T. Pl.ziekten 65 (1959): 116–118

THE EVALUATION OF ANTI-COAGULANTS AS RODENTICIDES IN THE LABORATORY[1]

Met een samenvatting:

Het laboratoriumonderzoek van anti-coagulanten als rodenticiden

BY

F. E. LOOSJES

Plant Protection Service, Wageningen

Everyone who wishes to market a pesticide in The Netherlands must have a licence, issued by the Director of Agriculture of the Ministry of Agriculture. To get such a licence a sample of the pesticide involved, its composition, directions for use, etc. have to be delivered at the Plant Protection Service. This regulation came into force in October 1948 and since that time all insecticides, acaricides, rodenticides etc. are screened for licensing in the laboratory and in practice. In this article we shall deal with rodenticides in particular.

Among rodenticides, arsenic compounds, strychnine, thallous sulphate, zincphosphide and red squill are known for a considerable time. When these substances are given, sufficient quantities should be eaten by the animals in the first day of intake in order to kill them. If insufficient dosages are eaten during the first time, the animals may only become ill and can get an abhorrence of the toxic substance concerned. It is possible that other specimens of the population also become suspicious and refuse to take up the poisoned food.

Within the last ten years anti-coagulants which make the bloodvessels pervious and prevent clotting of the blood have been added to the known series of rodenticides. The advantages of these substances are that the animals do not get any bait shyness so that they can be fed several days in succession until they are killed by internal bleeding. As a result the mortality of a population outnumbers that caused by other poisons. Furthermore one initial dosage is generally not sufficient to kill an animal; more uptakes are necessary.

This fact makes these anti-coagulants rather safe in use; accidental uptakes are usually without deadly effect. Moreover one gets the impression that killing rats by an anti-coagulant is in most cases much less repulsive than e.g. by scilla. As in addition anti-coagulants can be used economically it will be clear that among the rodenticides (offered for investigation) they soon held an important position so that methods for testing them in the laboratory had to be worked out. But it is also evident that these methods should differ from those used in the case of the classical rodenticides.

A starting-point for a testing-scheme was found in the work of HAYES & GAINES (1950). Of course our methods have altered since, following various other publications and as a result of our own experience.

We are evaluating anti-coagulant rodenticides now by means of the following methods:

[1] Accepted for publication 24 February 1959.

1. Estimation of the percentage of active substance in the chemical laboratory.
2. Stomach-tube tests to get information on the acute toxicity of new types of anti-coagulants to study the hazards for domestic animals, etc.
3. Estimation of the survival-time after daily intake until death. A series of increasing percentages of active substance in the food is given. Usually from 0.78 up to 800 p.p.m. is administered; the next percentage is always twice the previous one. Each of five or more rats is offered daily 20 g of poisoned food holding the same amount of anti-coagulant. Every day the intake is fixed. The results are plotted in log. dosage-survival-time curves, as far as the killed rats are concerned. An average survival-time of all members of a series of five cannot be given if one or more individuals survive. To overcome this difficulty we use according to POWERS (1918) the reciprocal value: $\frac{100}{S}$, which gives for the survivers $\frac{100}{S} = 0$. This so-called velocity of fatality is plotted with the log. dosage. In this way one gets an easy information on the smallest dose that gives the shortest survival time. This test is only done with new types of anti-coagulants.
4. Tests in which the animals are fed during a limited number of days. Brown rats are usually fed during three days, black rats and mice during five or seven days. They are usually fed with the amount of toxic substance prescribed in practice, incorporated in 20 g laboratory food for rats and in 5 g laboratory food for mice. The daily intake is registered. In this way we get an information on the lethal feeding period of a new brand, whether it is up to the standard or not.
5. Interruption-tests, i.e. tests in which the animals are fed every other day, every third or every fifth day. Again the daily intake is noted, also that of the plain bait of the interim days. Thus information is obtained on the influence of irregular intake of a bait under field conditions. In this connection it should be considered that one product might be excreted more rapidly than another. Also one product may accumulate more than another. The method also may give some information on the acceptance of the bait.
6. Acceptance experiments in which individually caged rats have a choice of food out of four containers. After feeding three to six days on oats, containers 1 and 3, or 2 and 4 (or another combination) are filled with the bait to be tested, while oats or laboratory food are given in the others. The daily intake is fixed regularly. The next day the bait is put in the alternating containers and oats in the other ones. Then all containers are filled again with oats for 3 to 6 days. Always the same amount (10 g) of food or bait is offered per container. Brown rats are given 3 days oats and 1 day a choice between oats and bait alternating. Black rats and house mice are given 6 days oats and 2 days a choice.
   Each product is tested on brown and black rats and on mice.

All brown or black rats are caught in the field and are first accustomed to laboratory cages and food before they are used in trials. The weight is taken at the start and at the end of an experiment. The animals are always caged separately and have fresh water available. The sex is determined. So far we could not find much difference between the susceptibility of both sexes for

anti-coagulants. Neither did we find indications in the literature in this respect.

Pregnant rats are not used in the experiments. Groups of rats of comparable weight are used for each separate treatment. Rats that have survived an experiment are killed. All symptoms are carefully noted and a *post-mortem* examination is made. White rats are used if there is a lack of wild animals or if we need animals of about the same weight for means of comparison.

Of course laboratory results are checked in practice as much as possible, especially when there is a reason for refusing the licence.

SAMENVATTING

Het onderzoek van anti-coagulanten als rodenticiden in het biologisch laboratorium van de Plantenziektenkundige Dienst, als onderdeel van de middelenkeuring ten behoeve van de bestrijdingsmiddelenwetgeving, wordt volgens de hierbij beschreven methoden uitgevoerd. Naast de chemische bepaling van het gehalte aan giftig bestanddeel wordt de giftigheid nagegaan in maagsondeproeven na éénmalige toediening (LD 50). Verder wordt de sterftetijd vastgesteld: bij het geregeld dagelijks opnemen van vergiftigd voedsel, tot de dood intreedt; bij dagelijks opnemen gedurende enkele dagen (3, 5 of 7 dagen-proeven) en bij opnemen van vergiftigd voedsel, afgewisseld met opnemen van niet-vergiftigd voedsel (onderbrekings-proeven). Ten slotte worden keuzeproeven gedaan om een eventuele afkeer van de dieren voor de middelen te kunnen vaststellen.

REFERENCES

ARMOUR, J. C. & S. A. BARNETT, – 1950. The action of dicoumarol on laboratory and wild rats and its effect on feeding behaviour. J. Hygiene 48: 158–170.

BENTLEY, E. W., L. E. HAMMOND & E. J. TAYLOR, – 1955. The comparative toxicity of 0.025 per cent and 0.005 per cent warfarin to Rattus norvegicus. Pl. Path. 4: 120–123.

BENTLEY, E. W. & M. ROWE, – 1956. Pival, an anti-coagulant rodenticide. J. Hygiene 54: 20–27.

HAYES, W. J. & F. B. GAINES, – 1950. Control of norway rats with residual rodenticide warfarin. Publ. Health Rep. 65: 1537–1555.

REIFF, M. & R. WIESMANN, – 1951. Untersuchungen über ein neues Rodentizid mit kumulativer Wirkung auf Basis der Cumarinderivate. Acta tropica 8: 97–130.

STEINIGER, F., – 1953. Ueber die Wirksamkeit des „Fumarins", eines neuen Anti-Koagulans zur Rattenbekämpfung und sein Vertrüglichkeit für Haustiere. Nachr.-Bl. dtsch. Pfl.-Sch.-D. 5: 167–168.

TAMMES, P. M. L. & H. DE VRIES, – 1955. Het beoordelen van anticoagulanten op basis van de sterftetijd. Versl. Plantenziektenkundige Dienst.