ADSORPTION OF DEOXYRIBONUCLEIC ACID AT MERCURY ELECTRODES FROM ALKALINE SOLUTIONS OF HIGHER IONIC STRENGTH

Viktor BRABEC[a], Viktor GLEZERS[b] and Valentina KADYSH[b]

[a] *Institute of Biophysics, Czechoslovak Academy of Sciences, 612 65 Brno and*
[b] *Institute of Organic Synthesis, Academy of Sciences of the Latvian SSR, 226006 Riga, USSR*

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Adsorption of double-stranded, thermally denatured DNAs and degradation products of DNA at mercury electrodes from alkaline solutions of higher ionic strength (0·5) has been studied with the aid of phase-sensitive alternating current (AC) and linear sweep voltammetry at the H.M.D.E. and phase-selective AC polarography (at the D.M.E.). It was found that the addition of DNA to the supporting electrolyte lowered the out-of-phase component of AC around the potential of zero charge. At potentials more negative than *ca.* −1·0 V nucleic acids yielded nonfaradaic peaks on voltammetric or polarographic curves. The model of adsorption of DNA at the mercury electrode from alkaline solution of higher ionic strength that fits best with the experimental data to date is proposed.

The behaviour of nucleic acids at mercury electrodes has been intensively investigated, particularly in the last decade. Among methods exploited in these studies alternating current (AC) polarography (tensammetry) has also been widely used. Since the pioneering work of Berg[1] a number of results have been obtained which have been interpreted particularly with the goal of contributing to an understanding of the properties of nucleic acids on their interaction with electrically charged surfaces.

AC polarographic measurements with nucleic acids can be performed in alkaline solutions when nucleic acids yield no faradaic current[2], so that the changes of AC correspond only to the changes of differential capacitance of the electrode double layer. Moreover, most of these experiments have been carried out with dropping mercury electrode (D.M.E) (drop time about 2 − 5 s) when a relatively slow direct current (DC) voltage scan rate was used (about 2 mV/s)(ref.[1,3,4]). The AC measured thus corresponded to the processes that took place on a D.M.E. charged only to the potential at which AC was measured. This means that the AC measured at a particular potential was not influenced by the history of the whole polarographic experiment, *i.e.* by the processes that took place at other potentials. Flemming was the first to use in these investigations a stationary hanging mercury drop electrode[5] (H.M.D.E.)

It has been shown[6-8] that nucleic acids are adsorbed on mercury electrodes, and that this adsorption may be at least partially irreversible. If, therefore, AC voltammetric (ACV) curve is recorded for DNA samples using one mercury drop, the AC is also influenced by interfacial processes taking place on the same drop at the potentials to which it has been polarized earlier.

ACV curves at the H.M.D.E. of some nucleic acids were different from those obtained at D.M.E. (AC polarograms)[5]. This difference was interpreted on the basis of the assumption that molecules of nucleic acids associate at the surface of mercury electrodes charged to more negative potentials (around -1.2 V). Later a number of papers were published aimed at describing the adsorption behaviour of DNA at the mercury electrode. However, many results of these studies were interpreted on the basis of a model according to which double-stranded (ds) DNA can be unwound at an electrode charged to more negative potentials[6,9] (around -1.2 V).

A different interpretation of the interfacial behaviour of nucleic acids at mercury electrodes was made on the basis of results obtained under different experimental conditions (particularly in different media and with samples of nucleic acids which might differ in their quality). The goal of this work was to perform further ACV, linear sweep voltammetric (LSV) and AC polarographic measurements with DNA which would make it possible to suggest a scheme of adsorption of DNA at a mercury electrode which would best comply with the experimental data obtained so far.

EXPERIMENTAL

Material and Methods

Calf thymus DNA was isolated and characterized as in our previous papers[2,6]. Thermal denaturation, sonication and measurements of DNA concentration were also carried out as described earlier[2,6,10]. Ds DNA was sonicated for 160 s, which led to a reduction of its molecular weight to the value of $2{\cdot}1 \cdot 10^5$. Apurinic acid was prepared from calf thymus DNA (ref.[11]). The pyrimidine bases were removed from apurinic acid in the same way as in our previous paper[3]. The electrochemical measurements were carried out in Britton–Robinson buffer, pH 9·85. The ionic strength of this buffer was adjusted by potassium chloride to a value of 0·5. Chemicals used for preparation of the supporting electrolyte were all of analytical grade.

Electrochemical measurements were carried out with a PAR Analyzer, Model 170. A three electrode system was used, including a working mercury electrode, platinum counter electrode and saturated calomel reference electrode. The working electrode was a Metrohm H.M.D.E., type E 410, with a surface area of $1{\cdot}77 \cdot 10^{-2}$ cm^2 or D.M.E. with a mercury flow rate of 0·644 mg/s. Phase sensitive ACV or AC polarographic measurements were carried out with a modulation voltage of 22 or 920 Hz and 0·013 V peak-to-peak. Phase angles of 0° and 90° with respect to the applied alternating voltage were employed (in-phase and out-of-phase component of the AC being measured, respectively). AC polarographic measurements (at the D.M.E.) were performed with the drop time control of the PAR Model 170 set at 1·0 s and with a DC voltage scan rate of 0·002 V/s. In this work AC was always measured in peak-to-peak values. Unless stated otherwise voltammetric measurements at the H.M.D.E. were carried out with a presweep delay

of 60 s at the initial potential (E_i). All measurements were made at 25°C. The solutions were deoxygenated by passing a slow stream of pure argon through them. The argon was allowed to pass over the solution during the measurements to maintain the inert atmosphere. The other details of our electrochemical measurements have been published elsewhere[3,6].

RESULTS

All electrochemical measurements described in this report were performed in a medium with pH 9·85. At this pH nucleic acids yield no faradaic current[2], so that all responses obtained in this work had a nonfaradaic character. Moreover, the results described in this report were obtained in medium of ionic strength 0·5. This means that DNA had negative charges of phosphate groups neutralized by cations of the background electrolyte so that it had, under the conditions of our measurements, the character of an electroneutral compounds.

AC *Voltammetry at the* H.M.D.E. *and* AC *Polarography* (*at the* D.M.E)

AC polarographic behaviour of our samples of DNA (Fig. 1) basically agreed with that observed earlier[1,3-5]. The out-of-phase component of AC was markedly lowered in the presence of ds and thermally denatured DNAs in the potential region −0·2 to −0·9 V. This lowering confirmed the adsorption of both forms of DNA in this potential region. Both forms of DNA yielded peak *1* at −1·12 V on out-of-phase AC polarograms. If the in-phase component of AC was recorded, both forms of DNA yielded peaks as in the case when the out-of-phase component of AC was measured. Only ds DNA yielded a still more negative peak 2 at −1·34 V if low frequency of the modulation AC voltage (22 Hz) was used.

AC voltammetric measurements carried out with thermally denatured DNA at the H.M.D.E (Figs 2*e*−*h*, 3*e*−*h*) yielded qualitatively identical curves as compared with AC polarograms (at the D.M.E.) (Fig. 1*e*−*h*). The results of AC voltammetric measurements were independent of E_i, DC voltage scan direction, frequency and phase angle. ACV curves of ds DNA were, however, more complicated. The course of ACV curves of ds DNA was strongly dependent on E_i (Fig. 4), DC voltage scan direction (Figs 2*a*−*d*, 3*a*−*d*), frequency and phase angle (Fig. 2*a*−*d*). If the DC voltage scan direction was negative and E_i more positive than c. −0·9 V ds DNA yielded on ACV curves at higher frequencies peaks *1* and *3* independently of phase angle; peak *2* was absent. On the other hand at lower frequencies all three peaks *1*, *2* and *3* were obtained on both in-phase and out-of-phase ACV curves. If, however, E_i more negative than −1·1 V was used, ds DNA yielded on ACV curves only peak *3*.

The heights of ACV peaks *2* and *3* of ds DNA were dependent on E_i as shown in Fig. 4. Peak 3 was only slightly dependent on E_i in the potential region −0·2 to −0·8 V (region *T*). If more negative E_i were used (region *U*) the height of peak *3* started to increase as E_i became more negative. After E_i had reached c. −1·25 V

the height of this peak started to fall with a further decrease of E_i. Peak 2, appearing only at low frequencies, was also slightly dependent on E_i in the potential region *T*, but disappeared completely if E_i more negative than $-1{\cdot}1$ V was used.

ACV peak 3 of thermally denatured DNA was practically independent of E_i for E_i ranging from $-0{\cdot}2$ to $-1{\cdot}4$ V. Only peaks 3 obtained for E_i around $-1{\cdot}2$ V were slightly higher (by c. 10%).

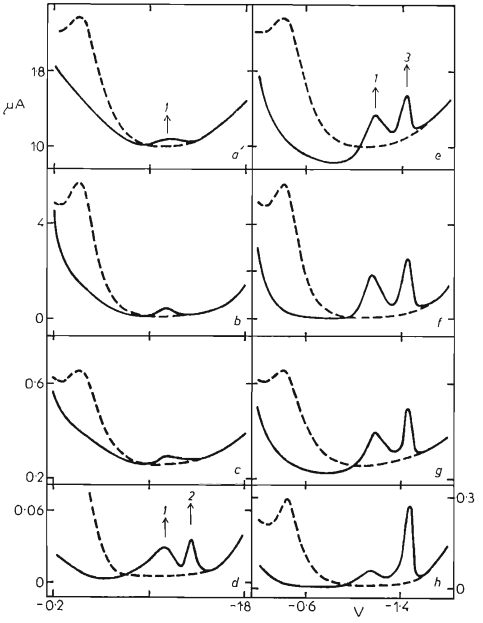

FIG. 1

AC polarograms ($I_\sim/E$ curves) of native DNA *a*—*d* and thermally denatured DNA e—*h*. DNAs at a concentration of 370 μg/ml. *a*, e out-of phase component of AC, 920 Hz; *b*, *f* in-phase component of AC, 920 Hz; *c*, *g* out-of-phase component of AC, 22 Hz; *d*, *h* in-phase component of AC, 22 Hz. (– – – – –) supporting electrolyte

The effect of some factors on the lowering of out of phase AC induced by DNA addition to the background solution (ΔAC) (Fig. 1) and on the height of out of phase ACV peaks *1* and *3* (h_{AC-1} and h_{AC-3}) (Fig. 2) was observed under conditions when the H.M.D.E. surface was not fully covered by adsorbed DNA molecules. It was found that ΔAC, h_{AC-1} and h_{AC-3} of ds and thermally denatured DNAs (peaks *1* and *3* were recorded with the H.M.D.E. charged to $E_i = -0{\cdot}6$ V and with DC

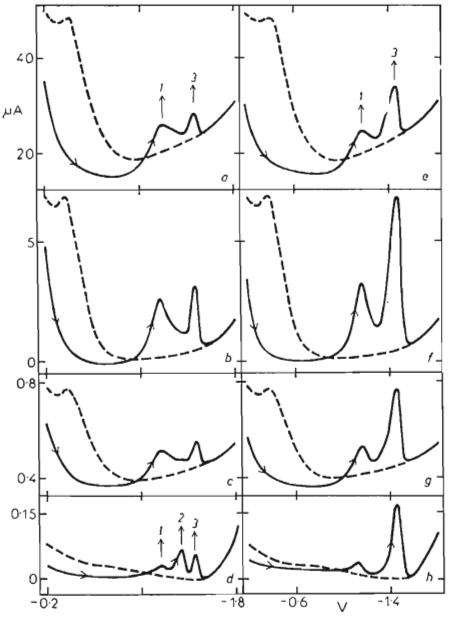

FIG. 2

AC voltammograms ($I_\sim/E$ curves) of native DNA *a*—*d* and thermally denatured DNA e—*h* at the H.M.D.E. DNAs at a concentration of 100 μg/ml. *a*, e out-of-phase component of AC, 920 Hz; *b*, *f* in-phase component of AC, 920 Hz; *c*, *g* out-of-phase component of AC, 22 Hz; *d*, *h* in-phase component of AC, 22 Hz. Initial potential (E_i) −0·2 V, waiting time at E_i 60 s; DC voltage scan rate 20 mV/s. DC voltage scan direction negative. (– – – –) supporting electrolyte

voltage scan rate of 20 mV/s) were directly proportional to the concentration of DNA in the bulk of solution and to the square root of the time for which the electrode (charged to E_i) was in the contact with DNA solution.

Contrary to denatured DNA ACV curves of ds DNA had a different course if positive DC voltage scan direction was used (Fig. 3*a*–*d*). In the range of E_i = = −1·4 to −1.7 V ds DNA yielded peak *1* and peak at −0·9 V, which was designated

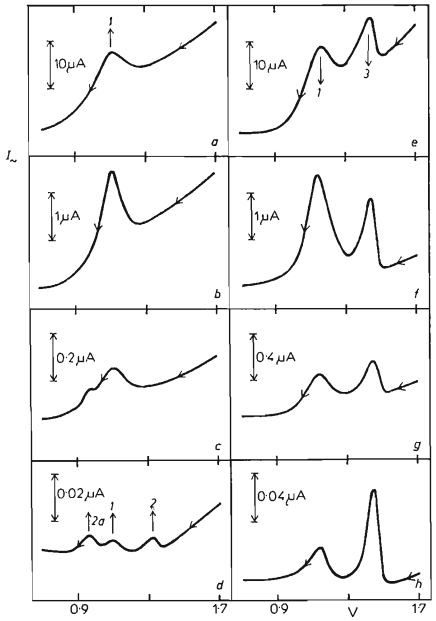

FIG. 3

AC voltammograms ($I_\sim$/—E curves) of native DNA *a*—*d* and thermally denatured DNA **e**—*h* at the H.M.D.E. *a*, *c* out-of-phase component of AC, 920 Hz; *b*, *f* in-phase component of AC, 920 Hz; *c*, *g* out-of-phase component of AC, 22 Hz; *d*, *h* in-phase component of AC, 22 Hz. Initial potential −1·7 V, DC voltage scan direction positive. Other conditions as in Fig. 2

peak *2a* (Fig. 3*d*). It was interesting that this peak *2a* was yielded only by ds DNA and at DC voltage scan rates higher than *ca.* 10 mV/s and E_i more negative than −1·4 V. At more positive E_i peak *2a* appeared only at very low concentrations of ds DNA or at short waiting times at E_i.

ACV curves at the H.M.D.E. were also recorded for sonicated ds and thermally denatured DNAs. These ACV curves had qualitatively the same course as those yielded by nonsonicated nucleic acids. All ACV peaks were, however, markedly higher than those yielded by nonsonicated samples.

The ACV measurements described in the preceding text were performed with the H.M.D.E. first kept for 60 s at E_i, the potential sweep only then being commenced. In earlier reports devoted to a similar subject so-called potentiostatic double step sweep method was also used[6–8]. If the latter technique was used in our work, DNA was first in contact with the H.M.D.E, charged to E_i for a period t_1 = 60 s, and then for another 60 s (t_2) with the same drop of mercury, charged to E_{i2}. Only after these two steps of prepolarization the negative DC voltage sweep was applied. If E_{i1} was −1·2 V and E_{i2} −0·4 V ds DNA yielded only ACV peaks *1* and *3*, peak *2* being totally absent. The height of this peak *3* was identical to that obtained with the potentiostatic single-step sweep technique if E_i was −1.2 V and waiting time at this E_i was 120 s. If the same experiments were carried out with thermally denatured DNA identical ACV curves were obtained independently of the selection of E_{i1} and E_{i2}. Thermally denatured DNA always yielded only ACV peaks *1* and *3*.

Linear Sweep Voltammetry

For the purpose of comparison LSV curves were also recorded for the same samples of nonsonicated DNAs. At slow voltage scan rates (comparable with those used for ACV measurements) ds DNA yielded three LSV peaks if negative voltage scan

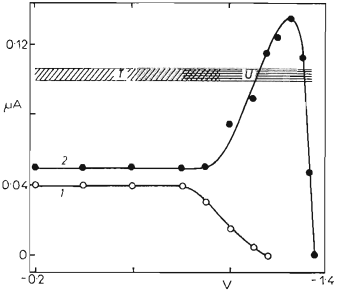

FIG. 4

Dependence of the heights ($i_\sim$) of in-phase AC voltammetric peaks *2* and *3* of native DNA at the H.M.D.E. on initial potential (E_i). The conditions of measurement were the same as in Fig. 2*d*. *1* peak *2*; *2* peak *3*

direction and E_i more positive than c. −0·9 V were used (Fig. 5*a*). These peaks corresponded, as regards their potentials, to the ACV peaks 1, 2 and 3 obtained at low AC voltage frequencies (Fig. 2*d*). As a matter of fact the LSV behaviour at slow scan rates of ds DNA and thermally denatured DNA was quantitatively identical to ACV behaviour at low frequencies (Fig. 2*d*,*h*), including E_i dependences and peak

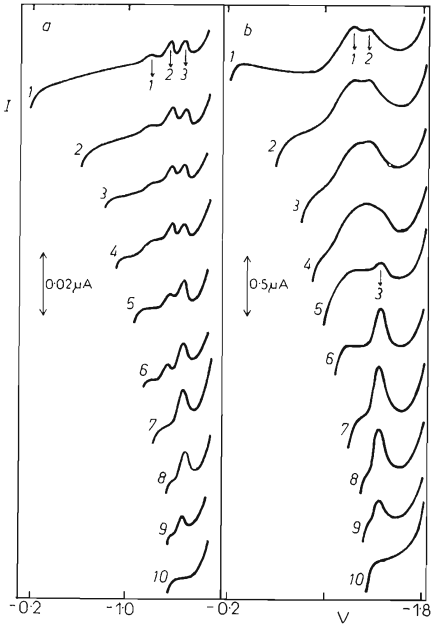

FIG. 5

DC linear sweep voltammograms (I/E curves) of native DNA at the H.M.D.E. Dependence on initial potential (E_i). *a* voltage scan rate 20 mV/s; *b* voltage scan rate 1·0 V/s. E_i values (V): *1* −0·2; *2* −0·6; *3* −0·8; *4* −0·9; *5* −1·0; *6* −1·1; *7* −1·2; *8* −1·3; *9* −1·325; *10* −1·35. Other conditions as in Fig. 2

2*a* appearance. If, however, the scan rate was increased (to a value c. 1·0 V/s) ds DNA did not yield LSV peak *3* unless E_i more negative than c. −0·8 V was used (Fig. 5*b*).

LSV curves of ds DNA obtained with $E_i = -0{\cdot}6$ V at various voltage scan rates (Fig. 6*a*) and at scan rate 1·0 V/s at various E_i (Fig. 5*b*) indicated that the process corresponding to the origin of LSV peak *3* took place in the potential region −0·8 to −1·35 V, and that ds DNA had to be in contact with the H.M.D.E. charged to the potentials of this region at least 1 − 2 s. The same conclusion followed from the LSV experiments in which potentiostatic triple step-sweep technique[8] was used (E_{i1} and E_{i3} were −0·6 V, $E_{i2} = -1{\cdot}2$ V, t_1 and t_3 were 60 s and t_2 was changed within 0·5 to 50 s). If t_2 shorter than c. 2 s was used ds DNA yielded only peaks *1* and 2, peak *3* being totally absent. Only t_2 longer than 2 s led to the appearance of peak *3*, which continued to increase with growing t_2. The peak 3 of ds DNA thus obtained reached a limiting height if t_2 of 30 − 40 s was used. Further increase of t_2 had no influence on the height of this peak *3*. Thus in the case of ds DNA t_2 about 30 s was needed to obtain the maximum yield of the interfacial reaction responsible for the origin of peak *3*.

Voltammetry of Degradation Products of DNA

Samples of apurinic acid (prepared from DNA) which was gradually released from the pyrimidine bases were subjected to LSV and ACV under conditions identical to those used for voltammetry of DNA. The decrease of the base content in apurinic acid resulted in a diminution of LSV and ACV peak *3*, whereas peak *1* hardly changed, even when the number of bases in apurinic acid was reduced by half; if the content of bases in apurinic acid was under 3%, LSV and ACV peak 3 disappeared while peak 1 was present.

DISCUSSION

Adsorption of DNA *on Mercury Electrode Charged to Potentials around that of Zero Charge*

All conclusions on the adsorption of DNA on mercury electrodes presented in this report concern only adsorption from an alkaline medium of higher ionic strength.

The models suggested up to now for adsorption of DNA at the mercury electrode almost agree, at least as regards the adsorption of electroneutral DNA in the potential range −0·2 − c. −0·9 V. It has been assumed for this adsorption that ds DNA is adsorbed particularly *via* sugar-phosphate residues and also *via* a few bases present in ds distorted regions[3,4,12]. The results described in this report (Fig. 5*b*) have revealed that even relatively long polarization (about 60 s) of the mercury electrode to these potentials does not lead in the case of ds DNA adsorbed from alkaline solutions

to a significant change in its structure. Thermally denatured DNA is thought to be adsorbed in the potential region *T* particularly *via* bases[3,4,6,13].

Adsorption of DNA *on Mercury Electrode Charged to the Potentials of Peak* 1

Several different explanations have been submitted for the origin of an ACV or AC polarographic peak *1* appearing around $-1{\cdot}1$ V. It has been suggested[5,12] that DNA molecules associate at the potentials of this peak and that the associated molecules are adsorbed on the mercury electrode more firmly than the isolated molecules. This interpretation derives from an assumption that even ds DNA is adsorbed on the mercury electrode *via* two-dimensional totally adsorbed sequences alternating with three dimensional loops protruding into the bulk of solution[5,12]. However, such a configuration can be hardly adopted by ds DNA molecules having a molecular weight around $2 \cdot 10^5$. It follows from hydrodynamical measurements[14] that such a short molecule of rigid ds DNA exists rather as a single rod. On the basis of this the reduction of molecular weight of ds DNA to $2 \cdot 10^5$ should have led to a marked reduction of the yield of associated molecules of DNA (monitored according to this model[5,12] by peak *3*). The sample of sonicated ds DNA, however, yielded an even higher ACV peak *3* as compared with nonsonicated high-molecular weight ds DNA, in which case it is possible to admit the existence of loops. Moreover, it also appears reasonable to assume that the yield of this association reaction would be lower if the possibility of association among segments belonging to different DNA molecules were lowered. This lowering was achieved by a marked reduction of surface concentration (by shortening the time of adsorption and by diminution of the DNA concentration in the bulk of solution). However, not even in this case was peak *3* lowered more than would correspond to the reduction of the surface concentration of DNA. For these reasons the possibility of the associates' origin on the mercury electrode surface appears, particularly in the case of ds DNA, to be less likely.

Later, on the basis of AC polarographic measurements, it was concluded[4] that ds DNA is not adsorbed *via* sugar phosphate residues at the potentials more negative than $-1{\cdot}0$ V and that the origin of peak *1* at $-1{\cdot}12$ V (Figs 1, 2 and 5) corresponds to reorientation of base units in DNA into a more perpendicular position with respect to the surface[13]. As follows from the results also presented in this report the removal of bases from DNA did not lead to disappearence of ACV or LSV peak *1*. This means that nucleic acids can be adsorbed *via* sugar-phosphate residues even at the potentials of peak *1*, *i.e.* even at potentials more negative than $-1{\cdot}0$ V. It is apparent that if more hydrophobic and thus more adsorbable bases in DNA are accessible for a reaction with the electrode, the nucleic acid molecules will be adsorbed preferentially *via* these accessible bases. It has been suggested[6,13] that at the potentials of peak *1* (in the region *U*) unwinding of DNA takes place, during which bases become accessible for the reaction with the electrode. The question of the participa-

tion of base and sugar-phosphate residues in DNA adsorption on an electrode charged to the potentials of peak *1* could be thus resolved in terms of the extent of ds DNA opening (as will be discussed below the extent of this opening may be monitored by means of peak *3*). As follows from Fig. 6 and results of the experiments in which triple step-sweep technique was used, only contact of ds DNA molecules (in alkaline medium) with the electrode charged to the potentials of peak *1* lasting longer than c. 2 s led to the origin of peak *3*, and thus apparently to the opening of ds DNA molecules to a very small extent. If this contact was prolonged up to 30–40 s peak

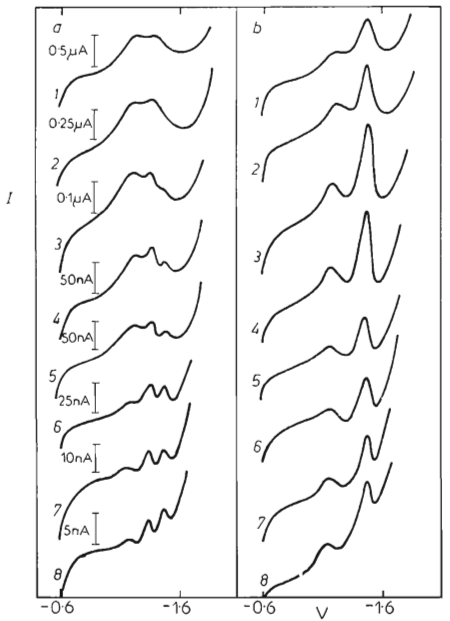

FIG. 6

DC linear sweep voltammograms (I/E curves) of native DNA *a* and thermally denatured DNA *b* at the H.M.D.E. Dependence on voltage scan rate (v, V/s): *1* 2·0; *2* 1·0; *3* 0·5; *4* 0·2; *5* 0·1; *6* 0·05; *7* 0·02; *8* 0·01. Initial potential −0·6 V. Other conditions as in Fig. 2

3 markedly increased, which could indicate that this opening hit a more significant part of adsorbed ds DNA molecules. It is thus apparent that in alkaline medium under AC polarographic conditions (when D.M.E. had a lifetime of, for instance, only 3–5 s) the molecules of ds DNA were in contact with the electrode for a very short time. During this short period only a very small number of base pairs could be opened. Thus an interfacial reaction at the D.M.E. was responsible for the appearance of the AC polarographic peak *1* in which adsorbed sugar-phosphate residues mostly participated. As for the ACV or LSV peak *1* or ds DNA observed at the H. M.D.E. for $E_i = -0{\cdot}2$ to $-0{\cdot}8$ V, the number of bases and sugar-phosphate residues adsorbed at the potentials of peak *1* depended particularly on voltage scan rate. At slow scan rates relatively long contact of ds DNA molecules with the electrode charged to the potentials of peak *1* was permitted so that the number of bases participating in adsorption at these potentials increased. On the other hand at very fast scan rates used in some LSV measurements (Fig. 5*b*, 6*a*) it appears probable that LSV peak *1* of ds DNA in alkaline medium is connected particularly with an interfacial reaction of adsorbed sugar-phosphate residues. The results of ACV and AC polarographic investigations of degradation products of DNA also indicate that sugar-phosphate residues cannot be adsorbed at potentials more negative than those of the negative foot of peak *1*. It thus seems probable that on the level of peak 1 it is particularly those ds DNA segments adsorbed *via* sugar-phosphate residues which are desorbed.

The conception of participation of sugar-phosphate residues in the interfacial reaction at the potentials of peak *1* might seem to be in contradiction with AC polarographic or ACV behaviour of thermally denatured DNA or of some other ss polynucleotides, which also yield peak *1*. As has been shown earlier[3] the origin of this peak of ss polynucleotides corresponds to the presence of helical regions in the samples of ss polynucleotides. The interfacial reactions of these helical segments in alkaline medium have been suggested[3] to proceed in an similar way as in the case of ds segments of DNA.

Adsorption of ds DNA *on Mercury Electrode Charged to the Potentials of Peak* 2

It has been suggested that the origin of AC polarographic or ACV peak 2 corresponds to desorption of ds segments of DNA, which were, at the potentials of this peak, adsorbed *via* bases present in ds distorted regions of DNA[3,4]. The interpretation of Nürnberg and coworkers, that the origin of this peak corresponds under LSV conditions to a reorientation of adsorbed double-helix[13], need not contradict that outlined in this report. This reorientation could be understood in such a way that ds DNA under voltammetric conditions at the H.M.D.E, at slow scan rate and at more negative potentials than those of peak 2 would be adsorbed only through ss segments which originated in it at the potentials of peak *1*. On the level of LSV or ASV peak 2 only ds distorted segments of DNA adsorbed only *via* bases would desorb, while

ss segments (if any) also adsorbed *via* bases would remain anchored on the electrode. The peak 2 appears only at low frequencies, which could indicate that the adsorption/desorption process taking place on the level of peak 2 is relatively slow.

Adsorption of DNA *on Mercury Electrode Charged to the Potentials of Peak 3*

It has been suggested[5,12] that this peak corresponds to the desorption of associates of DNA which originate at an electrode charged to the potentials of peak *1*. As also discussed in this report, the origin of these associates is unlikely.

All voltammetric or polarographic results obtained so far, particularly those obtained with degradation products of DNA, are explicable on the basis of the model which ascribes the origin of peak *3* to the adsorption/desorption process in which ss segments of DNA adsorbable *via* bases participate. The fact that ds DNA also yielded ACV peak *3* when a voltammogram was recorded with the aid of one drop of mercury (Figs 2*a*, 4*a*) can be explained by the origin of ss material as a consequence of the appreciable contact of ds DNA with the electrode charged to the potentials of the region U.

Quite recently it has been suggested[15] that base pairs in ds DNA adsorbed on the electrode can be temporarily opened. This process was called base fluctuation and was thought to be amplified at more negative potentials so that more bases would become accessible for reaction with the electrode charged to these potentials. Our results with the potentiostatic double-step or triple-step method indicate that bases that are released from the adsorbed double-helix can remain anchored irreversibly on the electrode even when the factors responsible for the release of bases at more negative potentials cease to be effective.

Adsorption of ds DNA *on the Mercury Electrode under Conditions of Peak 2a Appearance*

As follows from the results of this work peak *2a*, first reported by Flemming and Pospíšil[16], is yielded only by ds DNA. Further, this peak appears only if the surface concentration of ds DNA is very low, and when the adsorbed molecules of ds DNA are in the contact with an electrode charged to the potentials of peak *1* for a relatively short time (~ 10 s). If the opening reaction with ds DNA is assumed to take place on the level of peak *1* it is apparent that this opening, under conditions of peak *2a* appearance, hits only a part of the ds DNA molecule, *i.e.* ds DNA would become only partially denatured on the electrode surface. Owing to the fact that partial denaturation of DNA may be a reversible process[17,18] it is possible to suggest that peak *2a* might correspond to such a rearrangement of the adsorbed ds DNA molecules that it would result in renaturation of some DNA segments partially denatured on the level of peak *1*. This surface renaturation might take place due to the

fact that the action of factors responsible for DNA opening at negative potentials is already not so effective at the potentials of peak *2a* as compared with those acting on the level of peak *1*. The fact that ACV peak *2a* appears only at low frequencies could indicate that this surface renaturation would be a slow process.

CONCLUSIONS

On the basis of the experimental results obtained so far the model of adsorption of DNA from alkaline solutions of higher ionic strength at the mercury electrode is suggested, whose main features are the following.

Linear ds DNA is adsorbed in the potential range of −0·2 to −0·8 V particularly *via* sugar-phosphate residues and a few bases present in ds distorted regions. The ds segments of DNA adsorbed *via* sugar-phosphate residues may desorb on the level of peak *1* while the ds segments adsorbed through bases present in ds distorted regions of DNA may desorb on the level of more negative peak *2*. The participation of further bases in adsorption of ds DNA may result from longer contact of adsorbed ds DNA molecules with an electrode charged to potentials around −1·1 V. At these potentials an interfacial reaction can takes place, due to which an irreversible opening of adsorbed ds DNA segments may appear. This interfacial reaction leads to the origin of DNA conformation which has interfacial properties identical to those of thermally denatured DNA. These DNA segments desorb from the mercury electrode on the level of most negative peak *3*. The origin of ds DNA peak *2a*, which is well developed when positive DC voltage scan direction is used, is ascribed to a slow rearrangement of adsorption layer. This rearrangement might result in renaturation of DNA segments partially denatured on the level of peak *1*.

Thermally denatured DNA is adsorbed particulatly *via* bases. The single-stranded segments of DNA were thought to yield most negative AC voltammetric or polarographic adsorption/desorption peak *3*. The peak *1* of thermally denatured DNA corresponds to an interfacial reaction of helical or ds segments present in the denatured DNA sample.

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