

Characterization of Silver Anthranilate, a Promising Antibacterial Agent

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SUMMARY. The infrared spectrum of the $\text{Ag}(\text{NH}_2\text{C}_6\text{H}_4\text{COO})$ complex was recorded and briefly discussed in relation to its structural characteristics. The solubility of the compound in water, at 20 °C, was determined by conductimetric measurements. For comparative purposes similar measurements were also performed with the Ag(I) complexes of saccharin and sulfadiazine. The results suggest that the anthranilate and saccharinate complexes may be useful as slow silver releasing systems in pharmaceutical formulations.

RESUMEN. "Caracterización de antranilato de plata, un promisorio agente antibactericida". Se registró el espectro de infrarrojo del complejo $\text{Ag}(\text{NH}_2\text{C}_6\text{H}_4\text{COO})$ y se lo discutió brevemente en base a sus características estructurales. La solubilidad del compuesto en agua, a 20 °C, se determinó mediante medidas conductimétricas. Con fines comparativos se realizaron mediciones similares con los complejos que la Ag(I) forma con la sacarina y la sulfadiazina. Los resultados sugieren que los complejos de antranilato y de sacarinato podrían ser útiles como sistemas de liberación lenta de plata en formulaciones farmacéuticas.

INTRODUCTION

As part of a research project devoted to the synthesis and characterization of new metallopharmaceuticals, we are investigating the properties of different silver salts as potential antibacterial agents.

The microbiological activity of silver has been recognized for many centuries and the absolute minimum inhibitory concentration (MIC) of ionic Ag(I) is remarkably low at less than 0.1 µg/ml. The use of silver nitrate and colloidal silver in wounds dates from the mid-nineteenth century and the introduction of silver nitrate for the treatment of infantile blindness (*ophthalmia neonatorum*) in 1884 was considered a great medical breakthrough at that time. However, the development of silver complexes has not attained such a high grade of diversification as those of mercury compounds ¹.

Up to now, the most important and widely used Ag(I) complex is silver sulfadiazine, which was introduced in 1968 for burn treatment ², and which demonstrates a very effective activity especially against numerous strains of *Pseudomonas aeruginosa* and, consequently, won rapid acceptance from medical and drug agencies. It is dispensed in the form of a 1% cream in a water-soluble ointment base.

Baenziger and Strauss solved the crystal structure of this complex in 1976 ³ confirming its polymeric nature, with the Ag(I) cation coordinated to three N-atoms and one O-atom whereas a second Ag(I) ion is very close to it (Ag-Ag distance of only 2.916 Å), generating a distorted trigonal bipyramidal coordination around each of the cations. The polymeric nature of the complex renders it almost insoluble in water and in organic solvents and this prop-

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erty is in fact critical to its pharmacological activity. It is now widely accepted that the active species is actually Ag(I), generated by slow dissociation of the polymer. Because of the insoluble nature of the complex a controlled release of silver takes place at a moderate rate producing a kind of "reservoir" of silver ions ¹.

Taking into account the characteristics of this drug we are searching similar insoluble polymeric Ag(I) complexes that may have potential antibacterial activity. In this paper, we present the results of our investigation of the general physicochemical properties of the complex which Ag(I) forms with anthranilic acid (2-amino benzoic acid), a compound which presents some similarities with the sulfadiazine complex. On the other hand, some comparisons with the recently described Ag(I)-saccharinate are also made.

MATERIAL AND METHODS

3.5 g (25.5 mmol) of anthranilic acid were reacted with 40 ml of an aqueous solution, containing 1.02 g (25.5 mmol) of NaOH and heated at 65-70 °C. To this solution, 25 ml of another aqueous solution containing 4.33 g (25.5 mmol) of AgNO₃ was dropwise added, under continuous stirring. Silver anthranilate precipitates immediately in the form of a white flocculent product. It was separated by filtration and washed several times with hot distilled water. Its composition was confirmed by elemental chemical analysis: [Found: C, 34.45; H, 2.55; N, 5.66%. Calculated for C₇H₆O₂N Ag: C, 34.50; H, 2.43; N, 5.72%] and by X-ray powder diffractometry, analyzing the powder diagram on the basis of the published single crystal data ⁴ by means of the program POWDERCELL⁵.

Silver sulfadiazine was prepared by mixing very diluted equimolecular boiling ethanolic solutions of AgNO₃ and sulfadiazine (2-sulfanilamidopyrimidine). After slow cooling of the solution, an abundant crystalline precipitate is obtained ³.

Silver saccharinate was obtained by mixing equimolecular hot aqueous solutions of AgNO₃ and sodium saccharinate ⁶. After cooling the solution to room temperature, transparent needle-shaped crystals were obtained.

The solubility of the three compounds was investigated conductimetrically ⁷, working with saturated aqueous solutions at 20 °C and using a commercial conductimeter (L.PuSl, Munich, Type L 201) with a standard conductimetric cell (cell constant = 1.00/cm).

The infrared spectra of silver anthranilate were recorded with a Perkin Elmer 580 B spectrophotometer in the 4000-500 cm⁻¹ region, using the KBr pellet technique. Low-frequency (500-50 cm⁻¹) FTIR spectra were obtained on samples dispersed in polyethylene discs, on a Bruker 113v instrument. Unfortunately, Raman spectra of this complex could not be obtained as it decomposes rapidly in the laser beam, even if spectra were recorded at low temperatures. Attempts have been made using either the solid state Nd:YAG laser (1064 nm) with the FRA 106 Raman accessory of a Bruker IFS 66 FTIR instrument, or the 514.5 nm line of an Ar-ion-laser on a Jasco TRS-600 spectrometer.

RESULTS AND DISCUSSION

Crystal structure of silver anthranilate

The complex crystallizes in the monoclinic space group P2₁/n with two [Ag₂(C₇H₆NO₂)₂] dimers in the unit cell ⁴. The complex repeating unit comprises a distorted centrosymmetric bis-carboxylato-bridged dimer and each Ag(I) cation has a third bond to an amine nitrogen which extends the structure into a zigzag chain polymer. A plot of the structure, showing two sheets of chains, generated by the program POWDERCELL ⁵, on the basis of the published structural data ⁴ is shown in Fig. 1. As it can be seen, each Ag(I) cation is in the centre of a distorted trigonal plane (Ag-O bonds = 2.223(4) and 2.409(4) Å) and Ag-N bond = 2.301(5) Å). The Ag---Ag distance in the dimer is of only 2.9128(9) Å, whereas the Ag---Ag interdimeric separations are of 2.989(1) Å.

This structural analysis shows that a polymeric structure is present, but with a lower coordination of the Ag(I) cation than in the silver-sulfadiazine complex although similar short Ag--Ag interactions are observed in both cases.

Infrared spectrum

The IR spectrum in the most interesting spectral range, between 1700 and 500 cm⁻¹, is shown in Fig. 2. An approximate vibrational assignment is presented in Table 1. It is mainly based on general spectral data reported in standard references ⁸⁻¹⁰ as well as on recently studies on related complexes ^{11,12}.

In the higher frequency region, the stretching bands of the -NH₂ group clearly dominate the spectrum. In the free acid these vibrations ¹² are found at 3325 and 3240 cm⁻¹, and they are displaced to higher frequencies after coordination, in agreement with the participation of this

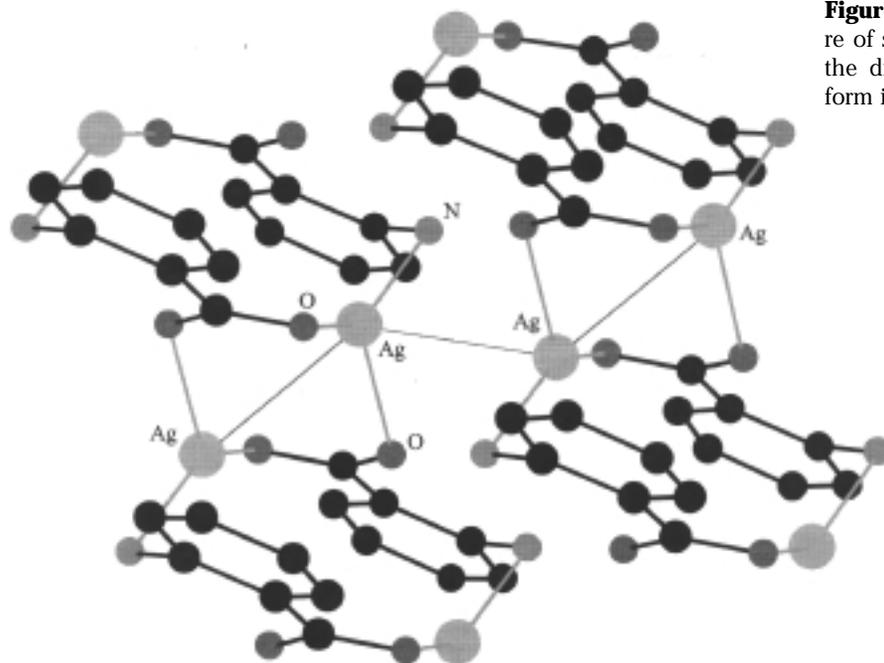


Figure 1. A view of the structure of silver anthranilate showing the dimeric units combined to form infinite polymeric chains.

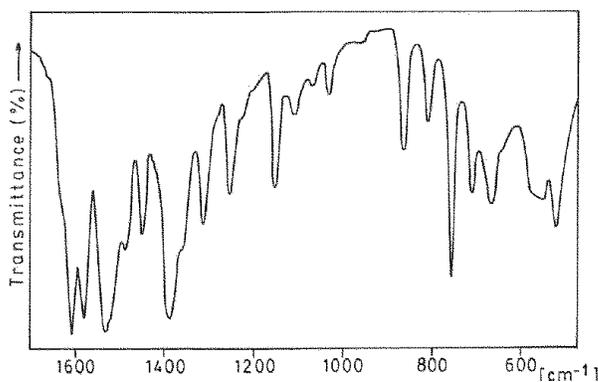


Figure 2. IR spectrum of silver anthranilate between 1700 and 500 cm^{-1} .

group in bonding. Other characteristic NH_2 -vibrations, such as the $\rho(\text{NH}_2)$ and the $\gamma(\text{NH}_2)$ modes located in the free acid at 1139 and 544 cm^{-1} respectively ¹², disappear after complexation. The characteristic scissoring deformation of this group, probably overlapped with one of the ring stretching modes, is identified as a very strong band at 1609 cm^{-1} .

The stretching vibrations of the C-H bonds are, as expected, relatively weak and not easy to identify. Some combination modes, probably of $\delta(\text{CH})$ vibrations, are found as very weak features between 1920-1750 cm^{-1} .

The behaviour of the carboxylate group also correlates with the structural peculiarities. Both stretching vibrations are seen as very strong IR bands. The energy difference between them ($\Delta\nu = 193 \text{ cm}^{-1}$) and the fact that both vibrations are

Band position (cm^{-1})	Assignment
3427 vs	$\nu_{\text{as}}(\text{NH}_2)$
3325 vs	$\nu_{\text{s}}(\text{NH}_2)$
3027 w	$\nu(\text{CH})$
1920 vw, 1860 w, 1750 vw	combination bands
1630 sh, 1609 vs	$\delta_{\text{sciss}}(\text{NH}_2) + \nu(\text{CC})_{\text{ring}}$
1582 vs	$\nu_{\text{as}}(\text{COO}^-)$
1532 vs, 1522 sh	$\nu(\text{CC})_{\text{ring}}$
1486 w, 1448 m	$\nu(\text{CC})_{\text{ring}} + \delta(\text{CH})$
1389 vs	$\nu_{\text{s}}(\text{COO}^-)$
1358 sh	
1312 m	$\nu(\text{C-CO}_2)$
1252 m	(C-N)
1151 m	$\nu(\text{CC})_{\text{ring}} + \delta(\text{CH})$
1109 w., 1067 vw	$\delta(\text{CH})$
1030 w	$\nu(\text{CC})_{\text{ring}}$
964 vw, 861 m, 807 m	$\delta(\text{CH})$
756 vs	$\delta(\text{CH})$
707 m	$\gamma(\text{C-NH}_2)$
664 m	$\delta(\text{CCC})$
576 m	
560 m	$\delta(\text{COO}^-) (?)$
523 m	

Table 1. Proposed assignment of the IR spectrum of silver anthranilate in the spectral range between 4000 and 500 cm^{-1} . [vs: very strong; m: medium; w: weak; vw: very weak; sh: shoulder].

close to the free ion values, is characteristic for carboxylate groups bridging two metal centers^{11,13}. The $\delta(\text{COOH})$ deformational mode, which is seen as a strong IR band located at 658 cm^{-1} in the free acid¹², is shifted after deprotonation and metal bonding. A medium intensity band found at 560 cm^{-1} in the complex has been tentatively assigned to this vibration.

For the ring a number of C–C and C–H vibrations could be confidently assigned as they clearly lie in the regions expected for an *ortho* substituted benzene ring⁸⁻¹⁰.

In the low frequency FTIR spectra a number of additional bands could be identified (472 (m), 435 (w), 419 (s), 330 (s), 304 (s), 235 (s), 173 (s), 146 (w), 121 (m) and 87(s) cm^{-1}) but they are not easy to assign. Those at the lowest frequencies are surely external (lattice) modes. In order to identify the position of the metal-to-ligand vibrations, which are also expected to lie in this spectral region, a comparison with the spectrum of the free acid was undertaken. This analysis showed that two bands present in the spectra of the complex (330 and 235 cm^{-1}) have not counterparts in the acid. Therefore, these bands are tentatively assigned to Ag–N and Ag–O stretching modes, respectively.

Solubility of silver anthranilate and comparison with other Ag(I) complexes

It was interesting to compare the solubility of the investigated complex with that of the pharmacological active silver sulfadiazine and with that of silver saccharinate, which also posses a

Compound	Solubility (g/100mL)	Solubility product constant
Ag-anthranilate	0.039	2.55×10^{-6}
Ag-saccharinate	0.019	4.29×10^{-9}
Ag-sulfadiazine	0.009	6.35×10^{-8}

Table 2. Solubility (in water and at 20 °C) and solubility product constant of silver anthranilate, compared with those of the corresponding saccharin and sulfadiazine complexes.

polymeric structure^{6,14}. The solubility determinations were performed conductimetrically at 20 °C in water. The results are presented in Table 2.

The results clearly show important solubility differences between the sulfadiazine and the other two complexes. Notwithstanding, the solubilities of the anthranilate and saccharinate complexes are also relatively low. This property, together with their polymeric nature, suggests that both complexes offer promising capacities for the slow release of silver in pharmaceutical preparations.

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