

NAS Committee on the effect of Herbicides in Vietnam

PRELIMINARY PROPOSALS FOR STUDIES ON THE
PERSISTENCE OF HERBICIDES IN FOREST AND MANGROVE SOIL

J. D. Fryer

G. E. Blackman

INTRODUCTION

When herbicides such as 2,4,5-T and picloram are sprayed from an aircraft on to forest only a small proportion will reach the soil direct. After emission from the spray nozzle some of the herbicide will be lost before reaching the canopy as a result of drift of small droplets out of the target area or volatilisation. The majority will be retained on the foliage of the forest plants and then either absorbed or left on the leaf surface. Of the fraction entering the leaf, some will be metabolised within the plant and some reach the soil when dead leaves, twigs or branches fall from the tree.

It may be expected, therefore, that considerably less than the nominal 3 gal/ac of the defoliant agents used in Vietnam will have reached the soil either directly or indirectly. The maximum levels will have been reached in areas where previous sprayings, forest clearing or agriculture, had greatly reduced the amount of vegetation cover. These levels may have approached the nominal dose in open places and occasionally exceeded it when overlap of spray swathes occurred.

The proposed main experiments are designed to investigate the rate of disappearance of 2,4,5-T, picloram and 2,4-D - in order of priority - when a dose of 3 gal agent/ac is applied direct to the soil. Under these conditions residues in the soil are likely to be greater than those generally resulting from the defoliation programme due to the elimination of some of the routes of loss already described. However, it should be borne in mind that where it is necessary in the case of upland forest or mangrove experiments to remove undergrowth from or clear-fell a site prior to spraying to allow reasonably uniform application of the herbicides there will be difficulties in the initial dose and the rate of disappearance of residues compared with the treatment of a relatively undisturbed forest. For this reason it is hoped that in Vietnam it will prove possible to arrange, in addition, aircraft applications of 'orange' and 'white' to allow assessment of the persistence of herbicide residues following an initial treatment of at least an undisturbed mangrove area and if possible a forest area also. The latter may, however, well prove impossible.

Experiments are also proposed to examine the effect of 4 doses of the agents on agricultural soil on establishment and subsequent early growth of a range of crops.

PROPOSED INVESTIGATIONS

The persistence of agents 'orange' and 'white' in mangrove and forest soils in the **Phillipines** and Vietnam.

I MANGROVES

It is hoped that the following experiment will be carried out in the Phillipines and, as far as possible, duplicated in Vietnam.

Object: To determine the rate of disappearance from soil of 2,4,5-T, picloram and 2,4-D applied shortly after clear-felling an area of mangroves.

Site: This should be at the higher end of the tidal range to allow adequate time for spraying and soil sampling when the mud is not covered by water.

Treatments: Orange

C

3 gal (US)/ac

= approx. 12.9 lb 2,4-D a.e./ac + 13.2 lb 2,4,5-T a.e./ac

White

C

3 gal (US)/ac

= approx. 1.6 lb picloram a.e./ac + 6.0 lb 2,4-D a.e./ac

Spray volume: Approximately 30 gal/ac according to equipment available and results of test spraying.

Formulations: 'Orange' is a rather viscous oily fluid which must either be diluted with oil or emulsified with water after the addition of a suitable emulsifier, before it can be applied at the desired rates by conventional plot spraying equipment. Diesel or kerosene are suitable diluents but might be objected to on the grounds that at high rates they might conceivably alter the rate of disappearance of the herbicides. If an oil/water emulsion is used, the possibility of the emulsifier having an effect on dissipation must also not be overlooked, e.g. by allowing greater mobility of the herbicide in tidal waters - a problem that could be overcome by having a quick-breaking emulsion. The advice of Dr. C. Minarick is being sought.

Plot size and layout:

To be determined on site but for the basis of planning it is suggested that an area not less than 90 x 30 yd be cleared to allow 3 x 30 x 30 yd plots, two to be treated with i) orange ii) white; the third as control. Stumps should be cut as near ground level as possible to facilitate spraying. Trash should be removed from area and could be stacked around perimeter. It should not be burned near the site as charcoal could confuse the results. To minimise the risk of cross contamination the cleared area should be orientated so that the main flow of tide is parallel to its shorter sides. If due to local topography it is not possible to have the 3 plots all in one block, each can be located in a separate clearing, in which case the sites should be as close together as is feasible.

Plot marking: Some way will have to be found to locate accurately each plot for the duration of the experiment. Permanent markers for each plot corner will be needed. Unless the area is securely guarded at all times, the removal of markers by local people must be anticipated. If the plots are located by triangulation to adjacent uncut, marked mangrove trees, care will have to be taken that these will still be identifiable in the event of their being cut down by local people.

Tide at time of spraying: Spraying should take place on the ebb tide after the water has drained from the plots. Provided the chemicals have been prepared and measured beforehand and the spraying equipment is in good order and has been calibrated, it should be possible to spray both treatments within 2 hours. Dr. Chandler and Professor Ho are requested to advise urgently on days in January when low tide occurs around noon in the selected areas in the Phillipines and the Rung Sat, Vietnam, respectively.

Precautions for spraying: It is suggested that the spray liquids are strained through a fine mesh filter whilst being poured into the sprayer. They should be made up into transparent containers to allow any crystallisation of 2,4,5-T - ester or emulsion - breaking to be detected. Unless previous tests confirm that adequate stability of an oil/water emulsion of 'orange' can be assured for the required time to allow detection of concentrate in the laboratory, mixing up must be done on site and jerry cans of clean water will be required (total 20 gal?). To prevent drift of spray on to adjacent plots (this is particularly important in view of the high doses used), two windshields may be required - one up-wind, one down-wind. Hessian stretched over a light wooden frame (about 2.5 x 5 ft) is much better than plastic film for this purpose. To assist accurate spraying, a stop watch should be available.

Spraying equipment: Note: The spray bar (with pressure gauge) and nozzle set-up should have been previously calibrated with the spray liquids to be used to provide reasonably uniform deposit over the spray swathe. The viscosity, throughout and spray fan angle may be appreciably different for whatever formulation of 'orange' is used and for 'white' - and even for the different concentrations. It is essential that a full calibration programme is completed before reaching the site and if necessary different spray bar and nozzle configurations provided.

Tests at WRO point to use of a Polyclair Knapsack sprayer fitted with a special lance and sprayboom. Promising results have been obtained with Allman fan jets operating at 10 p.s.i applying 30 gal/ac at 2 m.p.h. to a 2 yd swathe. The present plan is to take out this equipment to the Phillipines, use it for the mangrove and forest sites selected by IRRI and then take it to Saigon. If IRRI does not have suitable equipment, WRO can make available two sets - one for Vietnam, one for Phillipines.

Sampling for residues: Schedule. Suggested times of soil-sampling are: 0, 3, 9, and 27 weeks after treatment. It is necessary to sample immediately after spraying (shallow cores only - say 2 in.

deep) and before the plots become flooded by the tide to estimate the percentage recovery of the applied herbicide. This may vary from around 60 to 100% + according to previous experience in agricultural soils. If possible not less than 20 cores from each sprayed area should be taken at random from each treated area at each sampling date. Without preliminary work to measure variability of residues, there can be no statistical basis for choice of number of cores per plot and logistics are likely to prove the deciding factor. At the 2nd, 3rd and 4th sampling dates, cores should be taken to the full depth of the corer (c. 75 cm), each core being divided into the top 5 in. (12.5 cm) and two 26 cm sections. On each sampling occasion, 5 comparable random cores should be taken from the untreated area. The core sections should be placed in separate polyethylene bags, labelled and stored in a 'deep freeze' within a few hours of sampling until required for analysis. If 20 samples are taken from each treated area and 5 from the control, the number of samples for analysis will be $20 \times 1 \times 2 + 5 = 45$ from the first sampling date and $20 \times 3 \times 2 + 5 \times 3 = 135$ for each of the three remaining dates, i.e. a total of 450 samples. WRO experience suggests a maximum of 16 determinations of 2,4,5-T a day or 8 - 10 picloram per GLC unit. If it is necessary to reduce the number of samples by bulk-ing and sub-sampling, it must be realised that it is unlikely to be easy with this type of soil and that the mixing process will result in aeration of the samples and possibly a rapid change in pH and the microbiological status of the soil, which might affect the persistence of the herbicides. *

Equipment:

The WRO dual purpose 2 x 36 in. soil corer is already available in Saigon suitable for both forest and mangrove soils. A new light-weight corer is currently being constructed at WRO which should be suitable for the mangrove and volcanic soils of the Philippines. It will probably not be strong enough for really hard or stoney soils. The plan is to loan this to IRRI if suitable equipment is not already available there.

Residue analysis:

Samples from the experiments involving agent 'orange' should be analysed for 2,4,5-T and those from the experiment with 'white' for picloram. If possible 2,4-D should be analysed in samples from both experiments but this should be regarded as of lower priority since 2,4-D is known to degrade more rapidly in soil than 2,4,5-T. At the time of writing (22nd November) it is not known for certain which laboratories will carry out the analyses. Dr. Chandler has offered to explore the possibility of the work being undertaken by IRRI and Professor Ho and Dr. Lang will investigate facilities in Saigon. In either case, communication should be established with Dr. R. J. Hance of WRO who has experience of the techniques and problems involved and who, if necessary, is prepared to make a short advisory visit to the laboratories concerned.

Supplementary studies:

In addition to the basic experiments proposed above, it would be valuable if the work could be extended to include (i) plots of mangrove forest in a natural state sprayed by aircraft with 'orange' and 'white' at the standard rate of

* It follows from this that 50 residue determinations should be made immediately after mixing.

3 gal/ac (Vietnam only); (ii) some bioassays of residual phytotoxicity, in particular the planting of mangrove seedlings in situ; (iii) measurement of rate of disappearance when 'orange' and 'white' are applied at lower doses representing drift levels or a dose likely to reach the soil direct under a forest canopy (only if the aircraft applications described below prove not to be feasible or if assessment of the hand-sprayed plots 1 and 6 weeks after treatment indicate a slow rate of dissipation).

(i) Aircraft-sprayed plots. The size and replication of the plots should be decided by the ecology group but it seems likely that two unreplicated strips each of 'orange' and 'white' (at 3 gal/ac) will be all that is feasible with a buffer untreated strip between. As already noted, the route of entry of the herbicide into the soil is likely to be largely via the leaves and twigs of the treated forest and if possible an estimate of the herbicide dose reaching the soil directly or indirectly should be obtained by (i) use of an oil-soluble dye such as Waxoline Red in the spray solution, placing strips of filter paper in suitable holders a few inches above the soil surface prior to spraying, eluting the dye from the filter papers after spraying and assessing dosage by colorimetry; (ii) collecting samples of leaves falling from the trees at intervals after spraying and estimating the herbicide reaching the soil by this route. In either case variation in deposit from place to place within a sprayed plot is likely to be extremely large and unless a considerable number of random samples can be taken and analysed, the chance of any reliable data resulting will be small. Little guidance can, unfortunately, be given on the minimum number of samples required to produce a reasonably low coefficient of variation and it seems that at the best only a rough estimate of herbicide levels reaching the soil can be obtained. For the initial sampling of spray deposit, a reasonable procedure might consist of five rows of 10 samples at right angles to the direction of spraying, spaced at regular intervals along the swathe. Two-inch wide strips of filter paper have proved satisfactory for this purpose, being kept flat in light metal holders which can easily be made up from thin gauge sheet aluminium. If each plot consists of two spray swathes, soil samples should not be taken from the vicinity of the junction of the adjacent swathes, since the dose is likely either to be excessive or much lower than average according to exact position of the aircraft on each run and prevailing met conditions. In Vietnam, the procedure adopted for soil sampling for residue analysis of the aircraft-sprayed plots will doubtless be more a question of feasibility rather than of statistical requirement. If possible cores should be analysed separately as above. If not, bulking and sub-sampling can be done providing thorough mixing can be achieved (probably necessary to do this by hand) and there is only a short interval between mixing and analysis - or deep-freeze storage. Again 20 cores per plot should, if possible, be regarded as a minimum with a selection of sites representative of different densities of canopy. Since the total dose reaching the soil is likely to be considerably less than in the cleared plots and the period of dosage will cover days, if not weeks, following

5 rows of
10 samples

Two spray
swathes
per treatment.
at 100 ft.
each 400 ft
long

Initial
20 vertical
cores per
plot

Dates of
Sampling

treatment according to the rate of leaf fall, more frequent samplings will be required, particularly in the early stages, e.g. 0, 1, 2, 4 and 8 weeks after spraying. If it is decided to collect leaf samples for herbicide analysis, wicker or woven grass baskets of local manufacture should be considered. They would be cheap, light, inconspicuous and easily secured above high tide level to 3 canes.

(ii) Bioassays. In addition to the chemical residue determinations outlined above, it would be valuable if simple bioassays could be undertaken to provide a qualitative assessment of the disappearance of phytotoxic residues. Considerable experience of field and laboratory techniques for bioassay herbicide residues in soil is available at WRO but modification would be required for mangrove soils. For the latter the most realistic approach would seem to be sequential plantings of mangrove seedlings direct into the cleared hand-sprayed plots and the aircraft-sprayed strips.

(iii) Lower doses on cleared plots. A dose of about $\frac{1}{2}$ gal/ac of either agent ('orange' or 'white') might be appropriate, but should be adjusted according to the disappearance rate indicated by the early assessments in the hand-sprayed plots. This should not receive high priority, unless the latter indicate that little loss of herbicide has occurred or if in the case of mangrove soils it should not prove possible to undertake the aerial spraying trial.

II FOREST SOILS

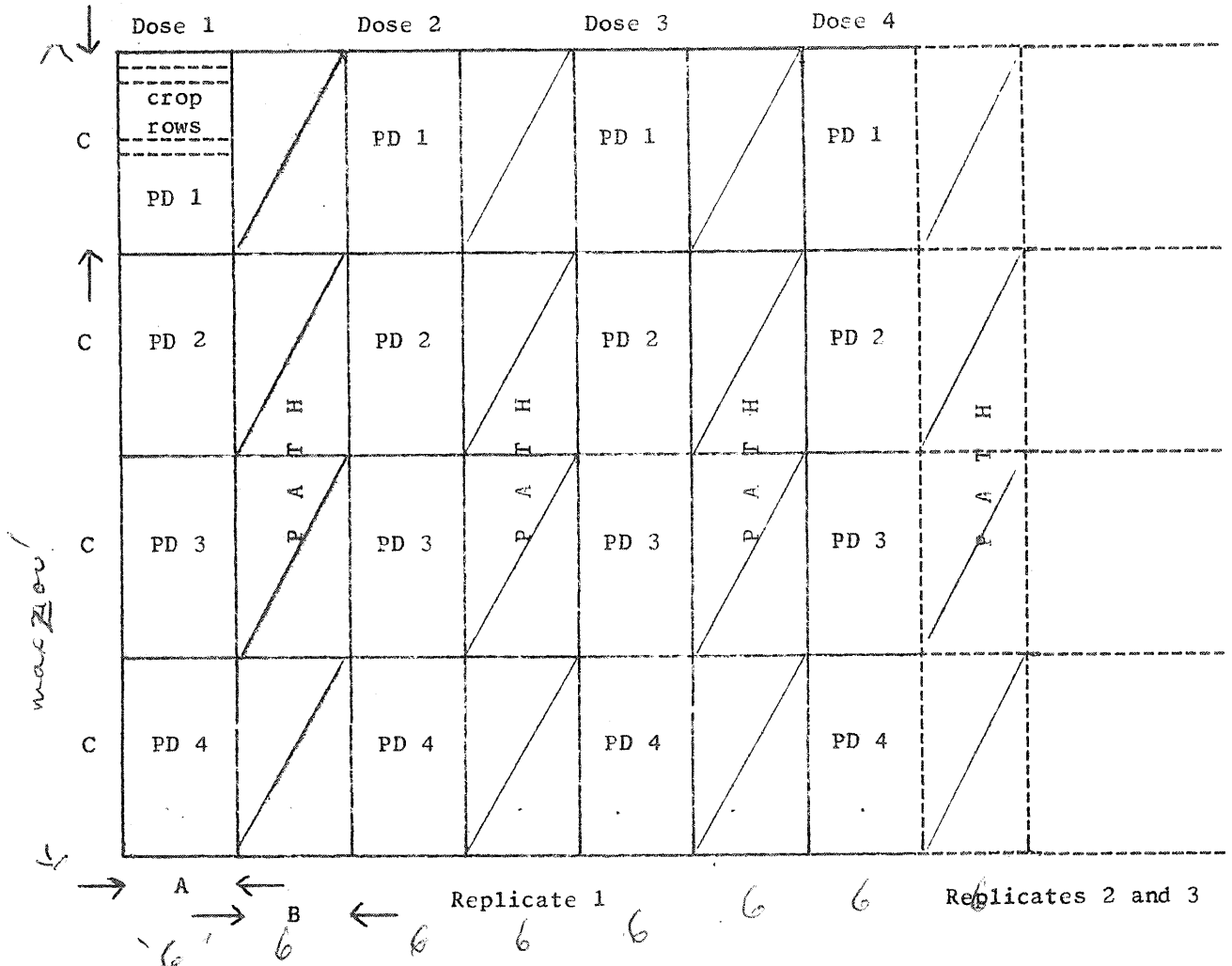
The general programme described above is also relevant to forest soils but will of course require modification according to local conditions. In the Phillipines Dr. Chandler has reported that it is likely to be possible to clear the undergrowth away to lay down the main experiments in a standing forest. In Vietnam a rubber plantation is likely to be the most suitable if a site, security and co-operation can be obtained. In both forest and mangrove there is some risk of damage to the trees, particularly from agent 'white' and agreement about this will need to be reached with the collaborator.

III AGRICULTURAL STUDIES

To obtain background knowledge of the persistency of residual effects in the soil arising from drift or the accidental application of the 'orange' and 'white' formulations to arable crops the following experimental layout is proposed (see diagram on page 7), but with separate experiments for the two formulations.

There are advantages of ease of application in having long relatively narrow plots which should be sprayed at rates of 0.0, 0.3, 1.0 and 3.0 gallons of the agent per acre. As stated earlier each formulation should be diluted to give a cover of 30 gal/ac.

SUGGESTED LAYOUT FOR AGRICULTURAL STUDY



$48'$
 48×3
 $\hline 144$
 16×3
 48
 $48 \times 30 \text{ yds}$
 480
 $\hline 1440$
 3
 $\hline 4320 \text{ yds}$
 0.8 acres

A - plot width determined by swathe width of sprayer but not less than 6 ft

B - path width not less than 6 ft (unless bunds between plots)

C - 50 ft (40 ft in rice excluded)

PD - Planting date

NOTE: Position of planting dates and doses should be randomised

At four intervals after spraying rows of seed in the case of corn (maize), sorghum, groundnut and mung beans should be sown transversally across the control and treated plots, together with cuttings of sweet potato and transplants of rice - see later. At each sowing there should be a minimum of 4 rows with records of plant establishment and growth made on the two middle rows. These observations should be restricted to the early vegetative phase - say 4-6 weeks from sowing or planting - when any abnormalities in the pattern of growth should be noted, together with final counts made at the end of the 4-6 weeks of leaf and stem or tiller number and maximum height, as minimum requirements.

After making the observations all the plants should be removed so that there will be no interference with adjacent younger plants that may have been sown at a later date.

The layout of the plots will be dependant on whether during the life of the experiment any dry period is likely to be so pronounced that it will not permit of the successful establishment and subsequent growth at each of the "sowing" dates. If drought is not a hazard, the ground is level and there is little risk of surface wash in heavy rain the plot unit should be one where the length is sufficient to accommodate the four sowing dates of all the selected crops. But, clearly these conditions will exclude paddy rice with its requirement of standing water. The simplest arrangement would be for a separate rice experiment with banded plots.

Assuming that the distance between rows will be about 1.5 feet for corn, sorghum, groundnut and mung bean and 3.0 feet for sweet potatoes the total plot length will be 200 feet (50 x 4) which would allow narrow gaps between sowing dates. If this is an awkward shape the plot could be split into 2 parallel 100 ft lengths, each with 2 sowing dates.

The width of the plots should be related to the effective width of the sprayboom and for uniformity of application plot width should be in multiples of boom width, with a minimum of 6 feet.

This split plot design is aimed at minimizing possible contamination between doses and so preferably the width of the paths separating the unit plots, compounded of dose and time of sowing, should be relatively wide - about 6 feet. Again, because of the greater mobility of the 'white' formulation the two experiments should be well separated.

If there is a danger of water stress and all the plots have to be watered from time to time the layout should be substantially the same but the details will be dependant on the method of irrigation.

The intervals between sowing dates will be determined by (i) when the experiment is started and (ii) the completion of the observations following the last sowing date. If as planned the experiments are initiated in January at the IRRI and in January or February at Mytho and all the data to be are/ready by the autumn for incorporation into the final report, the intervals between sowing dates on a logarithmic scale should be 3, 6, 12 and 24 weeks from the time of spraying.

Without a knowledge of local conditions only a guesstimate of the land requirement can be made. Allowing for a three-fold replication about an acre should suffice.

The plots should be hand-weeded as necessary to ensure that weeds do not interfere with i) growth of the crops ii) seedbed preparation and crop establishment.

Appendix

LIST OF EQUIPMENT LIKELY TO BE NEEDED

MARKING OUT PLOTS

Pegs
Labels
String
Tape measure*
Mallet or hammer

SPRAYING

Sprayer*)
Sprayboom*) pre-calibrated
4 plastic containers of 1.5 US gal capacity for mixing up spray solution
Clean water 20 gal in clean containers, e.g. petrol or jerry cans
Measuring jug 1 US gal
" " 1 pint
Stopwatch*
Plastic funnel with fine mesh filter or funnel with muslin for filtering
Rubber gloves
Light rubber or canvas-rubber boots

AERIAL APPLICATIONS

Dye
Filter paper strips*
Aluminium holders*
Baskets for collecting leaves
Canes for supporting basket clear of water

SAMPLING

Corer* (a new corer is being constructed at WRO and will be available for use in Philippines in addition to the one already in Saigon)
Double polyethylene bags* (500)
or
Metal containers (already in Saigon)
or
Plastic jars with screw tops
Plastic labels*
Pens with waterproof ink for marking*

NOTE: Items marked with * will be provided by WRO