

## A note on the claimed toxicity of *Cortinarius gentilis*

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The toxicity of *Cortinarius orellanus* group of mushrooms became apparent in the 1950ies. *C. gentilis* was considered toxic in Finnish mycological publications. The opinion was primarily based on the study by Möttönen et al. (1975) and on a case study by Hulmi et al. (1975), which papers were then cited in later publications. When the specimens on which the first-named study was based were rechecked, it turned out that the original material used for the rat feeding test by Möttönen with his co-workers was not adequately documented. In order to examine the possible toxicity of Finnish *C. gentilis* mushrooms, the present authors studied 28 samples of this species. An unspecific cell culture toxicity test and a feeding test on mice revealed no toxicity in *C. gentilis*.

Key words: *Cortinarius gentilis*, toxicity

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### Introduction

The toxicity of the fleshy fungus *Cortinarius orellanus* Fr. became apparent in 1952 when 102 people in Poland fell ill after consuming this fungus. In this outbreak eleven people died 4–16 days after the meal due to the acute renal failure (Wieland 1996). This severe outbreak brought fear for the toxicity of *Cortinarius* fungi and this genus of mushrooms was suddenly regarded by general public to be very toxic and not edible at all. Research work was directed to find out toxic *Cortinarius* species and to develop analytical toxicological methods (Tebbett & Caddy 1983, 1984). A surprisingly long latency period was reported, more than two weeks, between the ingestion of toxic *Cortinarius* mushrooms and the appearance of the clinical symptoms, which makes difficult to diagnose field cases. Schumacher and Høyland (1983) published a detailed documentation of *Cortinarius* poisonings. Keller-Dilitz et al. (1985) introduced a thin layer chromatography method to show the presence of orellanine, the

main toxic compound in *Cortinarius* mushrooms. In addition to *C. orellanus*, *C. speciosissimus* was soon found to contain orellanine as well.

Recent taxonomic and nomenclatural studies on *Cortinarius* have changed the name of *C. speciosissimus* and modern DNA techniques have brought new information on *Cortinarius* phylogeny (Høyland & Holst-Jensen 2000). *C. speciosissimus* is now called *C. rubellus* Cooke, synonyms: *C. speciosissimus* Kühner & Romagn. and *C. orellanoides* Rob. Henry (Brandrud et al. 1990). Its toxicity is well known and confirmed (Brandrud et al. 1990, Svendsen et al. 2002). *C. rubellus* has caused poisonings even to animals (Överås et al. 1979).

The aim of this paper is to re-evaluate the toxicity of *Cortinarius gentilis* (Fr.) Fr. The original mushroom material used in a Finnish animal experiment (Möttönen et al. 1975) was rechecked and a new toxicity study on *C. gentilis* was carried out.

## Literature review on toxicity of *Cortinarius gentilis*

*Cortinarius gentilis* is not reported to be toxic according to an extensive literature survey (Danel et al. 2001), except in the Finnish paper by Möttönen et al. (1975). Another Finnish publication (Hulmi et al. 1974) has been interpreted to support the toxicity of *C. gentilis*. We shall examine these two papers more closely. They have often been referred in international literature (Tebbet & Caddy 1984, Prast et al. 1988, Bresinsky & Besel 1985). Based on these papers many popular mushroom guides, at least in Finland, have considered *C. gentilis* toxic in addition to *C. rubellus* and *C. orellanus*.

Hulmi et al. (1974) described in a Finnish medical journal a case where four persons, viz. the parents, grandmother and an 8-year old son fell seriously ill after a meal prepared of wild mushrooms. A part of the mushrooms had been frozen fresh, another part boiled before freezing. Most mushrooms represented *Boletus*, but among them were some additional mushrooms. According to the authors they did not know what species were eaten and whether the studied mushrooms were boiled or not before freezing. They suggested in their abstract, written only in Finnish, that *Cortinarius speciosissimus* (i.e., *C. rubellus*) might have caused the poisoning. The paper describes the symptoms, the laboratory experiments, the treatments in hospitals and the follow-up of the patients up to one year. The father was most seriously ill, but recovered after a kidney transplant.

## *Cortinarius gentilis* sample used by Möttönen et al. (1975)

The results of the feeding experiment by Möttönen et al. (1975), are likely partly based on a wrong species identification or mixing of two specimens. The herbarium specimen of *C. gentilis* Fr., HH. No. 72, Kuopio, which Möttönen et al. referred in their paper, and which was used as feeding material for the rats in their experimental study, was found in the Kuopio Collection under two names. The name *C. speciosissimus* is written on the outer cover of that specimen and the name *C. gentilis* written on the inner cover of the same specimen. The material used for feeding experiment as *C. gentilis* mushrooms encompasses specimens of two species and is therefore not satisfactorily documented.

## Toxicity studies of *Cortinarius gentilis* by cell culture and feeding test on mice

### Mushroom samples

*Cortinarius gentilis* Fr. samples were collected in South Finland, Tammisaari, Raasepori, Repubacka, on sandy heath coniferous forest on 28th September 1990 (Mauri Korhonen 10004, H) and deep-frozen on the following day.

### Toxicity studies

The toxicity of the *Cortinarius gentilis* samples was studied by two methods. All mushroom samples were first studied by a method in which an extract from every single fruitbody was individually tested in a cell culture system. Altogether 28 *C. gentilis* samples were studied by this method, which can be regarded as an unspecific screening test for materials e.g. too small for feeding tests. The main toxicity test was a feeding experiment on mice for studying especially the possible effects on the kidneys.

### Cell culture toxicity test

The size of the *Cortinarius gentilis* fruitbodies varied. A sector of the cap weighing 1.3–3.0 g was cut for cell culture toxicity test. Small caps were used as whole, and the smallest samples in those cases weighed 0.6 g. The material was ground in a mortar and physiological salt water added; 3.0 ml of water to 1.3 g sample. The samples were kept for two hours at +4°C, centrifuged at 3200 r/min. The supernatant was filtered and studied on U-cell cultures according to the method described in Hintikka (1978). The death of the cells is regarded as indicator of toxic substance extracted from the original material. The detailed method of toxicity test is described by Nikulin and co-workers (Pasanen et al. 1993). All tests were carried out twice. Out of the 28 *C. gentilis* samples 26 were non-toxic and two showed slightly toxic regarded rather to be caused by the raw extract of the mushroom material, not by a specific toxin.

### Feeding test to mice

Five grams of mushroom materials combined from about five *Cortinarius gentilis* samples were ground and mixed in 25 g of mice diet and fed to a group of five mice. Each mouse was supposed to eat about one gram of the original *C. gentilis* material. There were three experimental groups with *C. gentilis* mushrooms and a control group. The mice weighed  $25 \pm 2$  g in the beginning of the experiment. After the mice had finished their mushroom feeds the diet turned to the former mice ration. Animals had free access to water. The mice showed no symptoms for disease. After seven days they were killed. The *post mortem* examinations revealed normal findings and the histopathology of the liver and kidney samples showed no pathological changes.

## Discussion

Since 1970ies with confusion on the poisonous *Cortinarius* species the methods to analyze chemically mushroom toxins have developed. Oubrahim et al. (1997) describes three methods for identification and quantification of orellanine, the main toxin, from several *Cortinarius* species. According to Oubrahim et al. classical TLC or electrophoresis coupled with electron spin resonance are sensitive enough methods to detect orellanine.

In the rat experiment by Möttönen et al. (1975) the renal weight, serum urea value and microscopically demonstrated renal lesions were used as criteria for toxic effect caused by the mushrooms. Rats fed with *C. rubellus* showed variations by these criteria, even severe lesions in kidneys were found. Among the rats fed with *C. gentilis* material only one rat out of six animals showed some changes in the histological examination of the kidneys but no differences in the serum urea values. The microscopically demonstrated renal lesions of this rat were mild according to the authors. One may suggest that a possible mixing of material of *C. gentilis* with that of *C. speciosissimus* could explain the results gained by their animal experiment.

The toxicity studies in the cell culture test and the feeding test on mice gave no hints that Finnish *C. gentilis* is toxic. Experiments carried out on different animal species are not directly comparable, but on the other hand mice are used in similar toxicity studies of *Cortinarius* species (Prast et al. 1988).

It is concluded that an unfortunate mixing of material of two *Cortinarius* species and the difficulties to quote a paper written only in Finnish language may have been misleading to the later reports that *Cortinarius gentilis* is toxic. Until today no evidence is found that Finnish *C. gentilis* is toxic and thereby differs from the same species from other countries.

Recent taxonomic studies (Høyland & Holst-Jensen 2000) have shown that *C. gentilis* and *C. rubellus* belong to different phylogenetic groups. *C. gentilis* is a member of the subgenus *Telamonia* and related to *C. brunneus*. *C. rubellus* belongs to the subgenus *Cortinarius* but has no close relatives.

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