MUTUAL CATALYSIS IN SYSTEM Co(II)-GLYCYLGLYCINE-ASCORBIC ACID-OXYGEN

By M. T. BECK and S. GÖRÖG

Institute of Inorganic and Analytical Shemistry, The University, Szeged

Many papers deal with the phenomena observed on the oxygenation of different Co(II)- amino acid and peptide complexes. It was observed that the oxygenation takes place in two steps. The first step is a reversible uptake of molecular oxygen followed by an irreversible process. The most thoroughly studied systems are Co(II)-histidine and Co(II)-glycylglycine [1], [2]. In the earlier investigations it was supposed that the valence of central ion does not change in a Co(II)-histidine system either in a reversible or an irreversible reaction. Tanford and his coworkers' paper does not deal with this problem definitely.

Our investigations aimed to study the catalytic effect of these compleses on autoxidation reactions. Co(II)-glycylglycine was chosen as model, since - according to earlier findings - in the case of Co(II)-histidine complex disturbing by processes have to be taken into consideration. First of all we examined the validity of the earlier statement that the oxidation state of central ion does not change during even the irreversible reaction.

The simplest way for the solution of this problem is the comparative study of the properties of Co(III) complexes and the products of irreversible oxidation [3]. A new method to prepare Co(III)-glycylglycine complex was elaborated. The essence of this method is that the complex is prepared from hexammine Co(III) chloride and glycylglycine in the presence of active carbon as catalyst. The ammonium salt was obtained with 90 per cent yield, the free acid with 30 per cent yield. The properties of complex obtained are the same as that of the product of irreversible oxidation of Co(II) glycylglycine. The anion character of the complex is proved by its electrophoretic behaviour and that the compound can be prepared in the form of ammonium salt.

It may be expected that the complex formed during the reversible oxigenation can catalyse the autoxidation reactions since the attachement of oxygen into the complex molecule causes the weakening of the O-O bond. Really, our experiments showed that the Co(II) glycylglycine complex is an effective catalyst of different autoxidation processes e.g. autoxidation of ascorbic acid.

The gasvolumetric kinetic experiments were carried out in a Csürös apparatus [4].

Experiments carried out at different pH showed that the velocity of catalytic reaction and the amount oxygen consumed increase with increasing pH(Fig. 1).

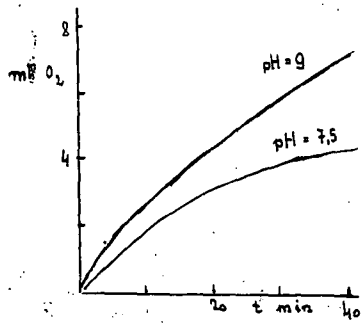


Fig. 1.

Catalyzed oxygen uptake of ascorbic acid at different pH. (Oxygen consumtion of blanks are substracted.)
$C_{asc} = 4.10^{-2}$; $C_{Co} = 4.10^{-3}$; térfogat 25 ml.

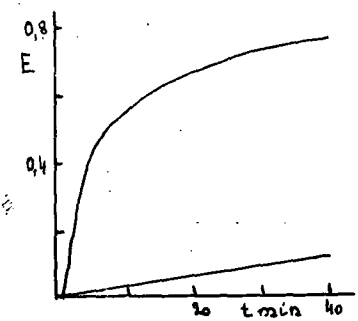
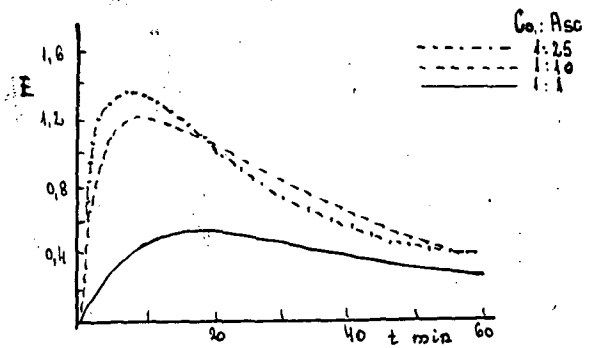


Fig. 2.

Change of absorbancy during the oxygenation of Co/II/-gycylglycine complex in the presence and absence of ascorbic acid.
$C_{Co} = 10^{-3}$ mole/1; $C_{asc} = 10^{-2}$ mole/1; pH = 8; cuvette 3,5 cm; filter S 52



ascorbic acid

Fig. 3.

Effect of pH on the formation of intermediate. $C_{Co} = 10^{-3}$ mole/1; pH = 7,5 cuvette 5 cm; wavelength 350 mμ.

Neither the Co(II) nor the product of irreversible oxidation (Co(III) glycylglycine complex) can catalyse the autoxidation of ascorbic acid; consequently, the oxygen carrying Co(II) glycylglycine complex is responsible for the catalysis observed. Below we shall present a direct evidence of this.

During these experiments an unexpected phenomenon was observed. Namely, in the presence of Co(II) glycylglycine complex not only the autoxidation of ascorbic acid is accelerated but the ascorbic acid also increases the velocity of oxygenation of Co(II) glycylglycine complex. This latter process can be easily followed by spectrophotometric measurements. The absorbancy of Co(II) glycylglycine is negligibly small in comparison to the absorbancy of both the oxygen carrying and the irreversible complexes. Simultaneous determination of these two compounds is possible because the brown intermediate has a selective absorbancy in the near ultraviolet while at 520 mμ the absorbancy both of the complexes is the same. So the measurement at 520 mu gives the sum of the complexes, while the measurement at 350 mμ gives the amount of the oxygen carrying complex only. Experiments were carried out partly with Pulfrich photometer in 3.5 cm cuvette by applying S 52 filter, partly with Beckman B spectrophotometer in 5 cm cuvette at 520 and 350 mμ.

Fig. 2 shows that the velocity of oxygenation of Co(II)-glycylglycine complex is greatly accelerated by ascorbic acid. The effect of the concentration of ascorbic acid on the oxygenation is shown in Fig. 3. For sake of a better perspicuity the data obtained at 350 mμ are plotted only. As can be seen from Fig. 3 the concentration of the oxygen carrying complex changes according to a maximum curve in time. The rate of oxygen consumption first increases on increasing the concentration of ascorbic acid, over certain concentrations it becomes independent of it. The absorbancy is not reduced to zero even after a longer time because the oxidation products of ascorbic acid also absorb at this wave length.

Fig. 4 shows the effect of pH on oxygenation. It is apparent that both the velocity of oxygenation and the maximum concentration of intermediate increase monotonously on increasing the pH. Concerning TANFORD and his coworkers' investigations - which are supported by our experiments - that the brown intermediate can be observed at higher pH values only (over pH 8), further, according to the date of Fig. 3 and 4, in the presence of ascorbic acid the intermediate exists for a longer time at relatively low pH, one must suppose that either the ascorbic acid or its oxidation products is built into the complex and hereby stabilises it. Here we note that the complex forming effect of the oxidation products of ascorbic acid exhibits in the autoinhibition phenomenon in this system what is evident from the curve d of Fig. 4. In this experiment the reaction mixture contained beside fresh ascorbic acid an amount of oxidized ascorbic acid, too. In this case the curve lies considerably lower than without oxidized ascorbic acid.

From the comparison of gasvolumetric (autoxidation of ascorbic acid) and the spectrophotometric (oxygenation of Co(II)-glycylglycine) measurements it can be seen that the velocity of autoxidation changes parallel to the concentration of brown intermediate and reduces to the velocity of non-catalyzed reac-

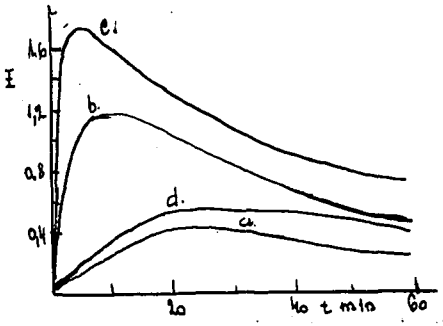


Fig. 4.

Effect of pH on the formation of intermediate. C_{Co} 10^{-3} mole/l; C_{asc} 10^{-2}; cuvette 5 cm; wavelength 350 mμ; a.) pH 6,9; b.) pH 7,5; c.) pH 8,1; d.) pH 7,5; the solution contains 10^{-4} mole oxidized ascorbic acid. too.

tion when the concentration of intermediate - the catalyst - is reduced to zero.

The autoxidation of ascorbic acid and the oxygenation of Co(II) glycylglycine are not simple reactions themselves. The simultaneous investigation of these makes almost impossible the quantitative treating og the data. Nevertheless the experiments show that in the system Co(II)-glycylglycine-ascorbic acid-oxygen mutual catalysis or otherwise the step-by-step activation of molecular oxygen have to be taken into consideration. From the molecular oxygen and ascorbic acid a labile adduct forms. This adduct can react with Co(II)glycylglycine in a fast reaction whereas the oxygen carrying complex and ascorbic acid form. The oxygen carrying complex having a higher activated oxygen oxidises the ascorbic acid with greater velocity than the molecular oxygen.

It is impossible to give a detailed mechanism because of the mentioned disturbing by-processes. According to our preliminary experiments the Co(II) glycylglycine complex can also catalyse the autoxidation of sulphite and the phenomena observed in the case of ascorbic acid appear in this system too. Further experiments with this and other simple redox systems will be carried out.

References

[1] Hearon, J.Z., D. Burk: J. Nat. Cancer Inst. 9, 337 (1949).

[2] Tanford, C., D.C. Kirk, M.K. Chantooni: J. Amer.Chem. Soc. 76, 5325 (1954)

[3] Beck, M.T.; Naturwiss. 45, 162 (1958).

[4] Csürös, Z., K. Zech, T. Pfliegel: M.T.A. Kém.Tud.Oszt. Közl. 1, 41 (1952).