From the culture liquid of mutants 47 and 48 of Act. olivovariabilis [1, 2] we have isolated a yellow crystalline substance with the composition C_{14}H_{12}O_{7} soluble in lower alcohols, acetic acid, and chloroform, and we have called it olivovarin (I).

The IR spectrum of (I) shows absorption bands that could be assigned to the stretching vibrations of hydroxy groups, of a carboxy group and of quinone carbonyls bound by hydrogen bonds.

The acetylation of olivovarin with acetic anhydride in pyridine formed a triacetyl derivative (II). The IR spectrum of the latter showed absorption bands indicating the aromatic and aliphatic nature of the acetylated hydroxyls. The NMR spectrum of (II) had three three-proton singlets, two of them (at 2.33 and 2.34 ppm) belonging to the protons of an acetoxy group in an aromatic nucleus and one (at 1.90 ppm) to the protons of an acetoxy group in a side chain.

Methylation with diazomethane gave the ethers (III and IV) the IR spectra of which showed the absorption band of the carbonyl of an ester grouping. The NMR spectra of these derivatives showed the presence in each of them of one methoxy carbonyl group (three-proton singlet at 4.08 ppm) and of aromatic methoxy groups: one in the ether (III) (three-proton singlet at 3.62 ppm) and two in the ether (IV) (three-proton singlets at 3.58 and 3.33 ppm).

The acetylation of (III) gave the diacetate of the dimethyl derivative of olivovarin (V); its IR spectrum lacked the absorption band of a hydroxy group. The NMR spectrum (V) showed, in addition to the signals of the protons of methoxy and methoxycarbonyl groups (singlets at 3.55 and 4.01 ppm) two three-proton singlets of acetoxy groups: in an aromatic nucleus at 2.32 ppm and in an aliphatic chain at 1.88 ppm. Thus, the olivovarin molecule contains one carboxy and three hydroxy groups two of which are phenolic.

The olivovarin molecule has three aromatic protons located on neighboring carbon atoms; they give characteristic one-proton signals in the NMR spectrum of (V): quartets at 7.23 ppm, \( J_{1,6} = 8 \text{ Hz}, \ J_{1,5} = 2 \text{ Hz} \) (H\(_1\)), and at 7.90 ppm, \( J_{5,6} = 8 \text{ Hz}, \ J_{5,7} = 2 \text{ Hz} \) (H\(_2\)), and also a triplet at 7.60 ppm, \( J_{6,5} = J_{6,7} = 8 \text{ Hz} \) (H\(_3\)). The oxidation of olivovarin with an alkaline solution of potassium permanganate gave hemimellitic (benzene-1, 2,3-tricarboxylic) acid,* which is possible only if (I) contains an aromatic nucleus with three unsubstituted carbon atoms and three C–C substituents on adjacent carbon atoms.

Analysis of the structure of the signals of the aliphatic protons (two two-proton doublets at 2.54 and 2.85 ppm and a one-proton quintet at 5.40 ppm) show that the (V) molecule contains the fragments \(-\text{CH}_2-\text{CH(OAc)}-\text{CH}_2-\) attached to carbon atoms not bearing protons.

Olivovarin reacts with o-phenylenediamine to form a quinoxaline derivative (VI), which shows the presence of an o-quinone grouping in it.

*The compound was identified by a comparison of the mass spectra of the methyl esters of the acid obtained and of an authentic sample of hemimellitic acid.
The facts given above show the presence of the following groupings in the olivovarin molecule:

These groupings include 12 carbon atoms out of the 14 present in the molecule of \( (I) \). The other two carbon atoms are bound to the two phenolic hydroxyls and are included in an o-quinone system. The latter condition is satisfied by two possible structures: \((Ia)\) and \((Ib)\).

In the NMR spectrum of the quinoxaline derivative \((VI)\), the two protons at \( C_3 \) gave separate one-proton quartets at 3.16 ppm, \( \delta J = 12 \text{ Hz}, J_{H_3',H_2'} = 9 \text{ Hz} \) \((H'_{3})\) and at 3.62 ppm, \( \delta J = 12 \text{ Hz}, J_{H_3',H_2'} = 4.8 \text{ Hz} \) \((H''_{3})\), which is possible only if the methylene group and the quinoxaline nucleus are spatially close, i.e., the side chain and one of the quinone carbonyls \((I)\) are in the peri position; this arrangement is found only in structural formula \((Ia)\).

The correctness of this conclusion was confirmed by the IR spectra of olivovarin and its derivatives in the 1600-1700 cm\(^{-1}\) region (Fig. 1). In the molecule of \((I)\), both quinone carbonyls are bound by hydrogen bonds, as is shown by the presence in the IR spectrum of olivovarin of only one absorption band, at 1625 cm\(^{-1}\), and by the increase in the frequency of this band to 1675 cm\(^{-1}\) in compound \((V)\) which lacks free hydroxy groups. In the trimethyl ether the OH group at \( C_2 \) remains unsubstituted, i.e., the formation of only one hydrogen bond, \( C_3=O \ldots HO^- \), is possible, as is shown by the two absorption bands in the IR spectrum: of free (1650 cm\(^{-1}\)) and of bound (1610 cm\(^{-1}\)) quinone carbonyls. Similar changes can be found in a comparison of the spectra of olivovarin and its anhydro derivative \((VII)\) which lacks an OH group at \( C_2 \). In this case, the spectrum of \((VII)\) again has the absorption bands of free \((C_3=O)\) and of hydrogen-bond-bound \((C_2=O \ldots HO^-)\) quinone carbonyls.

It follows from a combination of the facts given that olivovarin is \( 8-(1'-\text{carboxy}-2'-\text{hydroxypropyl})-3,4\)-dihydroxy-1,2-naphthoquinone. The results of the distillation of olivovarin with zinc dust agree with the structure proposed for it: In the reaction products we isolated and identified naphthalene, 1-methyl-naphthalene, and phenanthrene. The formation of phenanthrene may be represented as the result of the cycle...