A NON-CENTROSYMMETRIC POLYMORPH OF 5-HYDROXY-7-METHOXY-2-PHENYLCHROMAN-4-ONE

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ABSTRACT

The bioactive compound 5-hydroxy-7-methoxy-2-phenylchroman-4-one (pinostrobin, I), was isolated from aerial parts of Nolana ramossissima I.M. Johnst., a Chilean endemic species, using a combination of medium pressure column chromatography and high speed countercurrent liquid-liquid chromatography (HSCCC). The compound crystallized as a non-centrosymmetric polymorph in the orthorhombic chiral space group P212121. Its structure had been previously reported by Shoja (Acta Cryst. C45, 828, 1989) and Yamovoi et al. (Khim. Prir. Soedin (5), 361, 2001) in the centrosymmetric space groups Pbc a and P21/c, respectively.

INTRODUCTION

High speed countercurrent chromatography (HSCCC) is a support-free liquid-liquid partition chromatographic technique which eliminates the problem of the adsorption of a sample onto a solid support and has been widely used for the purification of natural products, including flavanones1. In this work this fast technique has been applied in combination with medium pressure column chromatography for the purification of the phenolic constituents in an organic extract of aerial parts of Nolana ramossissima I.M. Johnst. (Solanaceae), an endemic species of Paposo Valley, Northern Chile.

The main compound isolated, 5-hydroxy-7-methoxy-2-phenylchroman-4-one, synonym: pinostrobin is an important biologically active flavanone previously isolated from different plant materials. This flavanone is a constituent of Pinus strobus and many other pine species and, inter alia, Artemisia campestris, Boesenbergia pandurata, Boesenbergia rotundata, Sar candra glabra, and Polygonum lapathifolium, and has been identified also in honey2, propolis3, and trees frequently visited by bees such as Alnus spp. and Populus spp. Pinostrobin has shown antileukemic4, antioxidant5 and anti-bacterial activity6 and inhibits TNF-α and IL-1β production7. Pinostrobin is an inducer of mammalian phase 2 chemoprotective and antioxidant enzymes7 and an inhibitor of DNA topoisomerase I activity8. In our hands the title compound crystallized as a new non-centrosymmetric polymorph. The existence of polymorphic forms provides a unique opportunity for the investigation of structure-property relationships, since by definition the only variable among polymorphs is that of crystal structure, and one of the most effective strategies for studying structure-property relationships has been to follow the behavior of a physical property through a polymorphic phase change.

EXPERIMENTAL

General

Nolana ramossissima was collected in Paposo, Antofagasta, Chile in September 2011. A voucher herbarium specimen is deposited in the laboratory of Natural Products, University of Antofagasta, with the number 110403. Dried and finely powdered aerial parts of N. ramossissima (1562 g) were extracted with petroleum ether (2 L, 3 times) in the dark, 24 h each time). After evaporation of the solvent in vacuo at 35 °C, a dark honey extract (35 g) was obtained. The extract (20 g) was submitted to medium pressure column chromatography (Silica Gel 60 H, 7 x 40 cm) using n-hexane:ethyl acetate 9:1 and 0.82 Å for methyl, methine and methylene groups and hydroxyl groups or 1.2 Ueq (C) for methine and methylene groups and oxygen atom were positioned stereochemically and were refined anisotropic thermal parameters was carried o...
### Table 1. Crystallographic data, details of data collection and structure refinement parameters for compound (II).

| Crystal data | V = 1311.9(5) Å³ |
|--------------|------------------|
| C₇₆H₇₂O₆    |                  |
| Mᵣ = 270.27  |                  |
| Orthorhombic, P₂₁2₁2₂ (Nº19) |                  |
| a = 9.129(18) Å |                  |
| b = 9.5946(19) Å |                  |
| c = 14.977(3) Å |                  |
| Density (calculated)/Mg.m⁻³ | 1.368 |
| Data collection | Enraf Nonius CAD4-MACH3 |
| 2456 measured reflections | Rint = 0.055 |
| 1752 independent reflections | 2.521 to 25.948° |
| Theta range for data collection (θ) | -2 ≤ h ≤ 11, -11 ≤ k ≤ 11, -18 ≤ l ≤ 18 |
| Refinement | R[F²] = 0.035 |
| wR(F²) = 0.090 | Flack’s parameter = 0.000 |
| S = 1.05 | Δρmax = 0.25 eÅ⁻¹ |
| Extinction coefficient 0.040(4) | Δρmin = -0.25 eÅ⁻¹ |

### RESULTS AND DISCUSSION

The title compound (I) is a new polymorph of structures II and III reported by Shoja and by Yamovoi et al. In the Shoja report, the compound crystallized in the orthorhombic space group Pbc’a Z = 8, cell parameters a = 10.127(2), b = 15.270(3), c = 8.226(2) Å, V = 1270.9(5) Å³, whereas the structure reported by Yamovoi et al. is in the monoclinic space group P2₁/c, a = 10.172(2), b = 16.079(2), c = 8.079(3) Å, β = 91.74(1)°, V = 1320.75 Å³. In our case compound I crystallized in the orthorhombic chiral space group P₂₁2₁2₂, Z = 4, a = 9.129(18), b = 9.5946(19), c = 14.977(3) Å, V = 1311.9(5) Å³. In all three polymorphic forms, the tetrahydropyran ring has skew-boat conformation (Q₁ = 0.455(6) Å, θ = 123.1(7)°, φ = 249.0(8)° mean, for three polymorphs) and the phenyl ring has an equatorial ω-orientation relative to the tetrahydropyran ring, Figure 1. In Figure 2 structures I and II are superimposed in capped stick fashion. The overlay of molecular backbones clearly shows the conformational similarity between homologous atoms, except for the hydroxyl hydrogen atoms of the hydroxyphenyl group, where the H atoms point up and down in I and II, respectively. Another important difference between both structures is the orientation of the phenyl ring with respect to the plane defined by tetrahydropyran ring atoms O₁-C₈-C₉-C₁₀-C₁₅ (RMSD = 0.043 Å and 0.018 Å for I and II, respectively), 85.56(15)° vs. 109.4(3)°. Similarly, the main difference between I and III is the orientation of the phenyl ring with respect to the plane defined by O₁-C₈-C₉-C₁₀-C₁₅ (RMSD = 0.043 Å and 0.015 Å for I and III, respectively), 85.56(15)° vs. 38.5(3)°, Figure 3. The bond angles and distances are normal and similar to those in structures II and III. Table 2. In the three polymorphs the molecular conformation is stabilized by one intramolecular hydrogen bond between the carbonyl O atom and the H atom of the hydroxyl group with graph-set motif S(6)⁶, but the geometry of this interaction is different for each polymorph (H₂O₂⋅⋅⋅O₄ = 1.893(2) Å, O₂⋅⋅⋅O₄ = 2.618(3) Å, O₁⋅⋅⋅H₂ = 146.64(18)° for I, H₂O₂⋅⋅⋅O₄ = 1.598(5) Å, O₂⋅⋅⋅O₄ = 2.574(7) Å, O₁⋅⋅⋅H₂ = 174.2(4)° for II, H₂O₂⋅⋅⋅O₄ = 1.79(6) Å, O₂⋅⋅⋅O₄ = 2.685(7) Å, O₁⋅⋅⋅H₂ = 160(6)° for III).

### CONCLUSIONS

The main conclusion of this work is that a new, non-centrosymmetric polymorph of 5-hydroxy-7-methoxy-2-phenylchroman-4-one (pinostrobin), was determined by X-ray methods, was isolated from an organic extract of aerial parts of Nolana ramossisima Johnst., an endemic species of the Paposo Valley, Northern Chile, using high speed countercurrent chromatography (HSCCC) and slow evaporation of a methanol solution.

### Supplementary material

CCDC-1019211 contains the supplementary crystallographic data for this paper. These data can be obtained free of charge from the Cambridge Crystallographic Data Centre via www.ccdc.cam.ac.uk/data_request/cif.
Figure 3: Superimposed structures for the title compound (red) and polymorph III (blue).

ACKNOWLEDGEMENTS

The authors thank FONDECYT (Chile) (Grant 1140178) for financial support.

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