Molluscocide action of the cupric ricinoleate

by

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It is estimated that there are 150 million people in the world suffering from schistosomosis and the means at our command for fighting this disease are still very inadequate. An illness with such an alarming infection rate constitutes itself in a constant menace to public health; prophylactic measures are, therefore, required, especially through action against the molluscs responsible for the dissemination of the disease. This can be done by using low-cost chemical substances with immediate or delayed results, permitting an economic treatment of large snail infected areas.

A number of chemical substances containing copper in their molecule offer a high mollusc mortality coefficient; some of them, as the cupric sulphate and Schweinfurth green, were recommended by the authors. These copper compounds up to now used in the prophylaxy are either highly soluble in water, as the cupric sulphate, or they remain practically insoluble in water, as is the case of Schweinfurth green. When in contact with molluscs, highly soluble compounds develop a prevailing instantaneous mortality action; they must, therefore, be used in heavy concentrations, to counterbalance the more or less accelerated dilution to which they are subjected, depending upon the water movements. When using cupric sulphate, small portions of same are slowly transformed into less soluble compounds, as cupric carbonate or hydroxide, by the action exerted by the mud to be found at the bottom of water collections. Insoluble compounds are also poisonous to the touch; they form a deposit on the mud and kill molluscs that approach them. Insoluble compounds come in form of a very fine powder and large quantities of the material are necessary to treat fair-sized swamp areas.

On examining several organic copper compounds, especially the salts of fatty acids, we have come upon the cupric ricinoleate, whose properties result in making it very adequate as a molluscocide. The cupric ricinoleate, cheap and easily prepared, containing in its molecule an oxidril that favours the formation of stable emulsions with the water especially in great dilutions, develops a wide molluscocide action
in dilutions above the 1:10 million proportion, a factor of the utmost importance regarding its usage on a large scale in the prophylaxy of schistosomosis.

**Teneur of cupric salts, point of reference CuO**

It is quite clear that the molluscocide action of the cupric ricinoleate is due to the metal, while the fatty acid acts as an emulsification support. So as to be able to compare the activity of this and other organic copper compounds with relation to that of the cupric sulphate, we decided on the practice of using the copper oxide (CuO) as a reference point for the solution and compound teneurs. For this purpose, the process consists in measuring into a previously tared china crucible 0.5-1 ml of the solution, cause the solvent to be evaporated by means of a bain-Marie and whiteheat the crucible over a Bunsen burner, weighing the CuO residue after having allowed to cool.

**Preparation method for the cupric ricinoleate**

I — Preparation of the sodium ricinoleate:

**Formula:**

\[
\text{Ricinus oil F. B.} \quad \ldots \ldots \ldots \ldots \quad 1.000 \, \text{gr}
\]

\[
\text{Potassium hydroxide} \quad \ldots \ldots \ldots \ldots \quad 200 \, \text{gr}
\]

(or 150 gr sodium hydroxide)

\[
\text{Filtered water} \quad \ldots \ldots \ldots \ldots \quad 2.000 \, \text{ml}
\]

Dissolve the sodium hydroxide in water and, while the liquid is still warm, allow the ricinus oil to be stirred in slowly. Place in bain-Marie until it is possible to entirely dissolve in water a small sample taken from the mixture. Allow a 24 hours' rest.

II — Dissolve the soap obtained by the above process in 5 liters filtered water and slowly stir in a solution containing:

\[
\text{Cupric sulphate (CuSO}_4 + 5 \, \text{H}_2\text{O}) \quad \ldots \ldots \quad 400 \, \text{gr}
\]

\[
\text{Filtered water} \quad \ldots \ldots \ldots \ldots \quad 2.000 \, \text{ml}
\]

Let the copper soap not soluble in the water deposit. Pour out the water and submit the viscous greenish mass formed by the ricinoleate, palmitate and stearates copper soaps, to successive washings in 42°Bè heated alcohol, in small quantities, until the alcohol comes out colorless. The copper ricinoleate only may be dissolved in alcohol, this way separating from the other saturated fatty acid soaps. Copper ricinoleate can be preserved until the time when it is to be used either as a viscous mass or in a 24° Baumé solution.
Preparation of the emulsion

The emulsion may be obtained by slowly adding, under constant agitation, a cupric ricinoleate alcoholic solution with a 1½ cupric oxid (CuO) equivalent, dosed in accordance with the above-described method, to a water volume required for a final emulsion at a 1:5.000 dilution. This 1:5.000 emulsion seems to be the most convenient for pressure sprinklers, while the long arm of the apparatus should, whenever possible, be sunk into the water that is being treated. The more it is diluted, the greater becomes the stability offered by the emulsion.

EXPERIMENTAL PART

I—Laboratory tests

To obtain comparative data between mollusccocide action as exerted by the cupric ricinoleate against that of other commonly used agents, as the cupric sulphate and the sodium pentachlorphenolate, a series of experiments were effected in the laboratory, using 16 x 16 gawns, each of which had its bottom covered by a certain equal amount of washed sand and solution or emulsion liquid, each gawn containing 500 ml liquid, over which there floated trepoeraba branches. The dilutions used for each tested substance were the following: 1:400 thousand, 1:500 thousand, 1:1 million, 1:1.5 million, 1:2 million, 1:10 million, 1:20 million, 1:50 million; tests were always made with other gawns used as witnesses, which contained only common tap-water used in the preparation of the dilutions. The copper salt dilution teneurs, as mentioned above, refer to CuO. The 20 snails which were placed in each gawn were not laboratory-bred, but had been caught in a marsh existing in the vicinities of the laboratory, 1-2 days before the tests, and were of the A. glabratus species. It was established, initially, that two different actions may be distinguished among the mollusccocides, one immediate, the other delayed.

Attention must be called to the fact that the results obtained in the tests carried out under he above-described conditions showed much variation, a certain uniformity of results, however, being noticed for tests repeated at short intervals. These variations can be explained by several reasons, as the more or less pronounced resistance of the snails, due to biologic phases, and oscillation of temperature in the different periods of the year. These variations were especially noticeable in tests with cupric ricinoleate, and the explanation for this is that this is a very fine emulsion, which tends towards the formation of a delicate film adhering to the vegetation; in these the mortality action increases for, when the snail returns from its hibernal cycle, it requires larger food consumption.

From these tests one can conclude that both the cupric ricinoleate and the cupric sulphate have a pronounced precocious mollusccocide action, constant and more or less equivalent, and which is apparent in 1:500 thousand dilutions already, at the very first contact, occasioning retirement of all molluscs in the gawn; 50% of the snails were found
to be sensitive to a 1:1.5 million dilution. Under these circumstances, the precocious action of the pentachlorophenol or that of its sodic salt is unexisting. The delayed lethal action of the cupric ricinoleate is superior to that of the cupric sulphate, pentachlorophenol or its sodic salt.

After an action of 24 hours, in the best tests, made in the middle of a hot month, March, copper ricinoleate was still found to be effective, killing 65% of the molluscs in a 1:20 million dilution, against 45% of the cupric sulphate in 1:10 million dilutions, and 5% of the pentachlorophenol. These percentages have a relative value, for they show a large variation range, depending upon the time of the year in which the tests are carried out; as an example for this we may mention the relation obtained in tests effected in June, a period during which the temperature is at its lowest in Rio de Janeiro, the following results being obtained after a 24 hours’ contact with a dilution of 1:1 million: the copper ricinoleate caused the death of 75% of the snails in the gawn, cupric sulphate 60%, and sodium pentachlorophenolate 50%.

II — Field tests with the copper ricinoleate

For these tests we selected a small pool situated near Marechal Hermes, a suburb of the Federal District. This focus was formed by a clay-pit at a distance of some 50 meters from the high-way, an artificial excavation, which measured, in meters, around 30 x 70, with 1.5 maximum depth, resulting possibly from the landscaping and earth works carried out on the public high-way. Vegetation, which was more abundant at the margins, formed little tufts towards the centre, comprising trepoeraba, common grass, and other not identified vertical gramineae, wide-spaced and scarce. After 2 o’clock p.m. a light breeze stirs up the water surface, impelling the floating snails to one of the recesses of the excavation, towards the direction of the road. This place is cleaner and deeper, and children are in the habit of bathing there. Near the focus earth-works are being made; refuse of all kinds is found there, as well as some pigs. A little farther down lives a recently acclimatized population, original from the North-Eastern States of the country, employed in the local industries.

Our experiments were begun on July 5th 1954; after proceeding to a quick verification of the planorbidei (A. tenagophilus) density along the marginal contour, the first copper ricinoleate application was carried out at 3 o'clock p.m., in a 3% alcohol solution (calculated in CuO). Initially, spraying of the liquid was done by means of an insecticide sprayer but, as this apparatus was found be unsuited for the purpose at hand, application of the molluscoide was made directly into the water, with subsequent stirring. An hour later, a reading of this result was made as follows:

Dead planorbidi: 190
Living " : 78

Mortality rate: 70.8% (Map on fig. 1).
On July 12th 1954, a second ministration of copper ricinoleate was effected, with a better suited equipment, i.e. an adequate sprinkler working at 40 lb pressure, used by the National Malaria Service in DDT sprinklings. For the solution we used a 1.25% (CuO dosed) copper ricinoleate alcoholic solution, using 6 liters of this solution on this occasion.

With this solution we prepared an approximately 1:5,000 emulsion, slowly pouring the alcoholic solution into a can filled with water, stirring energetically all the time. This emulsion was then transferred into the sprinkler and sprayed on the pool (Map on fig. 2), at first with the apparatus above water level, then sunk into the waters, thus forming a stable emulsion. This way ricinoleate was applied all along the margins, counterclockwise, with the exception of the small recess marked on the map, formed by a small narrow gulf, where an abundance of snails was found, acting as witness to the test. Some small grass tufts off the margins, in a muddy and deeper area, could also not be reached by the sprinkler. The operation was over at 3.30 p.m., and the afternoon breeze began to blow. One hour later a reading was effected, with the following results:

1) Treated area —

| Dead planorbidii: | 400 |
| Alive " | 20 |

Mortality rate 95.2%

2) Witness area —

All planorbidii were found alive.

Twenty-four hours later a second reading was effected, as follows:

| Dead planorbidii: | 72 |
| Alive " | 200 |

As before, all planorbidii in the witness area continued alive.

It must be noticed that the majority of dead molluscs, not only these found by the first counting, but also those which died in the following hours, naturally have sunk into the mud and into deeper waters, so that only those at the margins could be subject to counting.

The molluscs which were still found alive after 24 hours did evidently migrate from the more distant central areas, where it was not possible to effect the spraying with copper ricinoleate emulsion.

The position of snails in the shallow marginal section separated as a witness area, and which could not be reached by water currents, as well as the position of those others carried by the afternoon breeze, are all clearly shown in the annexed map, giving a distinct idea of the results obtained.
CONCLUSIONS

1 — From preliminary tests effected in the laboratory and in an A. tenagophillus infected pool, it was verified that copper ricinoleate, when used in large dilutions and forming stable emulsions with the water, possesses a pronounced molluscocide action.

2 — The molluscocide action as exerted by this copper salt is much stronger than that of the cupric sulphate; like this one, it also develops an immediate and a delayed action; it is also much more powerful than the pentachlorophenol and its sodic salt, both of which have only a delayed action.

3 — The usage of this copper ricinoleate as a highly-active molluscocide is recommended, due to its easy preparation, low cost, insolvability in water, with a formation of emulsions of very small particles and tendency to form thin layers on vegetation, active not only in the emulsion but also after depositing.