

Antibacterial effect of substances from Cannabis indica L.

ZDENĚK KREJČÍ

INTRODUCTION

In the last few years, we have carried out a systematic survey of the flora of our climate zone at the Institute of Hygiene of the Faculty of Medicine of Palacký University in Olomouc for the content of substances with antibacterial effects. We have processed over 3,000 different plant species in this regard, some of which have shown relatively good antibacterial effects. And through this path of systematic research, we also came to Cannabis indica - Indian cannabis, which we subjected to thorough research. It was the discovery of this antibacterial effect in the extract of Cannabis indica, which was demonstrated several years ago at our institute, that sparked interest in cannabis in medicine and a whole series of works from individual institutes and clinics. The interest of clinicians in these substances was supported by very good experience with antibacterial substances from various plants, cited in world, especially Soviet literature.

I presented the preliminary results of this work as early as 1951 at the Association of Physicians in Olomouc and shortly afterwards published it in Lékařské listy. This paper addresses the issues of extraction of active substances with the aim of their isolation, and roughly evaluates the antibiotic effect.

I. INDICATIVE CHEMICAL AND BACTERIOLOGICAL EXPERIMENTS WITH CANNABIS INDICA (CI) EXTRACTS

a) Preparation of extracts

Extracts prepared in different ways using different solvents were tested for the presence of antibacterial substances by the modified Oxford method. The drug was subjected to Carlson extraction. In a weight ratio of 1:5, the individual parts of the plant (roots, stems, leaves, tops, seeds) were leached with ethyl alcohol, ethyl ether, physiological solution, sulphuric acid 1.5% and 1% acidic sodium carbonate, at higher, normal room temperature even at 0°C. From the beginning of the work, it was clear that the antibiotic principle of the drug is closely related to the content of its resinous substances, contained mainly in the leaves and in the cannabis tops of female inflorescences.

Organic solvents, especially ethyl alcohol, petroleum ether and benzene, proved to be the most suitable extraction substances.

We also tested the variety Cannabis sativa L. - hemp, commonly grown in our country, especially in Slovakia, for industrial processing into fibres for the content of active substances. This drug also contains antibacterial substances, but to a slightly lesser extent.

b) Bacteriological technique

The actual methodology for determining antibiotic power is very simple. We used common methods as we know it from practice to determine sensitivity to penicillin. A filter paper connected with an antibiotic is placed on the agar soil infected with the microbe being tested. The inhibition zone around the target after 24 hours of incubation is a measure of the effectiveness of the substance. To ensure standard conditions, especially the same number of microbes, we mostly carried out tests on bacteriological solid soils with inoculated microbes.

c) Spectrum of bacteria sensitive to Ci

From the bacteriological point of view, we were mainly interested in the spectrum of bacteria on which *Cannabis indica* extracts act in vitro. During the systematic investigation, we used representatives of gram-positive microbes *Staphylococcus pyogenes aureus haemolyticus* and *Escherichia coli* from the gram-negative microbes. Cannabis extracts had a significant bactericidal effect on *Staphylococcus aureus*, while *E. coli* proved to be insensitive. Based on this preliminary finding, we focused the whole work and proved that Ci extracts have a very good antibacterial effect on the following microbes: *Streptococcus alfa haemolyticus*, *Streptococcus beta haemolyticus*, *Diplococcus pneumoniae*, *B. subtilis*, *B. anthracis*, *Corynebacterium diptheriae* and *Corynebacterium cutis*, all gram-positive microorganisms. Particularly welcome was the finding of an effect on penicillin-resistant strains of *Staphylococcus aureus*.

This was one of the traits that put *Cannabis indica* at the forefront of our interest. We saw the possibility of using this antibiotic especially for local application in purulent diseases, without the danger of inducing resistant strains to another antibiotic, administered at the same time as a whole. It is also impossible to overlook the very good effect of Ci substances on *Staphylococcus aureus*, especially today, when a high percentage of staphylococcal diseases is beyond the possibility of penicillin treatment. Tests for other microorganisms, mainly from the gram-negative series, were mostly negative and thus limited the width of the antibiotic's action. On the other hand, our studies also confirmed the good effect of the isolated substance on *Mycobacterium tuberculosis*.

The advantage of our extracts is the possibility of accurately determining the effect of antibiotic and analgesic and then the fact that the intoxicating dangerous effect of oriental cannabis is missing.

If we overlook the main indications, we see that everywhere in folk medicine and among primitives I know the analgesic and antibiotic effect of cannabis, then the effect on the urogenital system, which was known even to ancient physicians and which official medicine has forgotten to the detriment of the sick.

SUMMARY

We examined the content of substances with antibacterial effects in 3,000 species of Central European flora. For more detailed research, we chose Indian cannabis – *Cannabis indica* – grown in Czechoslovakia.

We determined the most advantageous extraction methods and our in vitro experiments demonstrated the bactericidal effect of cannabis components on Gram-positive microorganisms: *Staphylococcus pyogenes aureus haemolyticus*, *Staphylococcus aureus* - resistant to penicillin, *Streptococcus beta haemolyticus*, *Streptococcus viridans*, *Pneumococcus* *Corynebacterium diptheriae*, and *Bacillus anthracis*. Gram-negative microorganisms of the typhoid-coli type were resistant similarly to *Pseudomonas aeruginosa* and *Proteus vulgaris*. It showed a great antibacterial effect on *Mycobacterium* in vitro even when diluted to 1:150,000.

A comparison of the bactericidal effect of isolated, amorphous and crystalline substances was performed and a thorough comparison of the sensitivity of two applied bacterial methods, the Oxford method and tests in liquid environment, was performed. We determined the limits of effectiveness at maximum dilution of the biologically active substance (1:100,000) and the speed of its action at different concentrations.

The influence of inactivating factors was studied in detail. Blood, plasma and serum had an inactivating effect, partially reducing their antibacterial effect.

Finally, a comparison of the effectiveness of these active substances with penicillin, streptomycin at different pH was carried out and an overview of cannabis preparations made for clinical application in dentistry, ENT, dermatology and phthysiology was elaborated.