# THE FAR-INFRARED SPECTRA OF ALKALI METAL ION COMPLEXES WITH VALINOMYCIN, BEAUVERICIN, NONACTIN AND PERHYDROANTAMANIDE IN SOLUTION

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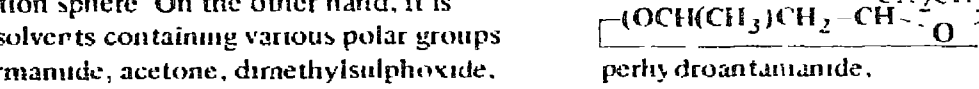
Received 25 December 1972

## 1. Introduction

The structure of membrane-active complexones of alkali metals (ionophores) and their complexes in solutions has been studied by a variety of spectral methods and theoretical approaches [1–5]. However, the results thereby obtained deal, as a rule, with the organic moiety of the molecules, and to a lesser extent with the metal ion's immediate environment, i.e. the internal coordination sphere. On the other hand, it is known that solvents containing various polar groups (dimethylformamide, acetone, dimethylsulphoxide, tetrahydrofuran etc.) form solvated complexes with alkali metal ions, whose metal–ligand linkages give rise to absorption bands in the far infrared (IR) region ($Li^+ \sim 410\ cm^{-1}$, $Na^+ \sim 200\ cm^{-1}$, $K^+ \sim 150\ cm^{-1}$, $Rb^+ \sim 120\ cm^{-1}$ and $Cs^+ \sim 110\ cm^{-1}$) [6, 7]. As the forces responsible for the ion binding by the ligands of macrocyclic complexones and for the ion–solvent interaction are basically of the same nature, it should be expected that far IR spectroscopy will be fruitful in studying the structure of the complexes in question. For example, this technique has been recently used for studying $Na^+$ and $K^+$ complexes with dibenzo-18-crown-6 [8].

In this work the IR spectra are studied in the 500–60 $cm^{-1}$ region of valinomycin [-(D-Val–L-Lac–L-Val–D-HyIv)–]$_3$, beauvericin, [-(L-MePhe–D-HyIv)$_3$–], nonactin,



perhydroantamanide,

[-L-Val-L-Pro-L-Ala-L-Cha-L-Cha-L-Pro-L-Pro-L-Cha-L-Cha-], and their complexes with alkali metal cations (Lac = lactic acid, HyIv = α-hydroxyisovaleric acid, MePhe = *N*-methylphenylalanine, Cha = β-cyclohexylalanine).

To facilitate interpretation of the spectral data, all measurements were taken in chloroform, a neutral solvent which does not in practice compete with the macrocycle for the cation. Beauvericin [5] was chosen from the antibiotics of the enniatin group as its complexes dissolve much more readily than those of enniatins A, B or C. When studying perhydroantamanide [9] whose complexing ability and conformational characteristics resemble those of antamanide [10],

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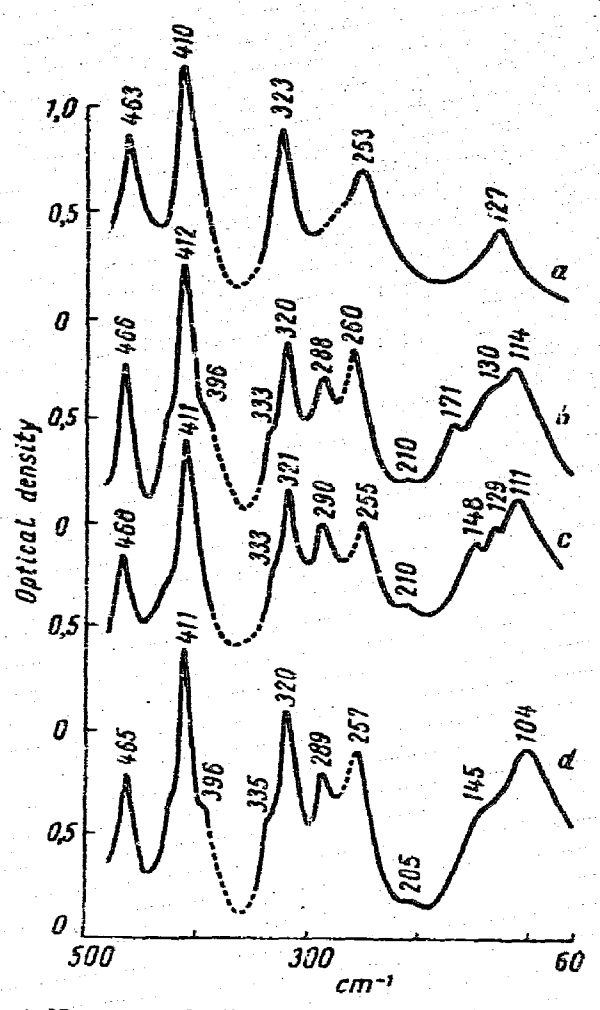


Fig. 1. IR spectra of valinomycin (a) and its complexes with $K^+$ (b), $Rb^+$ (c) and $Cs^+$ (d).

the higher solubility of its $Na^+$ complex in chloroform was also taken into account.

## 2. Materials and methods

Use was made of biosynthetic samples of valinomycin and nonactin. Beauvericin [5] and perhydroantamanide [9] were prepared by total synthesis. The complexes of the macrocycle and a 2–5-fold excess of the respective salt (KNCS, $KClO_4$ and KCl for valinomycin, KNCS and KCl for beauvericin, and rhodanides in the case of other complexes) were dissolved in methanol, the solution was evaporated, the precipitate dried in vacuum and treated with absolute chloroform, and the excess of the salt was filtered off.

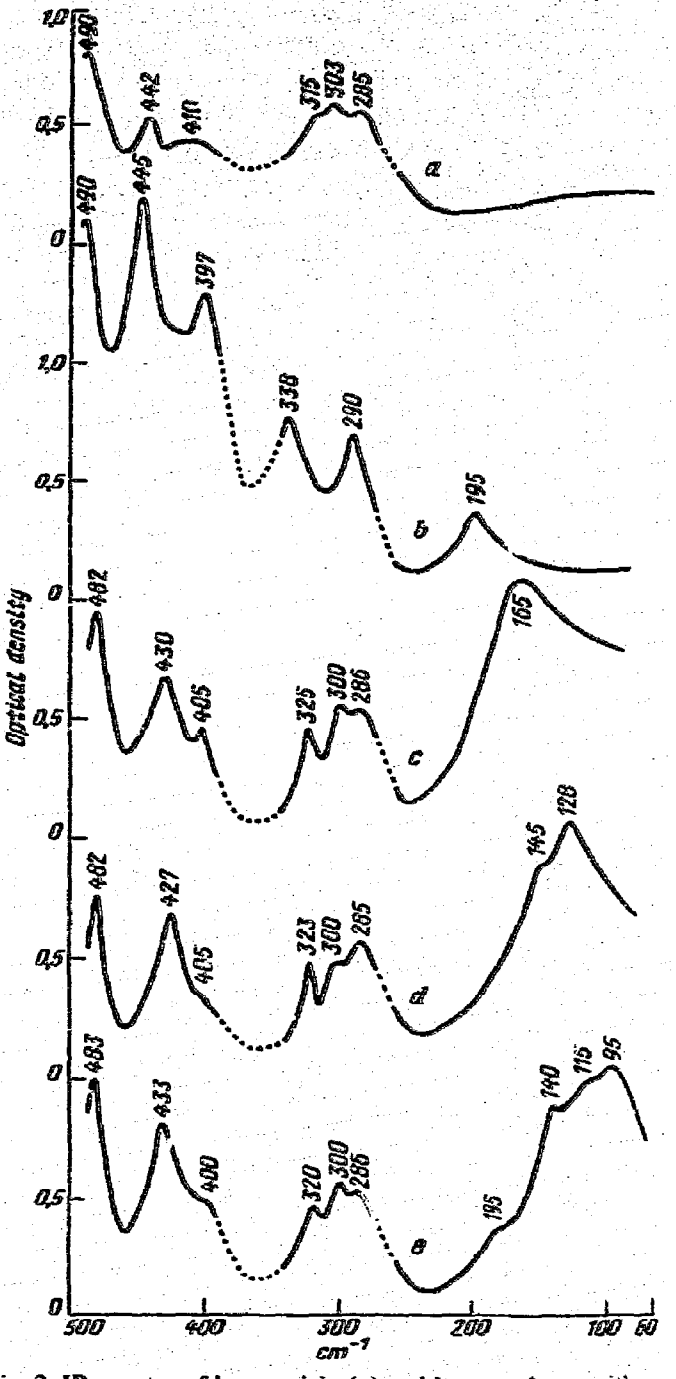


Fig. 2. IR spectra of beauvericin (a) and its complexes with $Li^+$ (b), $Na^+$ (c), $K^+$ (d) and $Cs^+$ (e).

Complex formation was monitored by the absence of absorption bands of the free macrocycle in the near IR region. The far IR spectra were measured in a FIS-21 single beam spectrophotometer (Hitachi) using ~0.2 M solutions. Vibrations of alkali metal ions

Table 1
$M^+...O$ stretching frequencies ($cm^{-1}$).

| Compound | Cation | | | | |
|---|---|---|---|---|---|
| | $Li^+$ | $Na^+$ | $K^+$ | $Rb^+$ | $Cs^+$ |
| Valinomycin | | | 171, 114 | 148, 111 | 145, 104 |
| Beauvericin | 445, 397 | 165 | 145, 128 | | 195, 140, 115, 95 |
| Nonactin | | | 150 | | 142, 102 |
| Perhydroantamanide | | 205 | | | |

were measured with an accuracy of ±4 $cm^{-1}$ and other bands with an accuracy of ±2 $cm^{-1}$. The instrument was calibrated by water vapour spectra.

### 3. Results and discussion

A comparison of the spectra of the free compounds with those of their $Na^+$, $K^+$ or $Cs^+$ complexes (figs. 1–4, the broken line corresponds to the absorption range of the solvent) has shown the latter to have new absorption bands at 104–205 $cm^{-1}$, which are ascribed to the metal–ligand bonds (table 1). The stereochemical properties of the ionophores studied prevent the cation from interacting with the anion [4, 10–12]. Hence, the spectral pattern in the region studied, unlike that of the dibenzo-18-crown-6 complexes [8], should not depend on the nature of the anion (except for the 462–480 $cm^{-1}$ band corresponding to the deformation vibrations of $NCS^-$); this has been demonstrated to be the case for $NCS^-$, $ClO_4^-$ and $Cl^-$, with the valinomycin·$K^+$ complex, and for $NCS^-$ and $Cl^-$ with beauvericin·$K^+$. With beauvericin·$Li^+$ the $Li^+...O$ vibrations overlap with the deformation vibrations of the depsipeptide skeleton. In accordance with the reported evidence [6, 13], the former are assumed to have 445 and 397 $cm^{-1}$ frequencies. Such an assumption is also favoured by a pronounced increase in the absorption intensity at 470–370 $cm^{-1}$ of the complex in comparison with the free macrocycle. The band at ~195 $cm^{-1}$ is to be assigned to the deformation vibration of the $O...Li^+...O$ angle. For most compounds the region of skeletal vibrations (> 250 $cm^{-1}$) noticeably changes its pattern on complex formation, i.e. the bands acquire more distinct contours and become more intensive, which testifies to the conformational homogeneity of the complexes. In the spectra of valinomycin, perhydroantamanide and their complexes there are also bands of stretching vibrations of the H-bonds at 130–104 $cm^{-1}$.

The most interesting feature of the spectra is that several $M^+...O$ bands are generally present; this was not the case in [6, 7], where only one band was reported whatever the nature of the anion. In accordance with the selection rules, the number of vibrations active in the IR spectra of coordination compounds is determined by the geometry of the inner

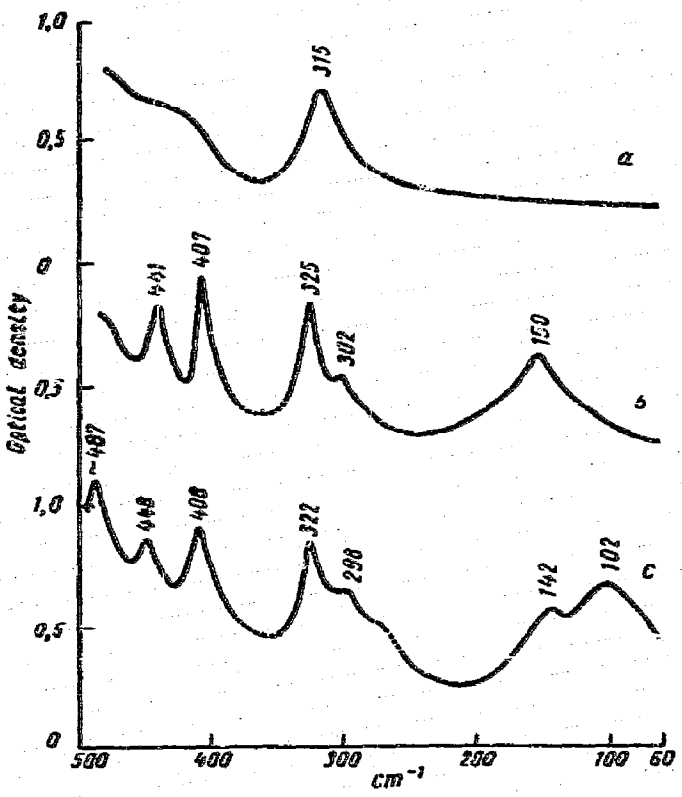


Fig. 3. IR spectra of nonactin (a) and its complexes with $R^+$ (b) and $Cs^+$ (c).

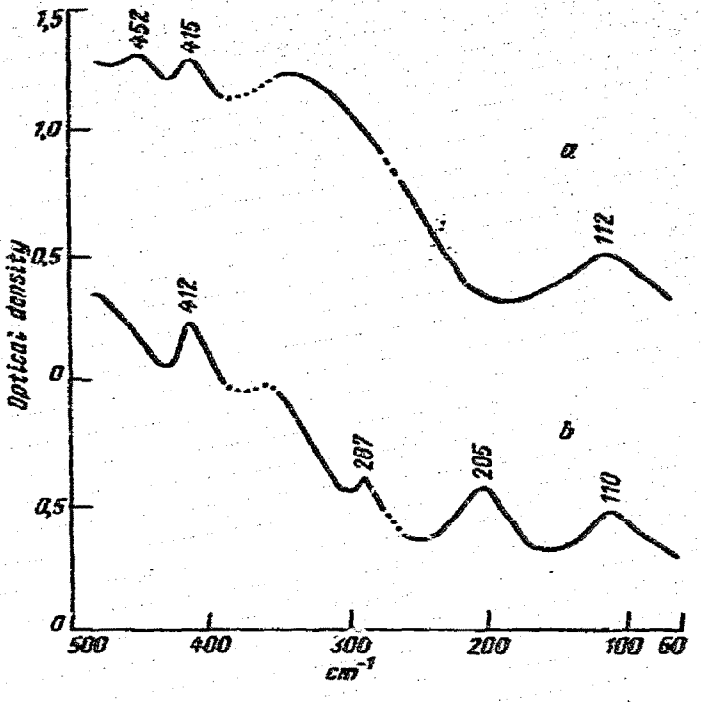


Fig. 4. IR spectra of perhydroantamanide (a) and its $Na^+$ complex (b).

coordination sphere. For example, one band in the metal–ligand vibration region should mean a highly symmetrical arrangement of the equivalent ligand atoms. This seems to be the case for the solvation shell of alkali metal ions in the solvents mentioned in the Introduction. In the complexes studied by us, participation of the ligand groups in the macrocyclic system considerably restricts their orientation with regard to the complexed ion and results in the formation of less symmetrical complexes producing several bands in the corresponding regions of the IR spectra. For example, the presence of two bands in the spectra of valinomycin complexes with $K^+$, $Rb^+$ or $Cs^+$ rules out the possibility of an octahedral coordination (local symmetry group $O_h$) of ester carbonyl oxygens forming the internal coordination sphere [4, 11] and suggests a trigonal antiprism type of coordination (symmetry group $D_{3d}$). Taking into account the NMR $^{13}C$ data [3], the more complex IR spectra may be explained by participation of amide carbonyls; however, rather large $M^+ \ldots O$ distances (> 4.0 Å, judging by molecular models) mean that the respective $M^+ \ldots O$ values should be out of the region studied ($< 100\ cm^{-1}$).

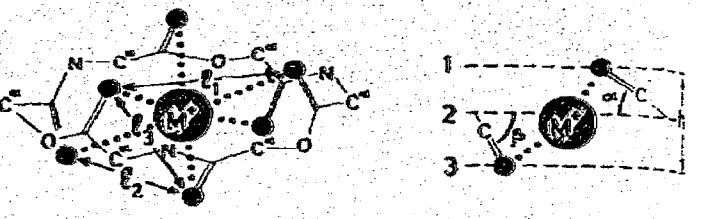


Fig. 5. Model of the depsipeptide skeleton of beauvericin complexes with alkali metal ions. 1, plane of esteric carbonyl O atoms; 2, plane of amide O atoms; 3, the average plane of $C^\alpha$ atoms; α, inclination angle of esteric carbonyls to plane 3; β, inclination angle of *N*-methylamide carbonyls to plane 3; *h*, distance between planes 1 and 2.

The IR spectra of the beauvericin complexes are interesting in that the number of bands depends upon the size of the cation: the $Na^+$ complex gives one band (although rather wide); $K^+$ and $Li^+$, two; and $Cs^+$, four. This surprising phenomenon was explained with the help of a geometrical analysis of the P type conformation of beauvericin which is characteristic for enniatin complexes [1, 11, 15]. The relevant calculations were made assuming bond lengths and bond angles as in [15] and *N*-methylamide and esteric bonds to have planar *trans* configuration ($\omega = 180 \pm 10^\circ$, for conformational nomenclature of peptides see [16]). The radius of the cation encaged in the internal cavity ($r_{M^+}$) was varied from 0.65 to 1.65 Å. It has been found that in a general case the centres of the oxygen carbonyl atoms form a distorted antiprism whose bases are unequal regular triangles with sides $l_1$ and $l_2$ lying in parallel planes (fig. 5, symmetry group $C_{3v}$). With certain $\phi$ and $\psi$ values the triangles become almost equal ($l_1 \approx l_2$, a trigonal antiprism, symmetry group $D_{3d}$); there are also structures with the carbonyl oxygens forming a subequilateral octahedron ($l_1 \approx l_2 \approx l_3$, symmetry group $O_h$). In conformity with the selection rules the above structures should have four, two and one $M^+ \ldots O$ frequencies, respectively. Summing up, the conformational transition of the beauvericin molecule in the complexes with $Li^+$, $Na^+$, $K^+$ and $Cs^+$ may be visualized in the following way: in the $Li^+$ complex the ligand oxygens form a flattened antiprism ($l_1 \approx l_2 > l_3$) which then becomes an octahedron ($Na^+$) and in the $K^+$ complex turns into an antiprism (flattened or stretched, $l_1 \approx l_2 \neq l_3$). In the $Cs^+$ complex the required dimensions of the cage are mainly achieved at

Table 2
Calculated geometrical parameters of beauvericin complexes with alkali metal ions*.

| $r_{M^+}$ | Amino acid residue | | Hydroxy acid residue | | h, (Å) | $l_1$, (Å) | $l_2$, (Å) | $l_3$, (Å) | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | $\phi$ | $\psi$ | $\phi$ | $\psi$ | | | | | | |
| 0.68 ($Li^+$) | − 65 ± 5 | 180 ± 5 | 60 ± 5 | −175 ± 5 | 1.9 ± 0.1 | 3.3 ± 0.2 | 3.3 ± 0.2 | 2.8 ± 0.1 | 35 ± 5 | 40 ± 5 |
| 0.98 ($Na^+$) | − 70 ± 10 | 165 ± 5 | 70 ± 10 | −170 ± 10 | 2.5 ± 0.2 | 3.5 ± 0.2 | 3.5 ± 0.2 | 3.4 ± 0.2 | 50 ± 5 | 50 ± 5 |
| 1.33 ($K^+$) | − 90 ± 10 | 145 ± 10 | 90 ± 10 | −150 ± 10 | 3.0 ± 0.3 | 4.3 ± 0.3 | 4.3 ± 0.3 | 3.9 ± 0.3 | 62 ± 5 | 66 ± 5 |
| 1.67 ($Cs^+$) | −103 | 171 | 74 | −136 | 2.8 | 3.5 | 5.0 | 3.6 | 46 | 74 |

* Computation of geometrical parameters was carried out as described in [19].

the expense of rotation of the *N*-methyl amide group, so that $l_1 < l_2$. The geometrical parameters of the beauvericin complexes fitting this model are listed in table 2; the $\phi$ and $\psi$ values of the $Cs^+$ complex are assumed to be the same as for the potential energy minimum of free cyclodepsipeptide [15]. Such conformational transition was previously reported [1] from an NMR study of (tri-*N*-desmethyl)-enniatin B, but no evidence for the symmetry of ligands in the complexes was offered**.

An X-ray analysis of the $K^+$ complex of nonactine has shown that its cation coordinated eight oxygen atoms located in the apexes of a cube (symmetry group $O_h$) [12]. Accordingly, there is just one vibration in the IR spectrum (fig. 3, table 1). As to the $Cs^+$ complex of nonactin, there are two metal–ligand vibrations in its spectrum, the fact indicating a disarrangement in the cubic coordination as the inner cavity grows in size. This may well account for the lower stability of the $Cs^+$ complex [18]. The data on the beauvericin and nonactin complexes testify to the fact, that the number of bands in the IR spectra of the macrocyclic complexones is chiefly determined by the geometry of the internal coordination sphere and does not actually depend on the nature of the functional groups carrying the oxygen ligand atoms (*N*-methylamide or ester carbonyls, ether groups). This, rather an unexpected, conclusion certainly requires additional experimental support.

It is known about the cyclopeptides of the antamanide group, that in their $Na^+$ complexes only two carbonyl groups have a direct contact with the central ion, whereas all the other oxygen atoms are located at greater distances [3, 10], so that their interaction with the metal ions should not contribute to the 300–100 $cm^{-1}$ region. The O...$Na^+$...O system has the $D_{\infty h}$ symmetry group and is to produce one band in the far IR region, which proved to be the case (fig. 4). On the other hand, in the case of antamanide and its analogues, water molecules seem to be involved in the coordination (fig. 6) [17] and the group of symmetry $D_{4h}$ thereby realized also has one vibration active in the IR spectra. When choosing between two above types of structures of the $Na^+$ complex of perhydroantamanide, attention should be paid to the Raman spectra which are being studied now.

Thus, the study of the far IR spectra of the macrocyclic compounds complexes with alkali metal ions furnishes valuable information about their structure. A more detailed analysis, involving an estimation of

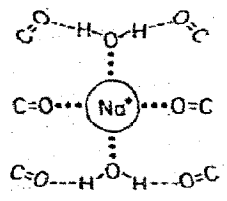


Fig. 6. Schematic representation of participation of water in antamanide complexes.

** This paper having been prepared, we found by NMR followed salt titration that enniatin B gives appreciable amounts of 2:1 complexes with $K^+$ and $Cs^+$ (but not $Li^+$ or $Na^+$) in methanol and dimethyl sulphoxide. The relevance of these findings to the present work is now under study.

force constants for the metal–oxygen linkages, may be of interest for elucidating their nature.

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