Table 2

Molarity BCDR	Slope ratio = R
10^{-5}	1·304 ± 0·026
10^{-4}	1·74 ± 0·01
10^{-3}	2·56 ± 0·04

Where R = ratio of the slope of the bromodeoxycytidine (BCDR) treated cells to the slope of the controls. Standard error computed according to the relationships $s^2R = (s^2bt + R^2s^2bc)/bc^2$. Where b_t = slope of treated and b_c slope of controls.

increase in slope ratio a decrease in the shoulder of the survival curve is noted. In spite of a substantial change in radiosensitivity at 10^{-4} M 5-bromodeoxycytidine no manifestation of toxicity was evident since the population of cells in the stock bottle was slightly greater than in the control stock and the plating efficiency was equal to the plating efficiency of the untreated controls (Table 1). At 10^{-3} M 5-bromodeoxycytidine some toxicity due to the drug alone was evident.

Although the results do not permit quantitation, experiment 3 demonstrated that most of the bromine-82 was recovered from the DNA fraction of cells incubated with ^{82}Br-5-bromodeoxycytidine. The nature of the moiety containing the bromine-82 was not determined. Cramer *et al.*[3] have suggested that 5-bromodeoxycytidine is incorporated into DNA as 5-bromodeoxyuridylic acid.

Although Djordjevic and Szybalski[4] have demonstrated an increase in radiosensitivity of cells pretreated with 5-bromodeoxyuridine *in vitro* clear confirmation of this effect *in vivo* has been difficult to obtain. Kriss and Revesz demonstrated in the rat that ^{82}Br-5-bromodeoxyuridine was rapidly debrominated by the liver[5]. Their detailed studies on the metabolic pathways of 5-bromodeoxyuridine and 5-bromodeoxycytidine also demonstrated that in the rat ^{82}Br-5-bromodeoxyuridine injected intravenously rapidly disappeared from the circulating blood, whereas ^{82}Br-bromide accumulated. Experiments also demonstrated that ^{82}Br-5-bromodeoxycytidine was debrominated more slowly than the ^{82}Br-5-bromodeoxyuridine. In addition the uptake of bromine-82 into ascites tumour cells was greater in mice receiving intraperitoneal injections of ^{82}Br-5-bromodeoxycytidine than in mice receiving an equimolar amount of ^{82}Br-5-bromodeoxyuridine. The *in vivo* evidence presented by Kriss and Revesz demonstrated different patterns of tissue distribution of the bromine-82 which depended upon whether the bromine-82 was administered as ^{82}Br-5-bromodeoxyuridine or ^{82}Br-5-bromodeoxycytidine. The experiments presented here indicate that 5-bromodeoxycytidine is an effective radiosensitizing agent for mammalian cells *in vitro*. Both studies provide groundwork for the *in vivo* evaluation of 5-bromodeoxycytidine as a radiosensitizing agent.

This work was supported by grant *CY*-4542, U.S. Public Health Service, National Institutes of Health.

I am indebted to Dr. Joseph P. Kriss for supplying the ^{82}Br-bromodeoxycytidine used in experiment 3.

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Regularities of Metal Excretion from Organisms on Late Application of Complexons

Complexons are of interest to biologists from various points of view: (1) their ability to affect sharply the behaviour of polyvalent metals in organisms; (2) their applicability as models for complex-forming substances (amino-, nucleic and other acids) naturally occurring in organisms. The ability of complexons to decrease the deposition of different metals in tissues and to increase their excretion from the organism have found a vast application in therapy of metal and radioisotopic poisoning.

At present, when considerable data on the subject[1–13] have already been accumulated, some attempts are being made to ascertain general effects of complexons on the behaviour of metal in organisms. The greater part of investigations in this subject was undertaken on the early application of complexons (some minutes after incorporation of radioisotopes), when the principal mass of metal is still in blood, for it is then that complexon efficiency is most pronounced. From the practical point of view, however, reactions at late complexon application, when the metal is already fixed in tissues, is of even greater interest.

Based on previously achieved results[11,12], an attempt was made to establish a relationship between excretion of radioisotopes in urine and, so called, calcium displacing constants ($K_{dsp.}$), which shows how many times the bond-strength of a given complexon (X) with a metal (M), expressed by stability constant $\left(K_M = \frac{MX}{M \times X}\right)$, exceeds the strength of its bond with calcium $\left(K_{Ca} = \frac{CaX}{Ca \times X}\right)$; otherwise $K_{dsp.} = K_M/K_{Ca}$. Logarithms of K_{Ca}, K_M and $K_{dsp.}$ (in brackets) for cations, which are interesting for us, are given in Table 1.

Table 1

	Complexons	Ca^{2+}	Ce^{3+}	Y^{3+}	Pb^{2+}
(1)	Diethylenetriaminepentaacetate (DTPA)	10·9	20·4 (9·5)	20·4 (9·5)	18·9 (8·0)
(2)	Ethylenediaminetetraacetate (EDTA)	10·8	16·0 (5·2)	18·0 (7·2)	18·3 (7·5)
(3)	1,2-Diaminocyclohexanetetra-acetate (CDTA)	12·5	16·8 (4·3)	19·2 (6·7)	19·7 (7·2)

Expediency of comparison of the efficiency of complexons not with the absolute K_M but with $K_{dsp.}$ (taking into account the high concentration of free calcium in blood and intercellular fluid) has been already demonstrated[7,8].

In our experiments[11,12] 100 μM of the complexons indicated in Table 1 (in calcium form) were injected intraperitoneally to 3–4 months-old male rats on the 8th, 13th and 38th days after intravenous injection of carrier-free radioisotopes (yttrium-91, cerium-144 and lead-210). The amount of daily excretion of metals in urine was measured.

The values of isotopes excreted (percentage of injected dose) during 24 hr. after administration of complexons were the highest in the case of DTPA, which has the largest $K_{dsp.}$ The effect of EDTA and CDTA was less pronounced; it is especially clear in the case of cerium, where differences in $K_{dsp.}$ between DTPA and the two above-mentioned complexons reach quantities of about 10^4–10^5.

In comparing excretion values of different metals under complexons treatment at each separate period with log $K_{dsp.}$ (Fig. 1) one can see that the experimental points fall well into straight lines, described by

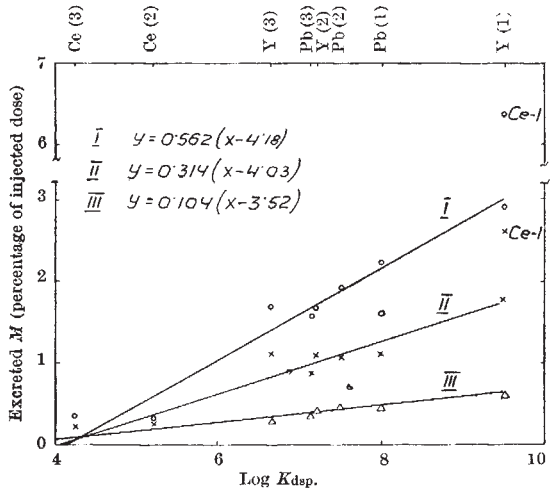

Fig. 1. Relationship between $K_{dsp.}$ and values of yttrium, cerium and lead excretion with urine, influenced by DTPA (1), EDTA (2) and CDTA (3), injected at eighth (curve I), thirteenth (curve II) and thirty-eighth (curve III) days of experiment

elementary mathematical function (deviation from linearity is statistically not significant, $P > 0{\cdot}2$). This permits us to make the following conclusions.

(1) Community of the curve for different complexons and different metals indicates their 'unspecificity', though it is quite probable that some other complexons with the same metals or other metals with the complexons considered will give an appreciable deviation from the curves presented, as is the case with cerium. Those deviations should demonstrate the specificity of a given metal or complexon. For example, the 'super-effect' of DTPA in the case of cerium (Fig. 1) appears to be a result of easy mobilization of this metal by complexon from liver (initial deposition of cerium in liver usually reaches 45–60 per cent, when that of yttrium and lead is only 7–10 per cent). With decrease in content of cerium in liver with time, this 'super-effect' is less pronounced (Fig. 1, curve II). A similar picture could be given by other 'liver' metals, too.

(2) The exponential nature of those curves allows us, by extrapolation, to evaluate the possible efficiency of stronger, as well as of weaker, complexons. As can be seen in Fig. 1, complexons with $K_{dsp.}$ less than 10^4 should be practically ineffective at the given stage of experiment.

(3) The shift to the left of the point of intersection of straight lines with abscissa (log $K_{dsp.}$: 4·18–4·03–3·52 on the 8th, 13th and 38th days; equations on Fig. 1) indicates a decrease of the efficiency-threshold with time, that is, complexons having a lesser value of $K_{dsp.}$ at later stages of the experiment become efficient. Confirmation of the existence of such a dependence would give an idea of changes in metal-tissues binding forces with time.

(4) Comparison of b (slope of the lines) for different periods indicates a linear regression in the bi-logarithmic scale. On this basis we can unite the equations given in Fig. 1 taking into account the alteration of b. However, we must refrain from such a generalization until we obtain further results, in particular, when radioisotopes will leave the soft tissues entirely and remain in the skeleton only. In such a case there is a possibility of considerable deviations in efficiency of the complexon in general (absolute efficiency) as well as in relative efficiency.

Thus, a quantitative dependence is obtained: effect—log $K_{dsp.}$ at a late application of complexons; this dependence is described by the elementary function. It allows us to make a quantitative interpretation of the experimental results and to elucidate interactions between complexons and metals in biological media. For practical purposes this dependence allows us: (*a*) to elaborate a method for quantitative determination of toxic metals or radioisotopes content in tissues according to the values of its excretion with urine under the influence of complexons; (*b*) to predict the efficiency of a new complexon if its $K_{dsp.}$ is known; (*c*) to determine the prospects of the synthesis of stronger complexons.

Further work on other metals is necessary for more precise development of regularities, dealt with in this work, taking into account the specificity of their metabolism and the age and sex of animals involved.

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BIOLOGY

Optical Rotatory Capacity of the Lens of the Vertebrate Eye

The lens of the eye can be considered as a transparent sac of proteins and as such should be amenable to examination with transmitted polarized monochromatic light. The results obtained when the lenses of sheep, lambs, steers, calves, dogs and rabbits were examined for their capacity, *in vitro*, to rotate the plane of polarized monochromatic light are reported.

The eyes used were obtained from local abattoirs or from the Department of Animal Care of this University, and all eyes were stored in the refrigerator until used. Three hours prior to the removal of the lenses, the eyes were allowed to warm up to room temperature (21–23° C.). No eye older than 3 days was used. The lens was removed by an anterior approach and at the same time as much of the adhering vitreous humor was removed from the lens. The capsule was carefully stripped off along with any adhering material. The lens was discarded if it did not have clear, unmarked anterior and posterior surfaces. The lens thickness was measured with a centimetre ruler graduated in millimetres. The lens was mounted in a brass holder so constructed that