbed quinone that is an oxidation product of serotonin.[27] It is interesting to note that the peak separation ΔE_p for this relatively reversible couple was ∼35 mV for the GC electrode, which is characteristic of a 2-electron process involving an adsorbed species. Wrona and Dryhurst[27] also observed a similar symmetric redox couple and confirmed the strong adsorption of the oxidation product species on a pyrolytic graphite electrode by the observation of the linear dependence of the peak currents on v. However, the voltammogram obtained for the diamond electrode exhibited a ΔE_p value of ∼110 mV, with relatively broad, asymmetric peaks. The shape of the voltammogram indicates the characteristic behavior of a diffusion-controlled reaction. The ΔE_p value for diamond was found to be pH-dependent; e.g., a ΔE_p value of ∼400 mV was observed at pH, 2.12. This value is in agreement with the value reported for anthraquinonedisulfonate in 0.1 M $HClO_4$ at an as-deposited diamond electrode.[15] Also, the peak current in the present study was linearly dependent on $v^{1/2}$, characteristic of a diffusion-controlled reaction.

A detailed study related to the adsorption of quinones, specifically, the polar compound 2,6-anthraquinonedisulfonate, on various pretreated carbon, HOPG, and diamond surfaces was reported by Swain and co-workers.[15] They observed similar differences between GC and BDD electrodes and attributed the inertness of the diamond surface to hydrogen termination. They further proved this point by intentionally hydrogenating the GC surface in order to obtain voltammograms characteristic of a diffusion-controlled reaction. Polar oxygen functional groups were believed to be responsible for the strong adsorption of this compound on the GC surface. We are now studying the adsorption properties of such polar molecules on diamond surfaces subjected to various treatments, including electrochemical anodization and oxygen plasma treatment, to determine whether the oxygen functional groups have any influence on adsorption. As a part of the preliminary work, we have obtained a voltammogram for 5-HT at an electrochemically oxidized diamond electrode (Figure 3). A moderate anodic treatment at +1.8 V for 10 min in phosphate buffer was shown to increase the oxygen content on the surface to ∼10%.[28] The purpose of the moderate oxidation was also to examine the electrode response under mild oxidative conditions such those that would be encountered during long-term use and exposure to the laboratory environment. The 5-HT oxidation peak for this electrode did not shift, and the peak current was also unchanged. Surprisingly, no decrease in the peak separation of the redox couple corresponding to the quinone was observed, indicating no enhancement of adsorption due to the introduction of oxygen-containing functional groups. On the contrary, the oxidation peak for the couple almost disappeared, and the reduction peak moved to more negative potentials. This preliminary result indicates that the diamond electrode, even after surface oxidation, behaves differently from a polished GC electrode having oxygen groups present on it. These results indicate that both as-

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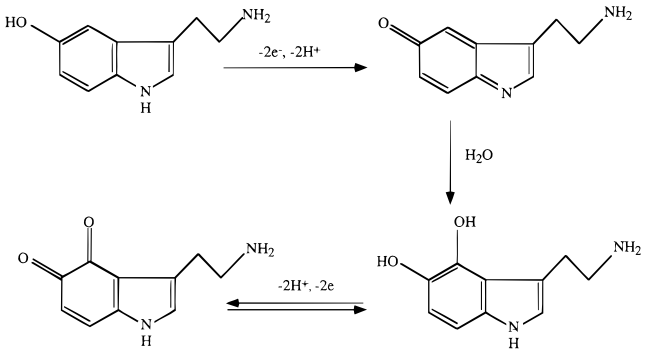
Scheme 1



deposited and oxidized diamond surfaces are inert with respect to adsorption, explaining the higher stability of response compared to GC. The remarkable stability of diamond for NADH determination was demonstrated by us earlier.[1]

The electrochemical oxidation of 5HT and 5-HIAA is believed to take place at the phenol group of the molecule to form the corresponding ketone (Scheme 1). The absence of the corresponding reduction peak indicates the instability of this oxidation product, which is thought to then undergo chemical reaction to form a product that is easily oxidizable. This product is believed to be the reduced hydroquinone.[27,29] The redox couple centered at −0.15 V can be attributed to this quinone/hydroquinone couple. The peak potential is in agreement with the value reported.[27] Strong adsorption of this quinone is undesirable for the electrochemical detection of 5-HT. Such strong adsorption was also observed by Jackson et al.[19] on carbon fiber electrodes, the adsorption being much more stronger than that of 5-HT itself. This fact is evident from the highly reversible couple observed in the present study at the GC electrode and by Wrona and Dryhurst on pyrolytic graphite.[27] The adsorption was found to be much lower in the case of diamond, as is evident from the diffusion-controlled peak behavior and also by the ability to recover the background voltammogram after removing the electrode from the 5-HT solution and placing it in fresh electrolyte.

It is also interesting to compare the background voltammograms for the diamond and GC electrodes (Figure 2). The background current for the GC electrode at 0.4 V (oxidation peak potential for serotonin) was at least 5 times higher than that for the diamond electrode. This indicates the possibility of obtaining high sensitivities at diamond electrodes. Furthermore, the S/B value obtained from the voltammogram for diamond was 3.5 times higher than that for GC. Although, the presence of oxygen on the GC surface is believed to be one of the reasons for its high background current,[5] surface oxygen does not appear to be a major factor in the case of diamond in determining the background current. For example, the background current observed for electrochemically oxidized diamond (Figure 3) was essentially identical to that of the untreated one. This suggests that the lack of porosity, rather than the oxygen content, is a main factor contributing to the low background currents for the diamond electrode. Popa et al.[3] demonstrated the high degree of sensitivity for diamond even after severe electrochemical oxidation. The high degree of resistance to electrochemical and chemical oxidative attack, which prevents the surface from acquiring porosity, allows the electrode to exhibit reproducible cyclic voltammograms even after oxidative treatment of the surface.

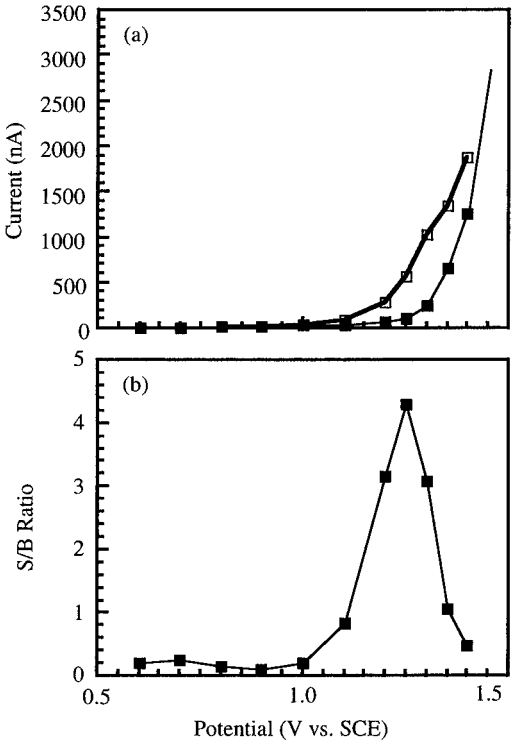


Figure 4. (a) Hydrodynamic voltammogram for 20-μL injections of 100 μM HI in 0.1 M phosphate buffer; (b) signal-to-background ratio. The flow rate was 1 mL min^{-1}.

Flow Injection Analysis. In the FIA measurements, an important observation was that the diamond electrode produced a stable background current within 15 min after switching to the desired oxidizing potential, in contrast to GC, which required ~45 min to attain a reasonably stable current. Figure 4a shows the hydrodynamic voltammogram obtained at a diamond electrode for 20-μL injections of 100 μM HI in 0.1 M phosphate buffer. The corresponding background voltammogram is also shown. Although the voltammogram for HI oxidation did not exhibit a sigmoidal shape, probably due to the high oxidation potential and high flow rate,[4] a significant current due to the oxidation of histamine was observed above ~1.2 V vs SCE. Figure 4b shows the S/B ratio calculated from Figure 4a as a function of potential. The S/B ratio reached a maximum value of ~4.5 at 1.28 V. Hence, this potential was selected for amperometric detection in FIA experiments. However, in the case of 5-HT, we obtained sigmoidal voltammograms with a half-wave potential (0.36 V vs Ag/AgCl) very similar to the peak potential obtained in the cyclic voltammogram. The detection potential in this case was set at 0.43 V vs Ag/AgCl, in the limiting current range. A similar sigmoidally shaped hydrodynamic voltammogram was obtained for 5-HIAA, with a half-wave potential of 0.41 V vs Ag/AgCl.

Figure 5 shows the amperometric response of a diamond electrode for repetitive 20-μL injections of HI at various concentrations in phosphate buffer at a detection potential of 1.28 V vs Ag/AgCl. Well-defined and highly reproducible signals were obtained for each concentration. The inset in this figure shows the response

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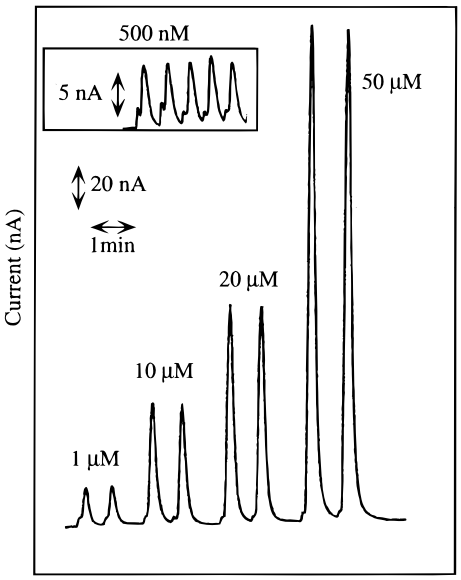

Figure 5. Electrochemical detection results for FIA with a diamond film and 20-μL injections of various concentrations of HI in 0.1 M phosphate buffer. The applied potential was 1.28 V vs Ag/AgCl, and the flow rate was 1 mL min^{-1}.

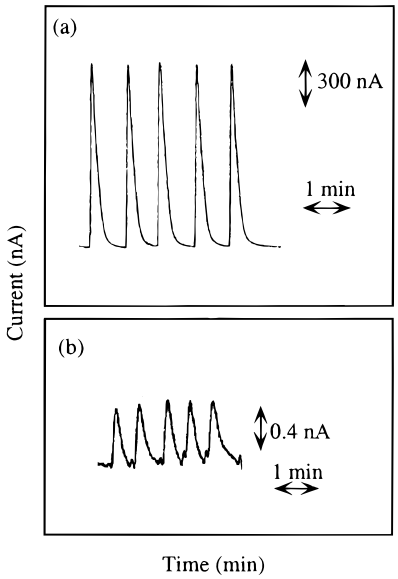

Figure 6. Electrochemical detection results for FIA with a diamond film and 20-μL injections of (a) 50 μM and (b) 10 nM 5-HT in 0.1 M phosphate buffer. The flow rate was 1 mL min^{-1}, and the applied potential was 0.43 V vs Ag/AgCl.

Table 2. Linear Dynamic Range and Signal-to-Noise Ratio for HI, 5-HT, and HIAA at Boron-Doped Diamond Electrodes

			at actual detection limit	
amine	linear dynamic range	sensitvty (nA/μM)	S/N ratio	peak ht variability (%)
HI	500 nM−100 μM (r = 0.9997)	4	13.8 (500 nM)	3−6
5-HT	10 nM−50 μM (r = 0.9992)	25	18.1 (20 nM)	3−7
5-HIAA	100 nM−100 μM (r = 0.9984)	17	32.9 (100 nM)	3−7

for the injections of 500 nM HI. The signal current increased linearly with increasing concentration from 500 nM to 100 μM (r = 0.99). The response variability was ∼5% (number of injections, 15), even for the detection of 500 nM. The signal-to-noise (S/N) ratio (15 injections) for the detection of a 500 nM was typically 13, indicating the remarkable performance of the diamond electrode for the electroanalysis of HI, despite its high overpotential. The electrode response was found to be reproducible from film to film within ±3%. Although, we have not tested diamond electrodes for long-term stability under continuous operation, they exhibited highly reproducible responses from day to day. Also, the response was reproducible after several days of exposure to the laboratory atmosphere.

Figure 6a shows a similar response for 50 μM 5-HT in phosphate buffer at a detection potential of 0.43 V vs Ag/AgCl. Excellent response reproducibility was observed, with the peak current variation being ∼±2%. The highly reproducible response of diamond for repetitive injections of even 10 nM 5-HT in the same medium is noteworthy (Figure 6b). The well-defined peaks were reproducible, with a peak variability of ∼3% (n = 15). The calibration plots showed excellent linearity from 10 nM to 50 μM (r = 0.99). We have obtained similar responses with 5-HIAA, with an experimental detection limit of 100 nM, with a S/N value of 32 (n = 15). A linear calibration plot (r = 0.99) was obtained up to 100 μM. A summary of the results is presented in Table 2. For all three analytes, detection limits in the nanomolar range were achieved, the values being within the range encountered in real samples, for example, ∼20 nM serotonin in human plasma[30] and ∼200 nM histamine in whole blood[31] detected by other methods. Diamond exhibited high sensitivities, with a wide linear dynamic range, which were not obtainable with GC. These results indicate the superior properties of diamond.

CONCLUSIONS

High-quality, conductive, boron-doped diamond thin-film electrodes are shown to exhibit excellent performance for the oxidative detection of the biogenic amines HI and serotonin. Well-defined voltammograms were obtained for HI at diamond electrodes, demonstrating significant advantages compared to GC. The oxidation of serotonin was found to occur at diamond electrodes, with a very low and reversible adsorption of the quinone oxidation product, unlike the case of GC, which is known to be vulnerable to deactivation and fouling due to strong adsorption of this oxidation product. The comparison of the voltammograms for diamond and GC indicates the superior behavior of diamond in terms of surface inertness to adsorption and response sensitivity. FIA results indicate that the two biogenic amines can be detected amperometrically without any derivatization or the use of pulse techniques. HI could be detected with a direct experimental detection limit of 500 nM (S/N = 13.8), corresponding to 10 pmol, and a response variability of less than 7%. A linear dynamic range from 0.5 to 100 μM was achieved. In the case of serotonin, an experimental detection limit of 10 nM (corresponding to 200 fmol)

(30) Cheng, F.-C.; Yang, L.-L.; Chang, F.-M., Chia, L.-G.; Kuo, J.-S. *J. Chromatogr.* **1992**, *582*, 19−27.
(31) Histamine Brochure, 1998 KMI Diagnostics, Inc., Minneapolis, MN.

was obtained, with a linear dynamic range from 0.01 to 50 μM. High sensitivities were also obtained for the serotonin metabolite 5-HIAA. These results demonstrate the analytical use of diamond electrodes for the determination of two amines that are known to experience complications with the use of GC electrodes. Further studies are in progress on the determination of these amines in real samples with diamond electrodes after first separating them by high-performance liquid chromatography. The advantage of the diamond electrode is that it is able to achieve highly stable and sensitive response with simple amperometric detection.

ACKNOWLEDGMENT

This research was supported by the Japan Society for the Promotion of Science (JSPS), Research for the Future Program, "Exploratory Research on Novel Artificial Materials and Substances for Next-Generation Industries".

Received for review August 3, 1999. Accepted January 24, 2000.

AC9908748