A study of the stability and radiochemical purity of some radiopharmaceuticals. 2. Labelled Chlormerodrin.

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SUMMARY

The chromatographic behaviour of chlormerodrin and its radiochemical impurities was studied in various solvents. The main product of thermal decomposition of chlormerodrin was identified as 3-chloromercuri-2-hydroxypropylurea. The thermal decomposition of chlormerodrin is accelerated by the presence of theophylline and suppressed by sodium chloride. The main product of radiation decomposition of chlormerodrin is inorganic mercury. The yield of radiation decomposition was studied as a function of the concentration of chlormerodrin. The radiation decomposition of chlormerodrin is not essentially influenced by the presence of some other substances, however these substances can under certain circumstances support the formation of 3-chloromercuri-2-hydroxypropylurea as the product of the radiolysis.

INTRODUCTION.

Chlormerodrin (Neohydrin), i.e. 3-chloromercuri-2-methoxypropylurea (I) has been used as mercurial diuretic. In last years this compound labelled with radioactive mercury isotopes ^{197}Hg or ^{203}Hg has been used in kidney scanning and for localization of brain tumors [1-3].

The labelled chlormerodrin has been prepared [1, 2, 4-7] by slight modifications of the procedure applied for unlabelled compounds i.e. by addition of mercuric acetate [8] or chloride [9] to allylurea in dry methanol. The product

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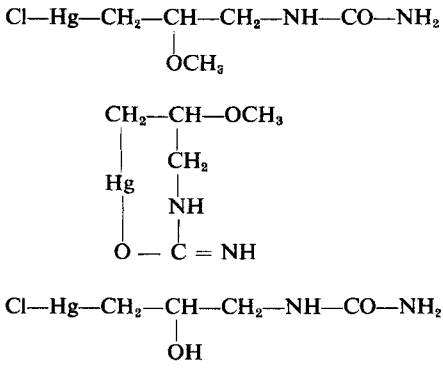
can be purified by repeated crystallization from dry methanol. The inorganic mercury has been removed by recrystallization [10], by adsorption on alumina [7] or on the anion-exchange resin [11]. The radiochemical purity of labelled chlormerodrin has been assayed by paper chromatography. The activity in free inorganic mercury is usually absent; the activity is mostly found in the spot of the chlormerodrin. In some cases; other organic labelled compound has been detected, either immediatelly after synthesis [12] or on storage [13]. Anghileri [12] considered the second labelled compound present in solution as the anhydro form of chlormerodrin (II), the content of which was dependent on the pH of the solution and reached in neutral solutions 30-40 per cent of the total activity. On the other hand, Herzmann [14] took for this compound to be 3-chloromercuri-2-hydroxypropylurea (III) which arose from water present during the synthesis.



(I)

(II)

(III)

At preparation of labelled chlormerodrin by isotopic exchange reaction [11] is has been also found that the composition of the product has been dependent on the reaction conditions, especially on the solvent used. The labelled chlormerodrin has been obtained only by using methanol as the solvent. By carrying out the exchange reaction in ethanol the activity has been found mostly in a new compound, which has been identified as 3-chloromercuri-2-ethoxypropylurea. Only 3-chloromercuri-2-hydroxypropylurea has been found using water as the solvent for the exchange reaction. In addition, the excess of chloride ions inhibits the exchange reaction.

The stability of chlormerodrin-^{203}Hg on storage has been studied by Anghileri [12]. Sarrach *et al.* [15] investigated polarographically the radiolysis of chlormerodrin in an aqueous solution. Various solvents for paper chromatography of chlormerodrin have been described [5-7, 10-12, 16], very useful are those mentioned in technical reports of various producers [10, 17, 18]. The thin-layer chromatography of chlormerodrin seems to be less convenient [6]. The choice of chromatographic solvent has been sometimes too influenced by opinions on structure assumed. Glenn and Kidwell [19] selected such a solvent

for the stability studies, that separated only ionic mercury from chlormerodrin, without distinguishing between organic mercury compound; they supposed that the distribution of activity between the regular and the "anhydro" form was caused by water present in the chromatographic solvent.

The purpose of this work was to study the processes which take place during the autoclaving and storage of labelled chlormerodrin. We used the method of accelerated experiments, discussed in our previous paper [20]. A brief description of our results was presented at the regional meeting [21].

1. — Experimental part.

3-chloromercuri-2-methoxypropylurea was prepared from allylurea and mercuric chloride in dry methanol in the presence of potassium acetate [9]. The product was purified by threefold recrystallization from ethanol [8, 11]. The labelled chlormerodrin-^{203}Hg was prepared in Nuclear Research Institute of Czechoslovak Academy of Sciences by the same manner. The labelled chlormerodrin was purified by threefold recrystallization from methanol. The initial specific activity was 3 mCi per gram of chlormerodrin. The solutions prepared from labelled chlormerodrin had the pH value of 6-6.5.

3-chloromercuri-2-hydroxypropylurea was obtained by addition of mercuric chloride to allylurea in very dilute aqueous solution [22].

3-chloromercuri-2-hydroxypropylurea-^{203}Hg was prepared by isotopic exchange reaction between mercuric chloride-^{203}Hg and chlormerodrin in aqueous solution [11].

Mercuric chloride-^{203}Hg was prepared in Nuclear Research Institute of Czechoslovak Academy of Science. The neutron-irradiated mercuric oxide was dissolved in hydrochloric acid. The last traces of acid were removed by manyfold recrystallization of mercuric chloride from hot water. The initial specific activity was 5 mCi per gram of mercuric chloride.

The other substances used in experiments (benzylalcohol, sodium chloride, theofyllin, solvents for chromatography) were of Pharmacopoeia purity or of "pro analysi" grade.

Water used for the preparation of solution was boiled with potassium chromate, then with diphenylpicrylhydrazone and distilled twice.

The samples were irradiated by gamma-rays of a cobalt-^{60}Co source under well defined conditions (each irradiation position was calibrated by a Fricke dosimeter. The dose rate of 2.97×10^{18} eV/ml per hour was used for most samples; the other values are mentioned in the text.

Solutions under study were irradiated mostly in 0.25-0.50 ml aliquots, sealed in normal glass ampoules. All ampoules were stored in the darkness, at room temperature.

The outgassed solutions and solutions for thermal experiments were filled into thick glass ampoules.

The outgassing was carried out by usual technique, i.e. by threefold freezing, and subsequent melting in vacuo (10^{-4} mm Hg).

The heating of samples was performed in water or Glycerine bath at a temperature constant within $\pm$ 1° C.

The mixture of ethanol — 1.5 N ammonia 8 : 2, containing 2g of sodium chloride per 100 ml of solvent was used for most chromatographic analyses in the stability studies.

Chromatographic paper Whatman No 4 was used in all analyses. The ethanolic solution of dithizone (0.05 percent) containing 1 percent of acetic acid was used for the detection of the spots.

The activity of paper chromatograms was counted under a thin-window GM tube (FHZ 15 *a*, mica 1.1 mg per cm^2) by a Frieseke-Hoepfner assembly.

2. — Results.

2.1. — *Chromatography of chlormerodrin and products of its decomposition.*

The chromatographic behaviour of chlormerodrin and of expected radiochemical impurities was checked in several chromatographic solvents. The R_F value of chlormerodrin, 3-chloromercuri-2-hydroxypropylurea and mercuric ions are presented in Table 1. All these test substances were applied

Table 1. The R_F values of chlormerodrin (A) 3-chloromercuri-2-hydroxypropylurea (B) and mercuric ions (C) in some descending chromatographic solvents.

Solvent	Reference	R_F			Front after 5 hours, cm
		A	B	C	
Ethanol-water-ammonia 6 : 2 : 1	17	0.40[a]	0.29[a]	0.1[a]	29-30
Ethanol-1.5N ammonia 8 : 2	11	0.42[a]	0.28[a]	0-0.1[a]	30-33
Ethanol-phosphate buffer (pH 7.4) 1 : 1	12	0.77	0.65	0-0.15[a]	26-27
		0.74[b]	0.62[b]	0-0.15[b]	
Butanol-pyridine-water-ammonia 5 : 7 : 1 : 3	18	0.32	0.20	0.09[a]	32-33
Butanol-pyridine-water 10 : 3 : 3	10	0.55	0.37	0.90	31-32

[a] The R_F values were influenced by the composition of the analysed solution;
[b] Ascending chromatography.

either as solutions in pure water or in 0.9 percent solution of sodium chloride, or as solutions containing equivalent amount of theophylline, to evaluate the possible influence of substances which are often present in commercial products. The influence of both substances was really observed in chromatographic solvents marked in Table 1., even the double-spots arose in some of them. This doubling can be removed by addition of the respective substance, i.e. sodium chloride or theophylline, to the chromatographic solvent. The results obtained for one of the chromatographic solvent are shown in Table 2. Also in other solvents such effect was observed, for example in Anghileri's solvent [12] the R_F value of mercuric ions changed from 0.0-0.15 to 0.7, when the analysed sample contained 0.9 percent of sodium chloride.

TABLE 2. The change in R_F values of chlormerodrin and its radiochemical impurities due to the addition of complexing substance to chromatographic solvent Ethanol-1.5 N ammonia 8 : 2.

Addition	R_F		
	A	B	C
None	0.42	0.28	0-0.1
2 g of sodium chloride per 100 ml of solvent	0.49	0.37	0.0
0.2 g of theophylline per 100 ml of solvent	0.64	0.50	0.40

A, B, C. See Table 1.

The influence of the pH value of the chromatographic solvent on R_F value of chlormerodrin was studied in the set of experiments, to check the Anghileri's hypothesis [12] concerning the transformation of chlormerodrin in the form I. to form II. This influence was studied in chromatographic solvents containing as buffer substances only acetic acid and sodium hydroxide (containing 10 percent of carbonate) to prevent the complexing action of various anions (chlorides, phosphates etc.). The solution of labelled chlormerodrin and 3-chloromercuri-2-hydroxypropylurea in the ratio of 4 : 1 was used for testing. The R_F values found are presented in Table 3. The ratio of measured activities in both spots remained unchanged in the whole pH range studied.

A certain decomposition of chlormerodrin on paper was observed during the analyses. The apparently high percentage of inorganic mercury was found on chromatograms which had been stored more than three hours after the applying of the analysed solution to the paper as compared with the chroma-

TABLE 3. The change in R_F values of chlormerodrin (A) and 3-chloromercuri-2-hydroxypropylurea (B) due to the pH of chromatographic solvent, containing in each case ethanol-buffer solution 1 : 1.

pH[a]	R_F	
	A	B
2.9	0.73	0.59
4.0	0.89	0.78
5.0	0.89	0.72
5.9	0.88	0.63
7.0	0.86	0.2-0.7
8.0	0.83	0.25
9.2	0.86	0.25
10.6	0.84	0.27
11.4	0.88	0.30
13	0.76	0.37

[a] The pH value was measured before the mixing of buffer solution with ethanol.

tograms developed immeditately after drying. The storing of chromatograms overnight led to entirely erratic results even when they had been stored in the darkness. The temperature of drying does not influence the results.

2.2. — *The thermal stability of chlormerodrin.*

The rise of a new organomercury compound was observed in the heated aqueous solutions of chlormerodrin. This compounds was identified by paper chromatography as 3-chloromercuri-2-hydroxypropylurea. The rate of transformation from 2-methoxy- to 2-hydroxy-compound increased with increasing the temperature. The results are shown in Figure 1. The increase of inorganic mercury was not observed during the heating. In solution of chlormerodrin containing 0.9 percent of sodium chloride no transformation of chlormerodrin was observed (within the error limit $\pm$ 1 percent) even at the temperature of 120° C after 30 minutes. On the other hand at the presence of theophylline the transformation rate was higher than in solutions of chlormerodrin alone (Fig. 1.). The presence of theophylline also accelerated markedly the transformation of chlormerodrin at room temperature. The results are presented in Table 4; in these experiments the initial radioactive concentration of ^{203}Hg was 11 μCi/ml.

2.3. — *The radiation stability of chlormerodrin.*

In preliminary experiments it has been found that the products of radiation decomposition of chlormerodrin were mostly the mercuric ions and, to less extent, 3-chloromercuri-2-hydroxypropylurea.

The radioactive concentration of ^{203}Hg was not higher than 12 μCi/ml in all radiation experiments. The results are presented as the percentage of decrease of chlormerodrin, in some cases the percentage of activity in the arising 3-hydroxy-compound are also shown. In all experiments, the results have been corrected to the zero time content of inorganic mercury as well as 3-hydroxy-compound, the sum of which never exceed 1.5 percent of the total activity.

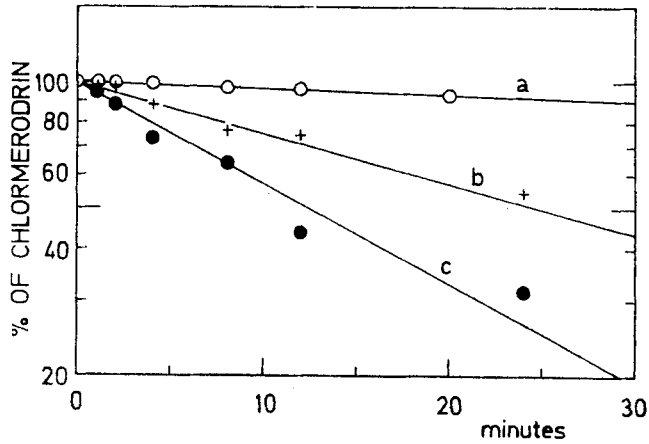


FIG. 1. The decomposition of 10^{-2}M solutions of chlormerodrin. (*a*) chlormerodrin alone, 100° C; (*b*) 120° C; (*c*) chlormerodrin with theophylline in molar ratio 1 : 1, 120° C.

A white opalescence, or at higher doses a white precipitate was observed in gamma-irradiated solutions of chlormerodrin. The sample had to be homogenized by shaking, before applying to the paper. The eventual inhomogeneity was partially eliminated also by repeated applying to the start spot. The results of parallel experiments showed that the error caused by this reason became higher only for samples decomposed by radiation to more than 50 percent.

It was found that the curves representing the decomposition of chlormerodrin as a function of absorbed dose of gamma radiation consist of two parts, the first one corresponding to the lower decomposition rate. The results obtained for various concentration of chlormerodrin are presented in Figure 2. From these curves the yields of radiation decomposition ($G_{(-M)}$) of chlormerodrin were calculated for both stages of decomposition. The plot of $G_{(-M)}$ values as a function of concentration is shown in Figure 3.

To explain the break on the curves mentioned above, the outgassed solutions of chlormerodrin were irradiated. The same course of decomposition, but higher yield of formation of 2-hydroxy-compound was found in these samples as compared with the aerated ones. The further experiments were

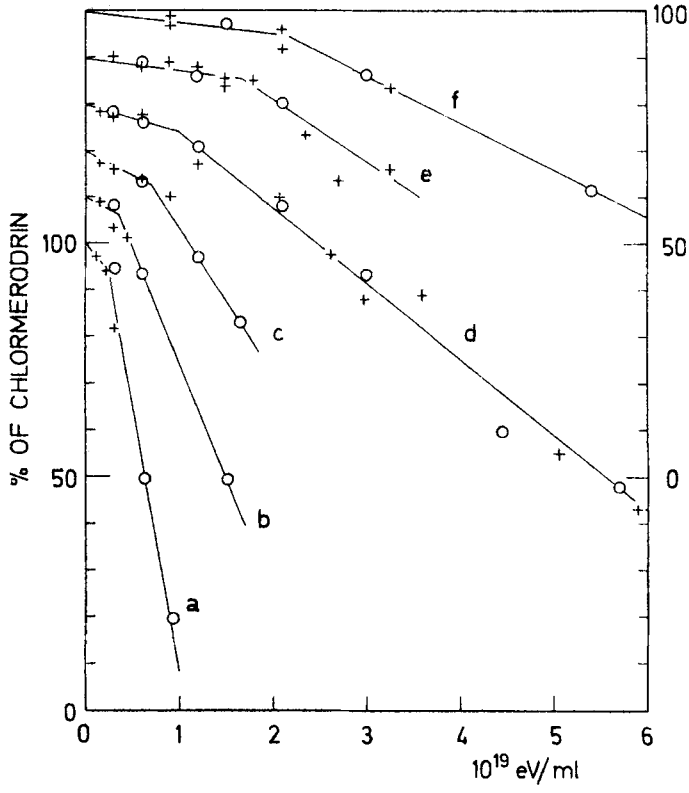


FIG. 2. The percentage of decomposition of chlormerodrin as a function of absorbed energy dose. The concentrations of chlormerodrin were as follows : (*a*) 3×10^{-4}M, (*b*) 1×10^{-3}M; (*c*) 3×10^{-3}M; (*d*) 6×10^{-3}M; (*e*) 1×10^{-2}M; (*f*) 1.77×10^{-2}M. Chlormerodrin was dissolved in water (+) or in 0.9 percent solution of sodium chloride (o). The individual curves are shifted along the y-axis by steps amounting to 10 percent.

carried out with normal samples irradiated at lower dose rate (3.58×10^{17} eV/ml/hod). The results are presented in Figure 4.

The presence of 0.9 percent of sodium chloride had no effect on the radiation decomposition of the chlormerodrin within the experimental error. The results are presented in Figure 2 and have been used in the calculations of the radiation yields $G_{(-M)}$. The presence of sodium chloride inhibited the formation of 2-hydroxy-compound so that only the inorganic mercury was found as the product of radiation decomposition.

The presence of 0.9 percent of benzylalcohol influenced markedly the course of radiation decomposition of chlormerodrin; the results are presented in the Figure 5. Also the formation of 3-chloromercuri-2-hydroxypropylurea was higher in these samples.

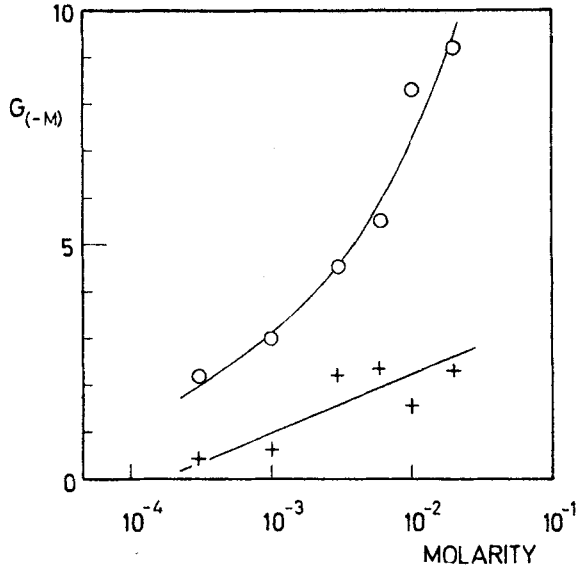


FIG. 3. The yield of radiation decomposition as a function of the concentration of chlormerodrin. The initial radiation yield (+), the radiation yield in the second stage (o).

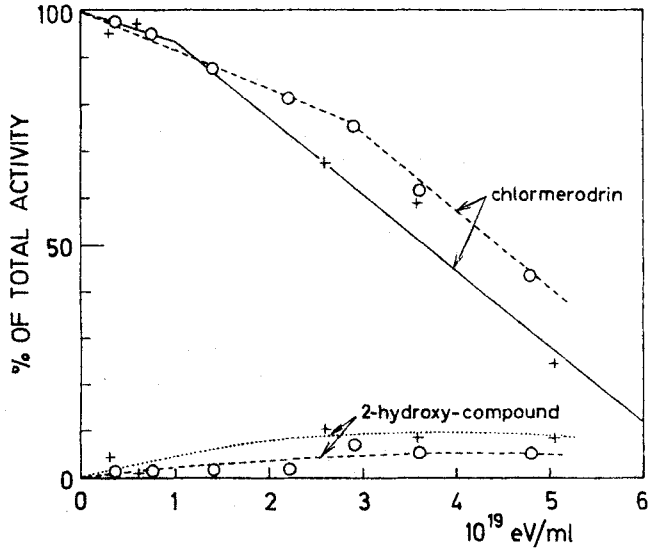


FIG. 4. The decomposition of chlormerodrin and formation of 2-hydroxy-compound as a function of absorbed energy dose. The concentration of chlormerodrin was 6×10^{-3}M. (o) Solutions irradiated at the lower dose rate. (+) outgassed solutions. The full line corresponds to the curve (*d*) from the Figure 2.

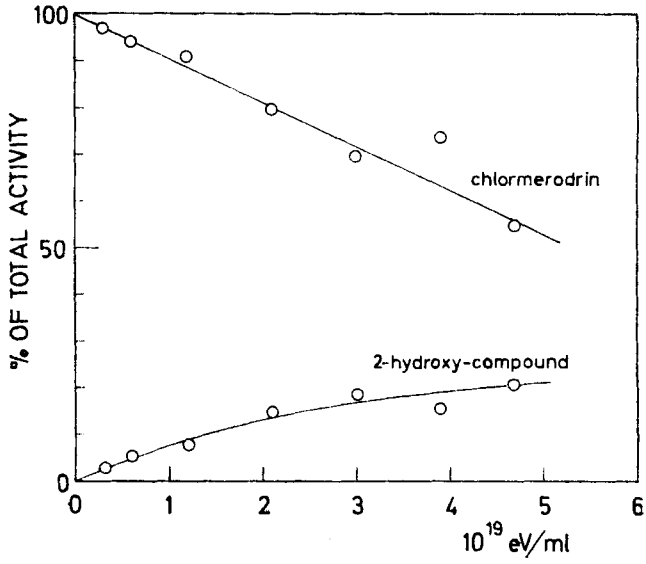


FIG. 5. The decomposition of chlormerodrin and formation of 2-hydroxy-compound as a function of absorbed energy dose. The concentration of chlormerodrin was 6×10^{-3}M, the concentration of benzylalcohol was 0.9 percent.

The influence of theophylline on the radiation decomposition could not be quantitatively estimated. Even the lowest doses applied (6×10^{18} eV/ml) caused in 1×10^{-2} M solutions a voluminous non homogenisable precipitate; thus the aliquot applied to the paper represents only the solution.

The 3-chloromercuri-2-hydroxypropylurea was the product of thermal, as well as radiation decomposition. The orientational experiments on radiation decomposition of this compound were therefore carried out. In these experiments the 6×10^{-3} M solution of labelled chlormerodrin was first heated at 120° C to obtain approximately 50 percent of transformation to 2-hydroxy-compound. Then this solution was irradiated by gamma-rays. The results are shown in Figure 6.

The survey of radiation decomposition of chlormerodrin under various conditions is presented in Figure 7 without experimental points, for easier comparison. The third component, which is not plotted, is the inorganic mercury; (the sum of chlormerodrin, 2-hydroxy-compound and inorganic mercury give the 100 percent in all cases).

3. — DISCUSSION.

The results obtained by chromatography of inorganic mercury, chlormerodrin and 3-chloromercuri-2-hydroxypropylurea were helpfull in explanation of discrepancies existing in literature.

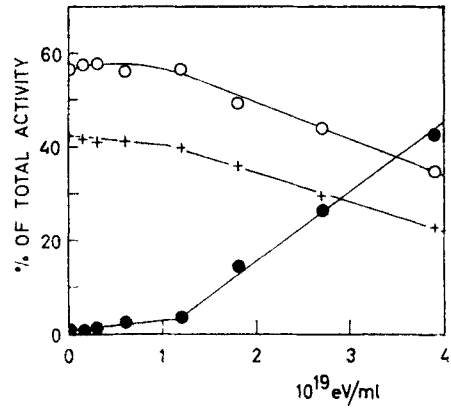


FIG. 6. The radiation decomposition of the mixture of chlormerodrin and 2-hydroxy-compound as a function of absorbed energy dose. The initial concentration of chlormerodrin (+) was 2.54×10^{-3}M; of 2-hydroxy-compound (o) 3.4×10^{-3}M; and of inorganic mercury (●) 6×10^{-5}M.

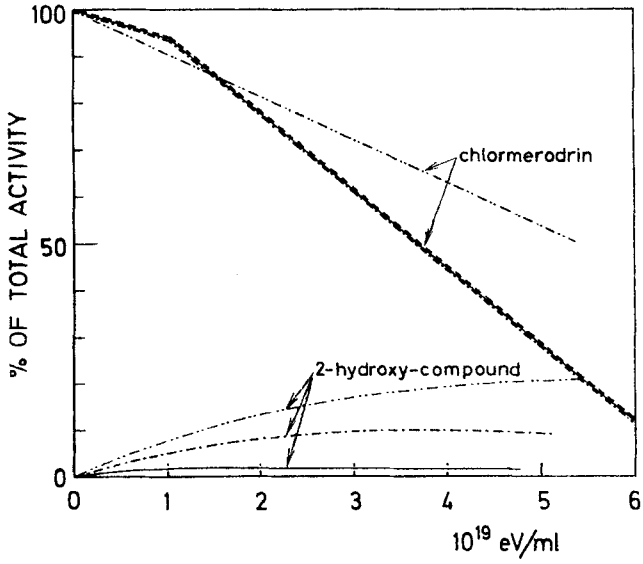


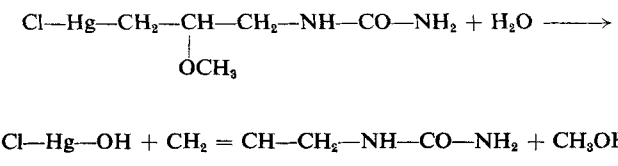
FIG. 7. The survey of radiation decomposition of chlormerodrin and formation of 3-chloromercuri-2-hydroxypropylurea at various conditions. The irradiation was carried out with 6×10^{-3}M solutions of chlormerodrin in water (—), in outgased water (—.—.—), in 0.9 per cent solution of sodium chloride (- - - -) and in 0.9 percent solution of benzylalcohole (—. .—. .).

The carefully recrystallized chlormerodrin gave only one spot containing more than 99 percent of the total activity of chromatogram. The results of elementary analysis showed [11] that the crystalline chlormerodrin contained chlorine in amount corresponding to the formula I. The substitution of the chlorine by OH group or other anion can easily occur in solution, as follows from the simple preparation of various substances differing only in the nature of anions; this can be done only by addition of the respective salt in excess (acetate, chloride, bromide, phosphate). The travelling of chlormerodrin as 3-hydroxymercuri-2-methoxypropylurea in alkaline chromatographic solvent can be therefore expected. The addition of chloride ions to the solvent, containing the comparatively low concentration of hydroxide ions, suppressed the above mentioned substitution. In such modified solvent chlormerodrin travels unchanged as 3-chloromercuri compound and this can explain the change of R_F value (Table 2). Theophylline has an analogous effects; the complex chlormerodrin — theophylline can dissociate during the development and the double spots occurs. The commercial preparations contain usually other substances, especially theophylline and sodium chloride (for isotonicity). The presence of these substances can explain the number of spots found in some chromatographic solvents [16]. It can be recommended for analysis of chlormerodrin to use only solvents without the above-mentioned effect (see Table 1), or alkaline chromatogaphic solvent with added sodium chloride, the high concentration of which suppresses the formation of other spots and, in addition, fixes the mercuric ions better on the start.

The R_F values of mercuric ions and of 3-chloromercuri-2-hydroxypropylurea are also affected by the nature of anion in analysed solution or solvent used; in some cases the presence of sodium chloride makes the solvent unapplicable.

The Anghileri's hypothesis [12] on transformation of form I to form II was not verified by our experiments. The R_F values assigned by Anghileri [12] to form I and II are identified according to our results as chlormerodrin (I) and 3-chloromercuri-2-hydroxypropylurea (III). The reversibility of transformation of form I to form II as a function of pH was also not observed. No changes in the activity ratio of both spots were found in the pH interval studied.

The behaviour of chlormerodrin during the heating or storage of its solution can be explained by the following mechanism. The chlormerodrin is first decomposed to the original compounds, i.e. allylurea, inorganic mercury and methanol



$$Cl—Hg—CH_2—\underset{\displaystyle OCH_3}{\underset{|}{CH}}—CH_2—NH—CO—NH_2 + H_2O \longrightarrow$$

$$Cl—Hg—OH + CH_2 = CH—CH_2—NH—CO—NH_2 + CH_3OH$$

This decomposition, which is characteristic for all mercury compounds prepared by addition of mercury to a double bond, is accelerated by acids. The inorganic mercury can be again added to allylurea. Owing to the fact that this addition occurs in aqueous solutions of components, the 2-hydroxy-compound arise.

$$Cl—Hg—OH + CH_2 = CH—CH_2—NH—CO—NH_2 \longrightarrow$$

$$Cl—Hg—CH_2—\underset{\displaystyle OH}{\underset{|}{CH}}—CH_2—NH—CO—NH_2$$

The overall reaction followed the first order kinetics, the decomposition of chlormerodrin being the rate determinating step. The above-mentioned mechanism is in full accordance with the earlier isotopic exchange results [11]. Similar mechanism has been recently proposed by Stanko and Burtseva [23].

The strength of mercury-carbon bond is considerably influenced by other substances present in the solution of chlormerodrin. Theophylline makes this bond unstable, the decomposition of chlormerodrin is then accelerated (Table 4, and Fig. 1.) not only during the elevated temperature but also at room temperature. On the other hand the mercury-carbon bond is highly stabilized by the presence of sodium chloride. Neither 2-hydroxy-compound nor inorganic mercury were found in heated solution of chlormerodrin in 0.9 percent sodium chloride. This fact has considerable practical significance owing to the higher uptake of 2-hydroxy-compound in kidney and its slower excretion as compared with chlormerodrin [24].

TABLE 4. The amount of 3-chloromercuri-2-hydroxypropylurea found in solutions of chlormerodrin on storage in darkness at room temperature.

The composition of the solution	Days	% of activity found in 2-hydroxy-compound
10^{-2}M chlormerodrin	3	0.8
	19	3.0
10^{-2}M chlormerodrin and 10^{-2}M theophylline	4	9.2
	13	30.2
	39	55.6

Only inorganic mercury and 3-chloromercuri-2-hydroxypropylurea were found as mercury-containing products of the radiolysis of chlormerodrin. The formation of 2-hydroxy-compound can be explained again by the addition of splitted mercury to allylurea, as in the case of thermal decomposition.

There are two causes for the inhibition of the complete re-addition of mercury : (i) The mercury which is lost by the original molecule of chlormerodrin is probably reduced to some extent to mercurous ions, and the insoluble calomel (the white precipitate observed) is incapable of the new addition. (ii) The second part of the broken molecule, i.e. the allylurea, undergoes the reactions with radicals. The fraction of mercury which remains in divalent state thus cannot be completely re-added owing to the lowering of double bond concentration.

The latter cause mentioned above is supported by the results of radiolysis of outgassed solutions and the solutions containing benzylalcohol, as compared with the normal solution of chlormerodrin. The formation of 2-hydroxy-compound was slightly increased in the absence of oxygen, while in the presence of benzylalcohol more than one half of the splitted mercury was found in the mentioned compound. The results are unsufficient for the accurate estimation of radical which predominantly reacts with the allylurea.

The presence of chloride ions does not influence the rate of decomposition of chlormerodrin but hinders completely the formation of 2-hydroxy-compound. This can be explained by the formation of $[HgCl_4]^{2-}$ complex, which is incapable of the addition (compare the complete inhibition of isotopic exchange at the presence of sodium chloride [11]).

The break on the curves of radiation decomposition of chlormerodrin has not been explained. The change of the radiation yield of decomposition is not caused by the consumption of oxygen during the radiolysis because the change is well observable even for the lowest concentration of chlormerodrin at the doses unsufficient for the removal of oxygen. Furthermore, the decomposition curve was identical, within the experimental error, for normal and outgassed solution.

The value of $G_{(-M)}$ 4.8. presented by Sarrach *et al.* [15] for 1.77×10^{-4} M solution chlormerodrin is much higher than our values in this concentration region (see Fig. 3), probably due to the different analytical method used. Other results of Sarrach *et al.* [15] are incomparable with ours; they were estimated for solutions containing several other substances.

The presence of free inorganic mercury in labelled chlormerodrin solutions is usually limited to 3 percent of the total activity. Only the values of initial $G_{(-M)}$ must be therefore used for the calculation of possible radiation decomposition of chlormerodrin. The results presented in Figure 8, show the calculated decomposition of chlormerodrin as a function of the absorbed energy for various concentrations of chlormerodrin. In evaluating the radiation decomposition of labelled chlormerodrin the following procedure is to be recommended.

(1) From the initial value of radioactive concentration of mercury-^{197}Hg or ^{203}Hg, and the storage time, the respective value of total absorbed energy (in eV/ml) is calculated using the Tables 4. and 5. (and corrections for geometrical shape, if necessary) of our previous paper [20].

(2) The radiation decomposition for this value of absorbed energy and the given concentration of chlormerodrin is found using the Figure 8.

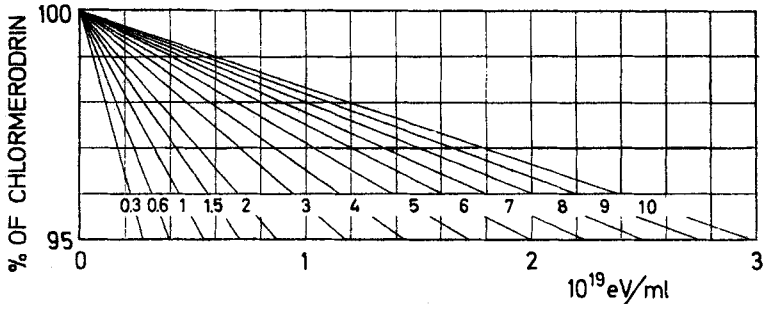


FIG. 8. The calculated decomposition of chlormerodrin as a function of the absorbed energy. Individual concentrations of chlormerodrin are expressed in mg/ml.

It is obvious that the value found must be corrected with respect to the initial conditions, i.e. the initial radiochemical impurities. The values found using the Figure 8 must be taken as rough owing to the fact, that the initial $G_{(-M)}$ values have unfortunately a great range of error.

The calculations mentioned above are correct only for the solutions of chlormerodrin alone or chlormerodrin containing 0.9 percent of sodium chloride. The presence of benzylalcohol increases the initial yield of radiation decomposition of chlormerodrin; its addition to chlormerodrin is therefore not convenient. The addition of theophylline to the solution of chlormerodrine also cannot be recommended; the white precipitate which appears at the lowest doses of absorbed energy makes the injections of chlormerodrin inapplicable.

The initial presence of 3-chloromercuri-2-hydroxypropylurea in the solutions of chlormerodrin does not influence the decomposition of chlormerodrin (Fig. 6)., the results of the Figure 8 are applicable also for the cases of thermaly decomposed chlormerodrin.

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REFERENCES

1. BLAU, M. and BENDER, M. A. — *J. Nucl. Med.*, **1** : 107 (1960).
2. MCAFFEE, J. G. and WAGNER, H. N. Jr. — *Radiology*, **75** : 820 (1960).
3. WAGNER, H. N. Jr. — Radioactive organic compounds of mercury. In Radioactive Pharmaceuticals. *Proc. Sympos.*, Oak Ridge, November 1-4, 1965. CONF-65 1111, USAEC April (1966), p. 399.

4. BLAU, M. and BENDER, M. A. — *J. Nucl. Med.*, **3** : 83 (1962).
5. ANGHILERI, L. J. — *Nucl. Med.*, **4** : 193 (1964).
6. BURTSEVA, L. N., MIRKINA, N. N., SYSOEVA, E. TS. and KHARLAMOV, V. T. — Metody analiza radioaktivnych preparatov. *Atomizdat*, Moscow (1965), p. 121.
7. MANI, R. S., DESAI, C. N. and RAGHAVAN, S. V. — *Indian J. Chem.*, **3** : 415 (1965).
8. ROWLAND, R. L., PERRY, W. L., FOREMAN, D. and FRIEDMAN, H. L. — *J. Amer. Chem. Soc.*, **72** : 3595 (1950).
9. BARZA, S. A. — *Brit. Pat.* 792, 992, Apr. 9, 1958; *Chem. Abstr.*, **52** : 18222e (1958).
10. PŘITASIL, L. and MATUCHA, M. — Laboratory Report of the Institute of Research, Production and Use of Radioisotopes, Prague, December (1965).
11. CÍFKA, J., KAČENA, V. and KRONRÁD, L. — Proc. Second Internat. Conf. on Methods of Preparing and Storing Labelled Molecules (Brussels 28 Nov.-3 Dec. 1966). EURATOM (1968), p. 461.
12. ANGHILERI, L. J. — *J. Nucl. Med.*, **4** : 410 (1963).
13. PŘITASIL, L. — Private communication (1966).
14. HERZMANN, H. — Private communication (1966).
15. SARRACH, D., HERZMANN, H. and VORMUM, G. — *Isotopenpraxis*, **2** : 145 (1966).
16. HEINRICH, H. C., GABBE, E. E., MEINEKE, B. and KÜHNAU, J. Jr. — Proc. Second Internat. Conf. on Methods of Preparing and Storing Labelled Molecules (Brussels 28 Nov.-3 Dec. 1966). EURATOM (1968), p. 1063.
17. Radiochemical Centre Amersham. — Specification M. B. 19 P, November (1963).
18. Radiochemical Centre Amersham. — Specification M. 19 P, December (1965).
19. GLENN, H. J. and KIDWELL, R. E. — Radioactive Pharmaceuticals. Proc. Sympos., Oak Ridge November 1-4, 1965. CONF-65 1111, USAEC, April (1966), p. 171.
20. CÍFKA, J. and BURIÁNEK, J. — *J. Labelled Compounds*, **4** : 107 (1968).
21. BURIÁNEK, J. and CÍFKA, J. — The Stability of Injections of Chlormerodrin-^{203}Hg (In Russ.). Presented on the Meeting on the use of chromatography and high-voltage electrophoresis in analysis and control of radioactive preparations, held by Council of Mutual Economic Cooperation in Mariánské Lázné (Czechoslovakia), October 1966.
22. CÍFKA, J. and KRONRÁD, L. — Unpublished results.
23. STANKO, V. I. and BURTSEVA, L. N. — *Zh. obshch. khim.*, **37** : 2633 (1967).
24. HERZMANN, H., LOECHEL, M. and SCHWARTZ, K. D. — *Radioaktive Isotope* in *Klinik u. Forschung. Sonderbände zur Strahlentherapie*, **65** : 480 (1967).