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METHODS FOR INCREASING THE SENSITIVITY, ACCURACY, AND SELECTIVENESS OF MEASUREMENTS IN DIFFERENTIAL CONDUCTOMETRIC SYSTEMS

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This paper summarizes approaches to solving problems limiting the sensitivity, accuracy, and selectivity of conductometric measurements. It presents the results of extensive research and development aimed at creating portable, affordable electrochemical analyzers with metrological characteristics that meet a wide range of practical needs in conductometry. The main factors that reduce the sensitivity of the measuring channel and cause measurement errors are discussed, as well as differential measurement methods that can eliminate or significantly reduce their impact. These results were obtained during the development of biosensor systems, but can also be applied to other tasks requiring rapid and accurate determination of changes in the electrical conductivity of solutions. The results of theoretical and experimental studies of biosensor conductometric transducers and measuring circuits for their use are presented. A new approach to further increasing the selectivity of differential sensory systems has been substantiated
References 22, tables 2, figures 5.

Keywords: conductometry, biosensors, differential measurements, sensitivity, common-mode interference.

Introduction. Conductometry – the determination of the electrical conductivity of electrolyte solutions – is widely used in many technological processes to monitor the composition of the working environment, raw materials, and finished products [1–5]. In the energy, pharmaceutical, and other chemical industries, as well as in the food industry, such measurements are used in the preparation of process water. In biomedical, environmental, and other laboratory studies, conductometric instruments are used in conjunction with selective electrochemical converters, in particular, with biosensors for the selective quantitative analysis of solution composition [6–13]. In all cases, strict requirements are imposed on the sensitivity and accuracy of the measuring instruments and, especially, on their selectivity with respect to uninformative influencing factors. This is connected, on the one hand, with the responsibility of this type of measurement, in particular, when used for medical diagnostics and quality control of food products [14–16]. On the other hand, this is also connected with the structure and operating features of the measuring channel, which contains conductometric primary converters (sensors) that are immersed in an electrolyte solution located in the measuring cell [17–20].

Conductometry is based on measuring the informative parameter of the signal at the sensor output (current I, voltage U, or their changes ΔI, ΔU), to which an alternating current test voltage is applied. The impedance of the such transducer is represented by a complex equivalent circuit [1–4]. In addition to the main, informative parameter – the active resistance of the solution R_s (or its electrical conductivity G_s) between the electrodes – it contains non-informative parameters. These parameters (inter-electrode capacitance C_g, double-layer capacitance C_{dl}, charge-transfer resistance R_{ct}, Warburg impedance W) are unstable and can significantly alter the sensor's conversion results. This also occurs when the initial

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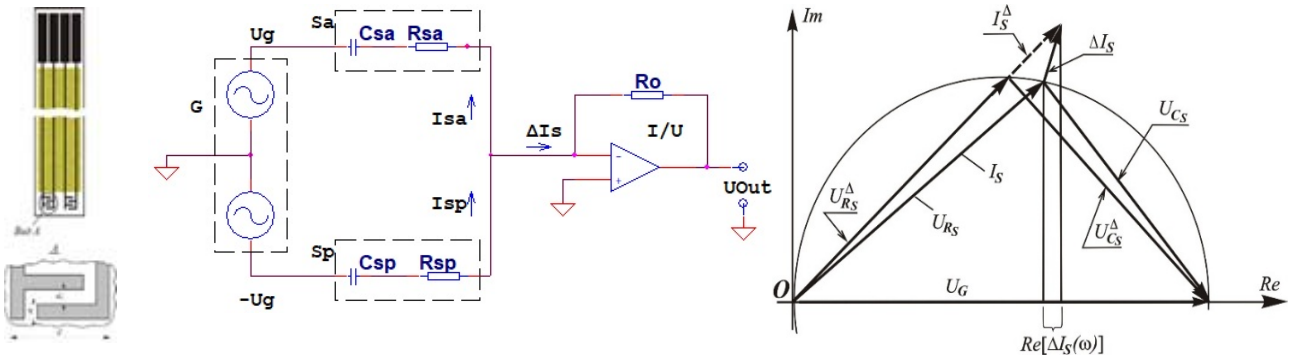
(background) parameters of the medium (solution composition, its temperature) change during the measurement process. Therefore, despite the positive qualities of the conductometric method, many specialists doubt its prospects for obtaining reliable results in complex, selective measurements over a wide range of determined quantities. In particular, conductometric biosensor systems have long been unused in practice due to their sensitivity to factors that alter the background conductivity of the solution in the conductometric cell. The purpose of this paper is to familiarize specialists with the possibilities for significantly reducing these problems, the results obtained by the authors in this area, and the prospects for further development and justification of a new approach to further increasing the selectivity of differential sensory systems.

Application of the differential measurement method. The influence of changes in environmental parameters in the conductometric cell was successfully reduced using the differential conductometric method with two planar two-electrode transducers (sensors) with an interdigital topology (Fig. 1, *a*), which are fabricated on a ceramic substrate using thin-film technology [11, 12]. Based on this approach, efficient conductometric biosensor systems were created [3, 5, 6, 10, 13].

The measuring channel contains a 4-arm AC bridge, two arms of which include a differential pair of conductometric transducers *S1* and *S2*, placed in a measuring cell with a buffer solution, and the other two arms include identical resistors [3]. Another, simpler version of the bridge circuit can be implemented as a current comparison circuit (Fig. 1, *b*). A selective biosensor membrane is applied to one of the transducers (the working transducer, S_a). This membrane generates charge carriers upon contact with the target analyte, causing a local change in electrical conductivity proportional to the analyte concentration. The other transducer is the reference transducer (S_p); it is located in the region of background electrical conductivity of the solution. The bridge is powered by a test alternating voltage generator G with two antiphase outputs U_g and $-U_g$. Its output signal U_{out} is received from the bridge output using an operational amplifier. From this signal, an informative component (in phase with the generator signal) is extracted using a synchronous detector and measured with a microvoltmeter [3, 20]. The value of this component is proportional to the magnitude of the difference in the active resistances of the solution R_{sa} and R_{sp} in the differential pair of conductometric transducers. Before the analyte is introduced, this difference is virtually nonexistent, and the other impedance components of the conductometric transducers and the bridge resistors are identical. Therefore, the bridge circuit is in a balanced state and its output signal is close to zero. When the working transducer comes into contact with the analyte, its active resistance changes (typically by about 1%), and a corresponding response appears at the bridge output [21].



a ***b*** ***c***

Fig. 1

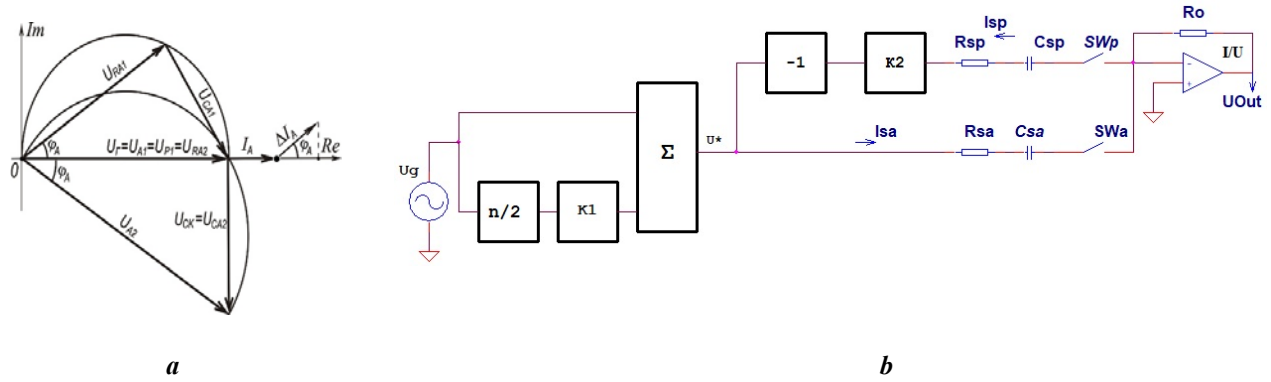
The described solution is implemented quite simply; however, it allows obtaining reliable results across a wide range of research tasks only with very small differences (no more than 1–2%) of the equivalent parameters R_s and C_s of a series two-element equivalent circuit in a differential pair of transducers. This is difficult to ensure during manufacturing and virtually impossible to maintain during operation of the sensors [22]. If the parameters R_s and C_s are not identical due to differences in the values of C_g, C_{dl}, R_{ct}, or W, then the sensitivities of the working and reference transducers (changes in current ΔI_s in them with identical changes in the solution resistance R_{sa} or R_{sp}) are different, and the effectiveness of the differential method is reduced. With a more significant difference in the values of these transducer parameters (up to 10%), sufficiently reliable results can only be obtained when studying analytes that do not change the background

conductivity of the buffer solution of the conductometric cell. Its change gives rise to an additive error proportional to this change and the relative difference in R_s and C_s.

In addition to the problem of the invariance of measurement results to changes in the background electrical conductivity in the conductometric cell, there is also the question of the dependence of the informative signal on the magnitude of the phase angle tangent tgφ of the conductometric transducers. This issue (for a bridge circuit with current comparison, similar in characteristics to a 4-arm bridge) is illustrated in Fig. 1, *b*, *c* [21]. The increment vector of the bridge output signal has a phase angle of 2φ, therefore the informative signal at the output of the synchronous detector of the conductometric channel **Re**(ΔI_s) drops sharply, (down to 0 at tgφ = 1). This is a systematic multiplicative error, which also limits the permissible parameters of the transducers and requires appropriate correction of the conversion result to stabilize the sensitivity and obtain an accurate measurement result in units of electrical conductivity.

Improving differential conductometric measurements by compensating for voltage drops across the near-electrode capacitances of sensors. The first developments in this area [22] were based on the use of a fully balanced bridge circuit or a bridge balanced by one (quadrature to the informative) parameter of the C_s transducers, which is determined primarily by C_{dl}.

The vector diagram in Fig. 2, *a* and the circuit in Fig. 2, *b*, illustrates this approach. In the first variant (with full balancing of the bridge), it consists of adding to the test signals U_g from the generator *G*, which are fed to the reference (passive) and working (active) converters (*S*2 and *S*1, respectively), a component U_{ck} that is quadrature to U_g and is regulated by the code *K*1, using the adder Σ. If the voltage U_{ck} becomes equal to the voltage U_{ca} (Fig. 2, *a*) across the capacitance of the working (active) transducer, it compensates U_{ca}. In this case, the voltage across R_{s1} is equal to the voltage U_g, and the current vector I_a rotates by an angle φ and coincides with U_g in phase. The in-phase and quadrature reference voltages of the synchronous detector (the **Re** and **Im** axes in Fig. 2, *a*) are also related to U_g. The resulting state (the bridge equilibrium point for the reactive component of the working converter impedance) is determined by the zero value of the **Im**(I_a) component when the reference converter is turned off by switch *Sw*2. In this case, the phase angle of the informative current increment ΔI_a in the converter decreases to φ.



a ***b***

Fig. 2

The sensitivity of the bridge at the synchronous detector output to changes in R_s is determined by the expression $S = \frac{1}{1+\mathrm{tg}^2\,\varphi}$ and can be well stabilized over a wide range of φ values. Bridge balancing for the active component of the converter impedance is achieved by varying the voltage amplitude on the reference converter *S*2 using the *K*2 code (based on DAC) adjustor until the common-mode component of the converter current difference, **Re**(I_{s1}–I_{s2}), is close to zero. After this, the change in this component's value will correspond to the change in conductivity in the differential sensor's working converter, regardless of the background conductivity values in the conductivity cell. At the bridge output, a certain component of the converters current difference, quadrature to U_g, remains in the presence of a difference in their phase angles due to incomplete compensation of the voltage drop across the reference converter's capacitance. This component does not affect the informative signal at the synchronous detector's output.

Research during the experimental operation of this version of the differential conductometer showed that the suppression of the influence of changes in the background electrical conductivity in the cell is worse in the case of differences in the active resistances of the converters (R_{s1} and R_{s2} in Fig. 2, *b*), compared with the

same relative difference in their capacitances C_s). This is because, even with equal currents I_{s1} and I_{s2} at bridge equilibrium, the voltages across the solution resistances R_{s1} and R_{s2} will differ when thes resistances are differ. Since these voltages determine the current increments in the transducers due to changes in background electrical conductivity, such increments will cause an error additive to the informative signal. To reduce this error, it is necessary to achieve equal voltages across the active resistances of the transducers even when their resistances and capacitances are differ. For this purpose, a second design of the differential conductivity meter was developed, with full compensation for voltage drops across the capacitances of both transducers.

The functional diagram of this design of the conductivity meter is shown in Fig. 3. Its difference from the circuit in Fig. 2, *b* is the presence of a second adder Σ2 on the base DAC 2, with code *K*2 regulation, providing small-scale regulation of the quadrature-to-U_g component of the test voltage on the reference transducer. This allows for precise compensation of the voltage drops across the capacitances of both transducers, obtaining identical voltages equal to U_g across the solution resistances in them, and significantly increasing the suppression of the influence of changes in background electrical conductivity [23]. With this balancing of the bridge, a small imbalance signal remains at its output of the in-phase component to U_g, which is the initial offset of the informative signal. However, this can easily be taken into account by simply offsetting the measurement results array.

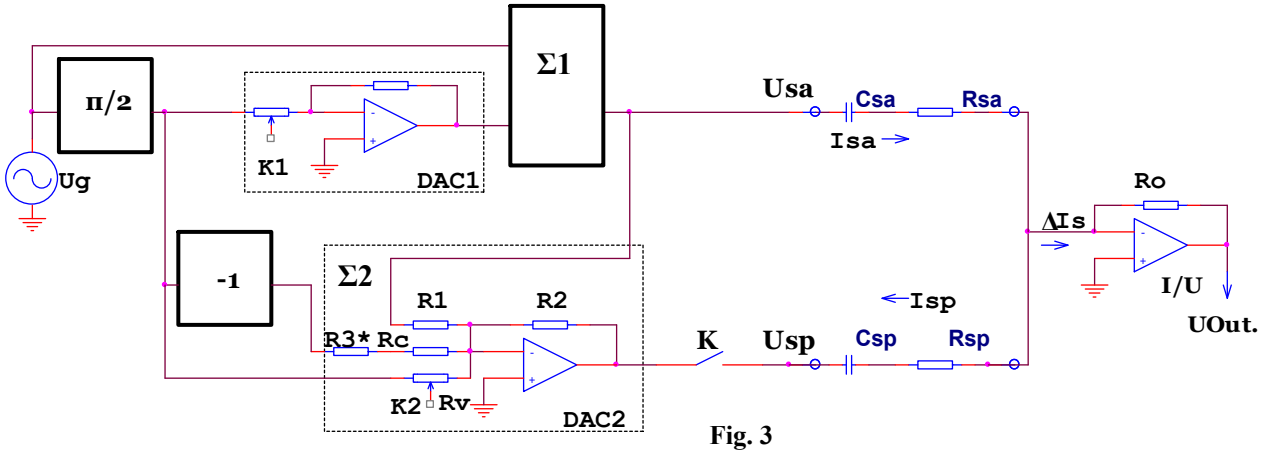


Fig. 3

The approach to improving differential conductometric measure-ments described above made it possible to effectively stabilize the sensitivity and normalize the output signal of the measuring circuit using fairly simple technical means, even with signi-ficant differences in the parameters of the transducers in the differential pair. The accuracy and noise immunity of measurements with this approach are quite adequate for studying analytes with low intrinsic conductivity (e.g., saccharides). A remaining problem is the significant additive error that arises when the intrinsic conductivity is comparable to or exceeds the informative increase in the solution's electrical conductivity upon introduction of the analyte into the measuring cell. When these conductivities are equal, this error is approximately 10% for a 20% difference in one of the parameters in a pair of transducers.

Differential conductometric measurements with profound suppression of background changes in the solution. To overcome the above problem, an approach was used to bring the bridge to a special quasi-equilibrium state, which is discussed below. A method for differential measurements in a quasi-equilibrium bridge circuit was developed in [24, 25], which allows for the theoretical complete suppression of errors due to changes in background conductivity in the conductivity cell. It consists of the following: Before adding the analyte to the buffer solution, the bridge is balanced by adjusting the current in the reference transducer in magnitude and phase angle to a minimum bridge output current value $\Delta\dot{I}_s$. The phase rotation of the adjusted current is $\Delta\varphi = \varphi_a - \varphi_p$. To bring the bridge to a quasi-equilibrium state, the phase of the current in the reference transducer is additionally adjusted by the same amount. In this case, the current $\Delta\dot{I}_s$ is not minimal. However, the changes in the currents of the transducers due to background changes in the conductivity of the solution in the cell become equal in magnitude and opposite in phase. Obviously, to reduce the error from background influences to the noise level, a very small relative non-identity of the converter gains in the differential circuit is required. Considering the differences in their parameters and the balancing accuracy of these differences, as well as the fact that in practice background influences can many times exceed the

informative values [23], this non-identity should not significantly exceed the sampling resolution of the informative parameter (approximately 0.01%). Achieving such bridge balancing accuracy using the analog converters used in the above-mentioned developments is technically impossible. Therefore, a bridge circuit with digital-to-analog converters of a constant reference voltage into quasi-sinusoidal test signals for the working and reference converters was developed [24] (Fig. 4).

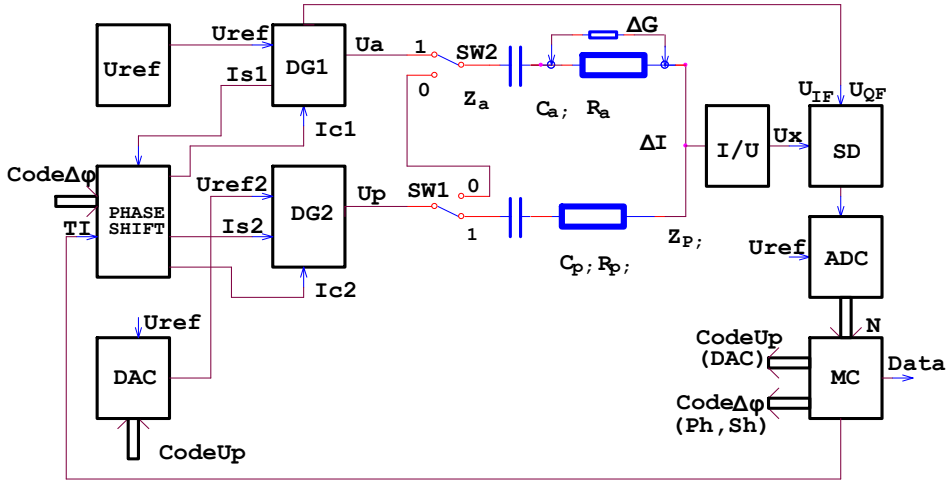


Fig. 4

The amplitude and phase relationships of these signals are controlled by digital codes, allowing the bridge to be balanced in magnitude and phase of the output signal, as well as to be precisely set to other calculated states, taking into account the differences in the parameters of the equivalent circuits of the conductometric transducers. Based on the results of the conducted research, a hardware and software complex of the differential conductometer for biosensor analyzers was developed, which is described in detail in [25]. The results of its metrological studies on the issue under consideration, the remaining problems, and possible solutions are discussed below.

The vector diagram of the bridge circuit currents in the quasi-equilibrium region is shown below in Fig. 5. In the described device structure, voltage compensation across the transducer capacitances (C_{dl}) is not performed; therefore, the current vectors in the transducers are located at an angle φ to the horizontal coordinate axis, and the phase angles of the current increments in them have values of 2φ (Fig. 1, *b*). This increases sensitivity variations with changes in φ and limits the maximum tan φ value of the transducers to 0.5–0.6.

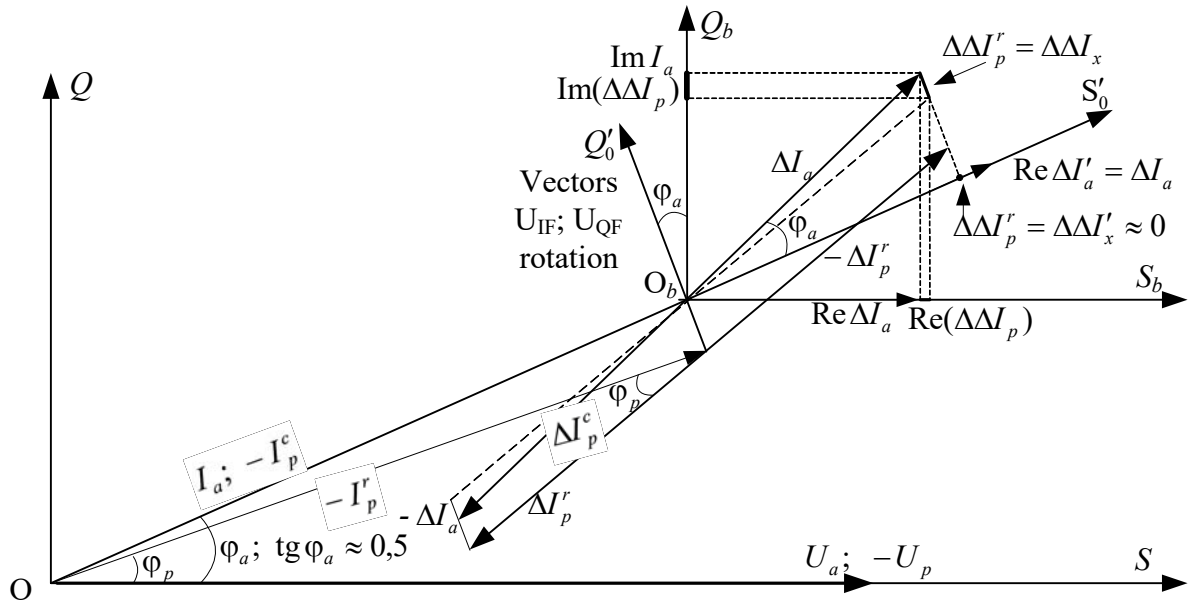


Fig. 5

The results of computer modeling of the error δ from changes in the background conductivity of the solution in the conductometric cell for the developed method of balancing a bridge circuit over a wide range of possible values are presented in Table 1. They demonstrate that, using the proposed method, almost complete (up to 1000-fold) suppression of background influences is theoretically possible.

Table 1

№	G_{AS}, мСм	C_{AS}, нФ	tgφ$_A$	G_{RS}, мСм	C_{RS}, нФ	tgφ$_R$	δ,%
1	1	5,44	0,4681	1	4,352	0,5851	0,08
2	5	40	0,3183	5	48	0,26526	0,04
3	5	1	12,733	4,167	1	10,61	0,017
4	0,2	1	0,5093	0,25	1	0,6366	0,11
5	1	4,5	0,5659	1	5,4	0,4716	0,066
6	1	5,44	0,4681	1,2	6,8	0,4496	0,01

Table 2 contains the values of the measuring channel responses (in ADC codes) to common-mode interference $\Delta(\Delta R_{ap}=1\%)$ and to the informative response (to ΔR_a = +1%). These data were obtained using a prototype conductometric analyzer with a quasi-equilibrium bridge circuit and an electrical equivalent of a differential conductometric sensor at maximum permissible deviations of one of the parameters of a pair of transducers. The last column contains the relative values of the obtained additive error $\delta = \Delta(\Delta R_{ap}/\Delta R_a)$. The data presented here showed a significantly lower suppression of the effect of background electrical conductivity compared to the computer model. The reason for this is the limited discreteness of the phase angle regulation (0.7°) at operating frequencies (more than 50 kHz), which, in turn, is limited by the clock frequency of the module microcontroller (32 MHz) and the speed of the digital microcircuits in the circuit in Fig. 4.

Table 2

Sensor parameter	R_a,Ohm	R_p,Ohm	R_p/R_a	tgφ$_a$	tgφ$_p$	$\Delta(\Delta R_{ap}=1\%)$	ΔG_a=+1%	δ_{add}, %
C_p=0.8C_a; R_p=R_a	1009	1010	1,001	0,440	0,525	18	1530	1,17
C_p=1,2C_a; R_p=R_a	1008	1008	1,000	0,440	0,351	12	1530	0,78
R_p=0.8R_a; C_p=C_a	1008	846	0,839	0,440	0,527	30	1519	1,89
R_a=0.8R_p; Cp=C_a	846	1010	1,1939	0,5268	0,4400	25	1680	1,49

Insufficient discreteness of establishing quasi-equilibrium by phase angle leads to deviations of the actual state of the bridge circuit from the calculated quasi-equilibrium state by this parameter. This problem and its possible solution are illustrated by the vector diagram of currents at bridge quasi-equilibrium in Fig. 5.

It shows the positions of the converter current vectors: the operating I_a and the reference I_p (inverted to enable comparison with I_a). The calculated position $-I_p^c$ coincides with I_a, and the actual one, after establishing quasi-equilibrium, is shown by the dashed line ($-I_p^r$) tgφ = 0.5. The possible *difference in their* angles (0.7°) is conditionally exaggerated. The increments of the converter currents ΔI_a and ΔI_p (they are at an angle φ to the vectors I_a and I_p) are also greatly exaggerated; in reality, their moduli are approximately 1% of I_a and I_p. After balancing the bridge, the origins of the increment vectors is moved toward the center of the coordinate system (the Q_b and S_b axes). In this coordinate system, if there were no difference in the angles between I_a and $-I_p^c$, the sum of the ΔI_a and $-\Delta I_p^c$ vectors would be zero. In reality, due to the difference in the phase angles ΔI_a and $-\Delta I_p^c$, a vector representing the difference between these currents, $\Delta\Delta I_p^r$, appears. When processing the signal in the synchronous detector, this vector yields a projection onto the S_b axis of **Re**($\Delta\Delta I_p$), which is a manifestation of the influence of changes in background conductivity.

This problem can be solved by the new approach with rotating the reference voltages U_{IF} and U_{QF} of the synchronous detector SD by an angle of φ. This causes the coordinate system axes to move to the Q_0^1 and S_0^1 positions. In this case, the phase angle inaccuracy of the regulated current vector in the reference converter will not affect the magnitude of the common-mode component of the bridge circuit output current Re(ΔI_{a1}). Furthermore, the angle between the S_0^1 axis and the ΔI_a vector is reduced by a factor of 2 to φ, which significantly increases and stabilizes measurement sensitivity. Thus, this solution combines the advantages of methods for compensating for voltage drops across converter capacitances and measurements with a quasi-equilibrium bridge circuit. The expected additional improvement in suppression of the influence of changes in background electrical conductivity is at least twofold. However, it should be kept in mind that this will require duplication of the Phase Shift and partially DG2 blocks in the circuit in Fig. 2, i.e., a certain complication of the device, its operating algorithm, and software.

Conclusion. Using differential measurements with a pair of planar two-electrode conductometric transducers and an AC bridge circuit enables direct, simultaneous comparison of solution conductivities in two localized regions of the measuring cell and the accurate determination of very small local changes, while

eliminating the influence of background instability on the results. This approach potentially enables rapid and highly sensitive analysis of solution composition using simple techniques and inexpensive measurement instruments, including selective analysis using chemosensors and biosensors. However, full realization of these advantages is limited by the complexity of the equivalent circuit of the conductometric transducers and the significant influence of its unstable, uninformative parameters on the measurement channel's conversion function.

The approach to performing measurements, employing bridge circuit balancing by compensating for voltage drops across the capacitances of conductometric transducers, developed during research, enable high-level stabilization of instrument sensitivity, regardless of the values of the informative and non-informative parameters of the equivalent circuit of the working conductometric transducer, over a fairly wide range of solution conductivity.

The measurement method, which places the conductometric bridge in a special state of quasi-equilibrium, deeply suppressing the influence of background changes on its output informative signal, has enabled significant invariance of measurement results to variations in the equivalent circuit parameters of differential sensor transducers.

A new approach to building a differential analyzer is proposed, consisting in a combination of measurement methods with a quasi-equilibrium bridge and compensation of voltage drops on the capacitances of conductometric converters. It will make possible to reduce the impact of changes in background conductivity, which have level of informative signals, to the level of resolution of the measuring channel.

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МЕТОДИ ПІДВИЩЕННЯ ЧУТЛИВОСТІ, ТОЧНОСТІ І СЕЛЕКТИВНОСТІ ВИМІРЮВАНЬ У ДИФЕРЕНЦІЙНИХ КОНДУКТОМЕТРИЧНИХ СИСТЕМАХ

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У роботі узагальнено підходи до вирішення проблем, що обмежують чутливість, точність та селективність кондуктометричних вимірювань. Наведено результати тривалих досліджень та розробок, спрямованих на створення портативних та недорогих електрохімічних аналізаторів з метрологічними характеристиками, що задовольняють широке коло практичних потреб у галузі кондуктометрії. Розглянуто основні фактори, що знижують чутливість вимірювального каналу та викликають похибки вимірювань, та диференціальні методи вимірювання, які дають можливість усунути або суттєво зменшити їхній вплив. Ці результати отримані у процесі розробок біосенсорних систем, але можуть бути застосовані й для інших завдань, де необхідні швидкі та точні визначення змін електропровідності розчинів. Наведено результати теоретичних та експериментальних досліджень кондуктометричних перетворювачів біосенсорів та вимірювальних ланцюгів для їхнього використання. Бібл. 25, табл. 2, рис. 5.

Ключові слова: кондуктометрія, біосенсори, диференціальні виміри, чутливість, синфазні перешкоди.

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