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Testing of Octopirox against Pityrosporum
ovale (orbiculare) in vitro

Summary

Octopirox was tested against Pityrosporum ovale (orbiculare) in vitro in an agar dilution test. Octopirox exhibited marked growth inhibition of this yeast at 62.5 and 125 $\mu\text{g/ml}$ (3- and 6-day values). The value for Octopirox has to be taken as about 6 $\mu\text{g/ml}$ because this active ingredient is partially antagonized by the growth medium used (Raether and Hänel, mycoses:33,191(1990)).

Introduction

P. ovale has been discussed since 1874 as a cause of dandruff (Malassez). The controversy is still continuing although it is increasingly being recognized that Koch's postulates are met (Shuster 1988). The yeast species is always present in dandruff, is not always encountered in its absence and reappears with the complaint after artificial inoculation. It therefore seems reasonable to treat dandruff with antimycotics. This strategy is being followed by an increasing number of suppliers, though whether antimycotics such as ketoconazole (shampoo), which have marked side effects, are suitable remains doubtful.

The yeast Pityrosporum sp. is noted for some peculiarities that make testing for antimycotic activity in vitro difficult. The need to add lipids to the growth media precludes the use of the common test methods. We have developed an agar medium in which olive oil is employed as an additive.

We tested Octopirox, which is marketed as an "anti-dandruff agent", on this medium.

Material and methods

- Substances:** Octopirox pure substance
- Concentrations:** ($\mu\text{g/ml}$) 500, 250, 125, 62.5, 31.25, 15.63, 7.8, 3.9, 1.95, 0.97
- Material:** Petri dishes of 5 cm diameter, Sabouraud's agar + 3 % olive oil + 1 % Tween 80^R, pH 6.5.
7 ml agar + 0.5 ml preparation suspension in the corresponding concentration were used in each petri dish.
- Stock preparation:** 7.5 mg preparation + 0.5 ml DMSO + 0.5 ml distilled water. Further dilution of this stock solution gives the required concentration.
- Inoculation:** *P. ovale* was removed from the above-mentioned oil agar after 3 days' culture and suspended in physiological NaCl solution. 100 μl of this were dropped onto each plate and incubated at 37°C for 3 days.
- Evaluation:** The growth was evaluated daily and documented photographically on the 3rd day.

Results

The result for Octopirox is shown in figs. 1 and 6. It can be seen that Octopirox has a strong inhibiting action at about 125 $\mu\text{g/ml}$. The 6-day value is no different from the 3-day value and was therefore not included in the figures. Delayed growth of the fungus occurs as a result of the use of Octopirox. It can be seen that prolonged incubation of the fungi at 37°C leads to increased growth of the yeast deposit on the plates containing the preparation. The curve for the 3-day value is similar to the control curve.

The DMSO control (solvent) similarly shows slightly inhibited growth in the upper dilution range (corresponding to the 500 and 250 $\mu\text{g/ml}$ preparation concentrations) compared to the growth control. (Figs 1, 2 and 3).

Discussion

The result shows that Octopirox in the concentrations used inhibits yeast growth. Inhibition can only be assessed visually owing to the deposit-like growth of the yeast. In a parallel operation all plates were examined microscopically for the presence of the yeast (fluorescence microscope). The results confirm earlier investigations by Engst and Krempf-Lamprecht (1983), who assessed cyclopiroxolamine as the most effective of 5 antimycotics tested. Cyclopiroxolamine can be classified as almost equal in effectiveness on the basis of our own tests with Octopirox.

A special fact must be mentioned in using Sabouraud's medium. It is known that some hydroxypyridones are antagonized by a wide variety of media. Thus, the related Rilopirox on Sabouraud's agar has an approximately 10 times higher minimum inhibitory concentration when used against *Candida albicans* (Raether and Hänel 1990). This means that the ascertained value for Octopirox needs to be lowered several places. This underassessment is unavoidable in view of the need to use Sabouraud's medium. However, Octopirox, like Cyclopirox, is noted for excellent penetration into skin and cutaneous horn. As a result, even deep-seated yeasts were reached by the active ingredient. This would then result in fewer recurrences of dandruff.

We investigated a recently published liquid medium for these tests (Marcon et al 1987) but observed only very slight fungal growth.

In conclusion it can be said that Octopirox has strong in-vitro activity against *Pityrosporum ovale* (*orbiculare*).

Literature

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