PRELIMINARY EXPERIMENTS FOR THE CONTROL OF CERTAIN EUROPEAN VINE-MOTHS BY FUMIGATING WITH CYANOGAS CALCIUM CYANIDE.[1]

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In western Europe there is no fruit cultivated as extensively as the grape. Travelers there are at once impressed by its general culture. Vineyards are exceedingly common in Portugal, Spain, Italy, France, Switzerland and western Germany, the parts of Europe with which the writer is familiar. It is natural, of course, to assume that a plant as common as the grapevine must have a large number of insect enemies. And it has. There are more than a dozen insects which may be considered of economic importance. The most notorious of them all, however, are two small Tortricids, commonly known in Europe as Cochylis (*Clysia ambiguella* Hb.) and Eudemis (*Polychrosis botrana* Schiff.).

Records show that for more than two centuries these two pests—and they are pests in the full significance of the word—have been present in the vineyards of Europe. Over a period of many years the annual loss to the vineyardists in each of the countries mentioned above has been millions of dollars. Owing to this enormous damage they have been the subjects of study by various noted entomologists in these countries, but till now no effective method of control has been discovered.

The investigations of the writer in the winter of 1925-26 brought forth the interesting information that thse two insects are unable to survive a dry season. While he was in Spain during the spring and summer of 1924 he was witness to a severe drought lasting from mid-April to late that autumn. This dry spell had reduced their numbers more than 95% for the season of 1925. The month of January, 1926, was spent in an exhaustive

[1] In November, 1925, the American Cyanamid Sales Co. of New York, sent the writer, who was then in their employ, to Europe to investigate the possibilities of Cyanogas calcium cyanide being used for the control of the vine pests mentioned in this article.

survey of the country for an infestation where methods of control with calcium cyanide could be studied, but without success, for the infestations throughout the country had been reduced to a minimum.

European entomologists have uncovered a formidable array of natural enemies during their constant studies of these two pests. There are recorded some forty hymenopterous parasites of the eggs, larvæ, and pupæ common to both pests. They are also frequent victims of several fungi and bacterial diseases. The annual loss occasioned by the two pests, despite these natural enemies, has always been enormous, for which reason many efforts at artificial control have been attempted.

One of the most promising artificial control measures was that attempted on a small scale by Dr. Friedrich Stellwaag of Neustadt a/d Haardt, Germany, in 1917-1918.[2] He conceived the idea of fumigating the vines with HCN gas by the use of sodium cyanide and sulphuric acid under cover, because of the success this method had produced in the California citrus groves. His preliminary experiments, well planned and thoroughly executed, were carried on between April 24 and June 12, 1917. In April the fumigations were made on the leafless vine stalks (winter condition) and in June they were fumigated in their summer condition. The results obtained in Dr. Stellwaag's experiments may be briefly summarized as follows: 1. Fumigation of the plants in their leafy (summer) condition, even when only small quantities of the gas are used, cause damage to the foliage without completely killing the pests. 2. On the other hand the vines in their leafless (winter) condition are able to withstand larger quantities of the gas, sufficient (under the conditions of the experiment) to kill the cocoons found under the tent in which the fumigation was done.

The results of his 1918 tests[3] checked substantially with those of the previous year, except that more injury to the vines was noted due to rapid formation of the gas. Furthermore, Dr. Stellwaag believed that this form of fumigation was "too fussy,

[2]"Der Weinbau der Rheinpfalz," No. 8, August 1917.
[3]"Der Weinbau der Rheinpfalz," No. 1, January, 1919.

too expensive and unreliable." Therefore he discontinued further experimenting along these lines.

With the development of calcium cyanide during recent years, it was believed that these objections were eliminated. It is a product convenient to handle, easy to apply, less dangerous, and much more economical. Its greatest asset, how-ever, is that the gas is not evolved suddenly and turbulently. When normal dosages are applied the chance of injury to living plant tissues is at a minimum, for the gas evolves gradually, the peak being reached some little time after the plant tissues have received small amounts of the gas produced soon after application. In the firm belief that calcium cyanide would succeed where the old process of sodium cyanide plus sulphuric acid had failed, the writer was sent to Europe in December, 1925, to investigate the possibility of control of Eudemis and Cochylis with Cyanogas calcium cyanide.

As mentioned above, no adequate location could be found in Spain, due to the scarcity of the pests in that country. However, two suitable sites were found, the first being near Bordeaux, France, the other in Lausanne, Switzerland. Dr. J. Feytaud, director of the Station Entomologique de Bordeaux, very kindly co-operated in the undertaking by allowing the use of his laboratory facilities and also permitting us to experiment in vineyards under his charge. Similarly, Dr. H. Faes, director of the Station Fédérale d'Essais Viticoles de Lausanne, courteously extended the use of laboratory and vineyards. Both thus facilitated the experimental work.

Life Histories of the Pests.

Before proceeding with the details of the experiments it is well to mention briefly the salient features in the life histories of these insects. Essentially they are alike, the only variation being that *Clysia ambiguella* (Cochylis) has two annual generations, whereas *Polychrosis botrana* (Eudemis) has sometimes a partial and sometimes a full third generation. Both insects pass the winter as pupæ within rather thick cocoons spun by the larvæ. The cocoons are ordinarily attached to the vine stalks, although at times they have been found on rocks, fences, the wooden supports placed beside the stalks, and other similar objects. In seeking locations for hibernation the fullgrown larvæ instinctively select such sheltered and secluded spots, in which to spin their cocoons, as will insure complete protection against the elements during the long hibernating period. Consequently the cocoons are to be found in the folds of the bark, in crevices, holes, fissures, cracks, etc. The newly formed cocoons of Eudemis are clean and snow-white; whereas those of Cochylis are invariably covered with dirt, dust and debris of all sorts, making them difficult of detection.

The adult moths emerge late in April or early May, simultaneous with the bursting of the buds, and are present thereafter until early in the fall. The Cochylis adults are nocturnal in habit, while those of Eudemis are more active at dawn and at dusk of day. The females, after fertilization, commence laying their tiny and solitary eggs in the flowers of the host plant. These eggs hatch in from 7 to 30 days. The larvæ of the first generation attack and destroy the flowers, while those of the following generations damage the fruits. The second generation adults of Eudemis appear about the third week of June (a cycle of 7 weeks) while those of Cochylis do so towards the middle of July (a cycle of 9 weeks). Thus it will be seen that from early May until cool weather stops their activities (late September) all four stages of these insects may be found in the field.

The Experiments

The aggregate damage caused annually by these insects is enormous. The idea was also gathered from entomologists interviewed that no difficulty would be experienced in locating heavy infestations. In view of these facts it was the original intention to experiment with Cyanogas calcium cyanide for the control of the pests by treating the infested vines without disturbing or removing the insects from their natural hibernating quarters. However, when the actual work was begun the plans were changed to suit the work to existing conditions. Bushels of bark were peeled off the vines in a number of infested vineyards within close proximity of the two laboratories, and all cocoons of the two pests were then removed from them in the laboratories. In detaching them from the bark many of the cocoons tore open, for they were attached very firmly. About 700 cocoons of the two species were recovered from about twelve bushels of peeled bark.

For their treatment with calcium cyanide, the insects (cocoons) were retained in a coarse muslin suspended on the stalks fumigated. No pupæ were used in the experiment which failed to respond to a gentle pinch with the forceps, prior to fumigation.

The vines.—The vines treated in Switzerland were of a species called "chasselas," indigenous to that country, while in France vines with American rootstock were selected. Vines of varying ages, from 5 years to 50 years, were treated in order to secure information concerning the resistance of plants of varying ages to the action of HCN gas.

In Switzerland the majority of vineyards are of the type where the vines are grown singly, trained on wooden supports about 1 meter or a little over in height, placed in the ground close to the pruned vine just as soon as growth begins in the spring. The French methods of cultivation are of three types: (1), individual vines (as in Switzerland) pruned close to the ground; (2), vines trained on a single line of wire stretched the full length of each row; and (3), taller vines trained on double lines of wire, the first line about 30-35 c. m. above the ground, and the second wire about an equal distance above the first. The wires are attached to strong wooden posts in the rows, placed about 12-15 meters apart. The majority of vineyards in France are of the third type.

The covering.—The protection afforded the insects by their heavy covering (the cocoons) and also the fact that their respiratory system is at the minimum period of functioning during this season of the year (late winter and early spring) and stage of their life, made it at once evident that open air fumigation would be futile. Consequently a canvas tent and a cylindrical galvanized iron can were used in covering the vines for the treatments. The tent, of heavy impermeable canvas, was made large enough to permit treatment of four vines together. Its dimensions were 1.25 m. x 1.25 m. x 0.65 m. On its four sides a flap 12 c.m. wide was planned to insure against leakage of gas during fumigations, by placing earth or stones or other weighty materials on the flaps. The galvanized iron can was 95 c.m. high with a diameter of 60 c.m. The galvanized iron can was originally thought of because of the certainty of its being airtight and rain proof, very important considerations for overnight fumigations. It has the further advantage in winter fumigation work over the canvas tent in that the temperature within it rises several degrees above that of the outside air. Tests made to determine this point showed an increase of 2 degrees C. after an exposure of one hour, when the outside temperature was 10 degrees C. and an increase of 5 degrees C. after an exposure of two hours with the outside temperature at 16 degrees C.

Dosages and exposures.—The cubic content of the tent was 940 litres. In order to secure a concentration of HCN gas equal to 1% of the volume of the space within this tent, it was calculated that approximately 45 grams of $Ca(CN)_2$ should be applied. For a dosage of 10% volume, therefore, it would be necessary to use ten times 45 grams, or 450 grams. Likewise it was found that 12.5 grams of $Ca(CN)_2$ would produce a 1% volume concentration of HCN gas when applied under the galvanized iron can, the cubic content of which was 270 liters. The dosages used in the 71 experiments varied from 0.275% volume to 10.00% volume, each dosage being tried at least twice with a given exposure. The exposures were varied also, these being dependent on the dosages. The exposures were for periods of 30 minutes, 45 minutes, 1, 1½, 2, 2½ and 3 hours. Some overnight exposures were also tried. In general, smaller dosages were used in the long exposures and larger dosages in exposures of short periods.

The treatments.—Dr. Stellwaag, in the experiments referred to above, found that the grapevine is an extremely delicate and tender plant and that it is unable to withstand heavy concentrations of HCN gas. In view of this, coupled with the desire to cause as little injury as possible to the plants, the dosages used in the first twenty experiments were less than 1% volume. Beginning with a dosage of 0.275% volume, with an exposure of 30 minutes, both dosage and exposure were increased to 1% volume and treated one hour, but witn no appreciable effect on the insects or on the plants. Gradually the dosages were increased even beyond that fatal to the insects up to 10% volume to note the effect of abnormally strong applications of HCN gas on the vines. It is interesting to note that even such a heavy dosage as 10% volume (450 grams to a space of 940 liters) just before the flow of sap had commenced (on March 25, 1926) did not prove fatal to the plants, merely "retarded growth" being the observation made on the following 12th of April.

In all the experiments fresh Cyanogas calcium cyanide grade "A" dust was used. The exact dosage of the dust, after being measured on balance scales, was placed in a foot-pump duster and applied under the tent or galvanized iron can. No difference was noted in the results when the nozzle of the duster was directed to the bottom of the covering or to its top. In the latter case the plants would be covered with a fine coating of the dust, whereas when the dust was directed to the ground there would be practically no dust on the vines. In either case the effect both on the insects and the plants treated was the same.

To discuss each of the 71 experiments in detail seems unnecessary, for the majority resulted in only partial kills or no kills at all. Only in nine of the experiments was complete kill of the insects exposed to treatment secured, the details of which are given in the accompanying table.

The primary object of Experiments 29, 30, 31 and 32 was to see if abnormally excessive dosages, during exposures of 30 and 45 minutes, under the tent and also under the galvanized iron can, would be fatal to *the vines*. Observations on the condition of the plants made on April 12, a short while after the leaf buds had burst, showed them to be slightly retarded in growth. No injury was caused to any of the plants in their dormant condition even with such strong applications of the poison.

Experiment 53 was conducted on a fair, warm and calm afternoon. 15 naked pupæ were exposed to 3.5% volume (45 grams of poison to 270 liters of space) for one hour, from 2:15 to 3:15 p. m., under the galvanized iron can. The nozzle of the

Exp. Number	29	30	31	32	33	53	55	57	59
Date	3–25	3–25	3–25	3–25	3–26	4–5	4–5	4–6	4–6
Hour[4]	3:03 P.M.	3:15 P.M.	3:59 P.M.	4:08 P.M.	7:32 A.M.	2:15 P.M.	3:23 P.M.	2:30 P.M.	4:33 P.M.
Sun	Yes	Yes	No	No	Hazy	Yes	Yes	Yes	Yes
Clouds	No	No	partial	partial	light	No	No	No	No
Wind	Weak	Weak	slight	slight	calm	No	gentle	calm	calm
Temperature[5]	54° F.	57° F.	55° F.	55° F.	44° F.	61° F.	63° F.	61° F.	64° F.
Relative Humidity[5]	59%	55%	54%	54%	78%	54%	54%	51%	47%
Exposure	45 min.	45 min.	30 min.	30 min.	45 min.	1 hr.	3 hrs.	2 hrs.	$1\frac{1}{2}$ hrs.
Dosage	450 gr.	125 gr.	450 gr.	125 gr.	225 gr.	45 gr.	25 gr.	32 gr.	38 gr.
Rate	10% vol.	10% vol.	10% vol.	10% vol.	5% vol.	3.5% vol.	2% vol.	2.5% vol.	3% vol.
No pupae treated	10	10	10	10	9	15	11	5	6
% pupae killed	100%	100%	100%	100%	100%	100%	100%	100%	100%
Effect on vines	retarded growth	growth retarded	none	much retarded	none	a little retarded	none	none	none
Covering	Tent	G. I. Can.	Tent	G. I. Can.	Tent	G. I. Can.	G. I. Can.	G. I. Can.	G. I. Can.

[4]Beginning of treatment.
[5]At start of fumigation.

foot-pump duster was directed upward, and at the termination of the treatment it was noted that the powder was very uniformly distributed over the ground and lightly over the plant. All of the pupæ were dead when examined 42 hours after fumigation.

In Experiment 55, eight naked pupæ and 3 pupæ in cocoons were treated for 3 hours (3:23 to 6:23 p. m.) on April 5. Only 25 grams of calcium cyanide was used (2% volume). The temperature at the beginning of the experiment was 63° F. and 59° F. at the end; and the relative humidity went up from 54% to 57%. There was a gentle breeze present, but really without effect since the treatment was under the galvanized iron can. There was again a uniform distribution of the dust on the vine and the ground. When examined on the 7th of April all the naked pupæ were dead and those in the cocoons were stupefied. They never recovered, for on April 10 the three pupæ in cocoons are recorded as being also dead.

Another combination of dosage and exposure, under similar atmospheric conditions and fatal to the insects treated, was found by the test under Experiment 57. Here five cocoons were fumigated under the galvanized iron can for two hours with a dosage of 2.5% volume (32 grams to the 270 liters of space). The temperature rose from 61° F. at the beginning to 64°, and the relative humidity dropped from 51% to 47%. When examined 48 hours after exposure all five pupæ in cocoons appeared stupefied, while three days later they were pronounced dead.

Perhaps what may be considered the best combination of dosage and exposure was that revealed by Experiment 59. On April 6 one naked pupa and five in cocoons were treated under the galvanized iron can from 4:33 to 6:03 p. m. 38 grams of calcium cyanide (3% volume) were used in this experiment. The temperature varied from 64° F. to 59° F. and the relative humidity was between 47% and 57%. The day was fair, clear and calm. Forty hours after the treatment the insects were found dead upon examination.

In all the remaining experiments mortality varied from none to 82%. The following chart illustrates the combinations of dosage and exposure resulting in 100% mortality to the insects when accompanied by favorable climatic conditions, as shown by these tests.

Exposures (in hours)

Dosages (in % volume)	$\frac{1}{2}$	$\frac{3}{4}$	1	$1\frac{1}{2}$	2	$2\frac{1}{2}$	3	
1%								
1.5%								
2%							X	
2.5%					X			
3%				X				
3.5%			X					
4%								
4.5%								
5%		X						

(Note: x signifies 100% kill of insects fumigated; e. g., a 3% volume treatment for a period of $1\frac{1}{2}$ hours resulted in a complete kill of the pupae exposed to the treatment.)

The following conclusions are presented only because they throw some light on the possibilities of the use of calcium cyanide for the control of these pests. However, it must not be forgotten that they are the results of only a few preliminary experiments.

Conclusions

1. Treatments of Cochylis and Eudemis with dosages between 2% volume and 3.5% volume during exposures of from one to two and a half hours give better kills than stronger dosages over shorter exposures, or weaker dosages over longer periods of fumigation.
2. Temperatures above 60° F. appear to be more effective.
3. High relative humidity does not appear to be so important a factor as high temperature.

4. A metal covering over the vines gives much better results in the number of insects killed than a canvas covering, under identical conditions. This appears to be due principally to two factors; (1) A metal covering is more air tight; (2) the temperature in a metal covering is higher after a certain exposure than it is under a canvas tent under a like exposure.

5. In the winter dormant condition vines are not injured by the use of Cyanogas calcium cyanide. Both young and old vines seem to withstand with safety such strong treatments as 10% volume for 45 minutes.