A COMPARISON OF TWO RODENT REPELLENTS IN BROADCAST SEEDING DOUGLAS-FIR

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INTRODUCTION

Broadcast seeding of cutover lands is a promising, yet still problematical, means of tree regeneration. Substantial progress has been made since 1950, particularly with the introduction of new rodenticides and repellents suitable for direct application to the seed. One of these is tetramine (tetramethylene disulpho tetramine), a highly lethal repellent that has shown considerable success in protecting broadcast Douglas-fir seed from rodents. However, the original supplier of tetramine soon discontinued production of the chemical, primarily because of hazards associated with its manufacture. This seemingly unfortunate occurrence intensified the search for alternative protectants that would equal or exceed tetramine in effectiveness. The Wildlife Research Laboratory of the U. S. Fish and Wildlife Service at Denver, Colorado, has offered "Endrin" as a possible candidate. To test the comparative effectiveness of the two chemicals, the U. S. Forest Service cooperated in establishing a replicated field experiment early in 1955.[1/]

The test was designed to show the merits of different seed treatments in establishing seedlings from artificially sown Douglas-fir seed. Specifically, how will seed treated with Endrin compare with tetramine-treated seed? Also, will treated seed give better results than untreated seed? And, finally, what are the results when no seeding is done? Four replications of the experiment were

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1/ Cooperating were: the Olympic and Siuslaw National Forests, the Willamette and Puget Sound Research Centers, and the U. S. Fish and Wildlife Service.

installed--two on the Siuslaw National Forest in Oregon, and two on the Olympic National Forest in Washington.

TECHNIQUE

While this test is directly concerned with effectiveness of two repellents in protecting broadcast seed from rodent consumption, some basic differences between the two chemicals should be noted. When applied with a solvent that transmits some repellent into the seed, tetramine gives a degree of post-emergence protection to the seedling itself. Endrin, however, does not possess this systemic potential. While both chemicals are highly lethal to rodents, bio-assay studies by the U. S. Fish and Wildlife Service suggest that mice develop some aversion to seed treated with tetramine, but much less aversion to Endrin-treated seed. Thus, broadcast seed may be protected primarily through education in one instance (tetramine); and through population reduction in the other (Endrin). Endrin also possesses insecticidal properties that may be of considerable importance.

Previous seeding tests with tetramine have shown that the carrier used with the repellent is highly important. Notable successes have been obtained with acetone, a solvent, and yellow dextrin, an adhesive. However, neither was ideal. The former often drastically reduced the viability of treated seed, while the latter did not stand up well under weathering. A hitherto untried adhesive, methocel-rhoplex, was used with both Endrin and tetramine in the present experiment. This carrier is the result of a quest for a substance that would combine qualities of maximum adhesion and weather resistance with minimum viability reduction.

The replicated design of this experiment represents an attempt to give some regional significance to the results. However, a number of unavoidable differences exist between replications, and these differences may considerably reduce the extent of comparability. Two replications, on the Hebo District of the Siuslaw National Forest, are situated on recent cutovers designated "Austin Creek Unit B" and "Austin Creek Unit D." The two Olympic replications, located within extensively burned areas on the Soleduck District, are identified as "North Fork Calawah" and "Bear Creek." Installation and seeding of the Siuslaw testing units took place during the first two weeks of February 1955. The Olympic replications were laid out and seeded during the last week of March and the first week of April 1955. A general description of each testing unit follows.

Austin Creek Unit B lies at 500 feet above sea level on a 72-acre clearcut setting with variable aspect, light to heavy ground cover, and moderate degree of slash burn (Figure 1).

Austin Creek Unit D is situated on a 52-acre clearcut setting with an 800-foot elevation. Generally light slash burn, variable aspect, and moderate to heavy ground cover are characteristic (Figure 2).

Bear Creek lies at an elevation of about 1,000 feet within the boundaries of a 1952 wildfire. Characteristics are light ground cover, southeast aspect, and heavy burn with little slash (Figure 3.)

North Fork Calawah, also within the boundaries of an extensive wildfire (1951), is the highest unit, with a 1500-foot elevation. Since it was logged after burning, slash concentrations vary from heavy to light. Aspect is southwest and ground cover is mostly light (Figure 4).

Selection of test areas was based on careful consideration of several factors. Location in all cases was on clear-cut Douglas-fir sites of sufficient size to reduce influence of natural seedfall. Each replication consisted of four 5-acre plots selected for closest possible uniformity of soil type, aspect, slope, seedbed, and vegetative cover. Such uniformity, however, was extremely difficult to attain, and variations between plots were evident in all testing units. On the Siuslaw installation, in particular, there was considerable disparity among plots in aspect, slope, and ground cover. All replications were within areas that had been burned, either by controlled means or wildfire, but different burning intensities caused extensive variation between plots in residual slash, brush cover, and exposed mineral soil. Proximity to seed source also differed considerably among both plots and replications. Natural seedfall, nevertheless, was not a serious complication on either the Siuslaw or Olympic, due to the generally light Douglas-fir seed crops throughout the Pacific Northwest during 1954 and 1955.

All plots were square or rectangular, and plot boundaries within each replication were separated by isolation strips with a minimum width of 330 feet (somewhat less on the Siuslaw). The four plot treatments, assigned at random, were: (1) Endrin-treated seed; (2) tetramine-treated seed; (3) untreated seed; and (4) unseeded. (See Figure 5.) Seed was from two sources, and originated from low elevation areas near the locality in which it was sown. One seedlot was used

Figure 1.--Austin Creek Unit B (tetramine-treated seed plot). Vegetation mostly scattered salmonberry, with mixture of sword and bracken ferns. Many openings on soil and rotten wood. (Summer photo)

Figure 2.--Austin Creek Unit D (tetramine-treated seed plot). Predominantly herbaceous vegetation with only scattered clumps of salmonberry. Much exposed soil and rotten wood. (Summer photo)

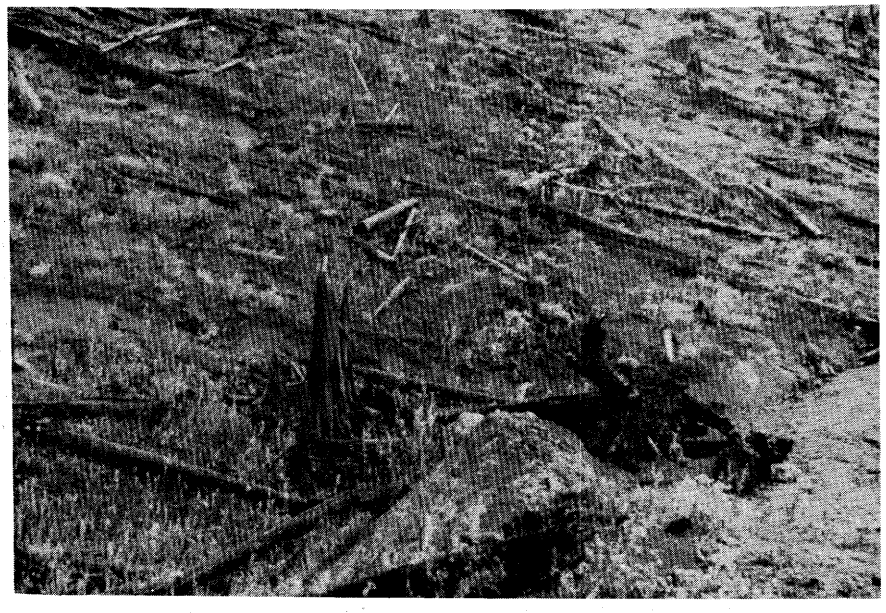

Figure 3.--Bear Creek (tetramine-treated seed plot). Severe burn with little slash. Fireweed predominates in moderate herbaceous cover. (Fall photo)

Figure 4.--North Fork Calawah (tetramine-treated seed plot). Heavy slash following logging of fire-killed timber. Herbaceous vegetation mostly light. (Fall photo)

in the Siuslaw tests; another, on the Olympic. The Wildlife Research Laboratory at Denver provided the repellent treatments for each seedlot. Seed was broadcast by cyclone hand seeders at the rate of one pound per acre on the Olympic, and three-fourths pound per acre on the Siuslaw.

To rate seeding effectiveness, each plot was sampled by 49 circular milacre sub-plots spaced systematically.[2/] Newly germinated seedlings present at time of examination were tallied and marked with painted wire stakes. Mortality could thus be determined on subsequent inventories, when stakes marking dead seedlings were removed. As a measure of seedling distribution, sub-plots were classified as "stocked" if they contained one or more seedlings.

A screened rodent exclosure was constructed on each replication to compare the effects of various treatments on germinative capacity under field conditions (Figure 6). Each exclosure contained 200 each of Endrin-treated, tetramine-treated, and untreated seed, sampled from the seed broadcast on the area. The samples were broadcast sown and covered lightly with soil for further protection. Additional samples from each seedlot were subjected to laboratory germination tests. Siuslaw seed was tested by the Oregon State College Seed Laboratory at Corvallis, Oregon; Olympic seed, by the Capital Forest Nursery near Little Rock, Washington.

Each testing unit was examined three times after seeding. Early summer examinations were made in 1955 to observe and record initial results of seeding and seed treatment. Later examinations, following both the 1955 and 1956 growing seasons, evaluated cumulative and net results. The Siuslaw tests were first examined on June 20-24, 1955, and re-examined on September 19-20, 1955 and January 30-31, 1957. The Olympic tests were examined July 12-14, 1955, November 28-30, 1955, and October 30-31, 1956.[3/]

RESULTS

On all testing units the artificially sown seed, whether treated or not, produced sufficient seedlings for successful area restocking at the end of the first growing season. However, results vary widely

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2/ Two plots have 48 milacre subplots, and another has 51.

3/ The 1956 Olympic examination, temporarily halted by snowfall, was completed November 8-9, 1956.

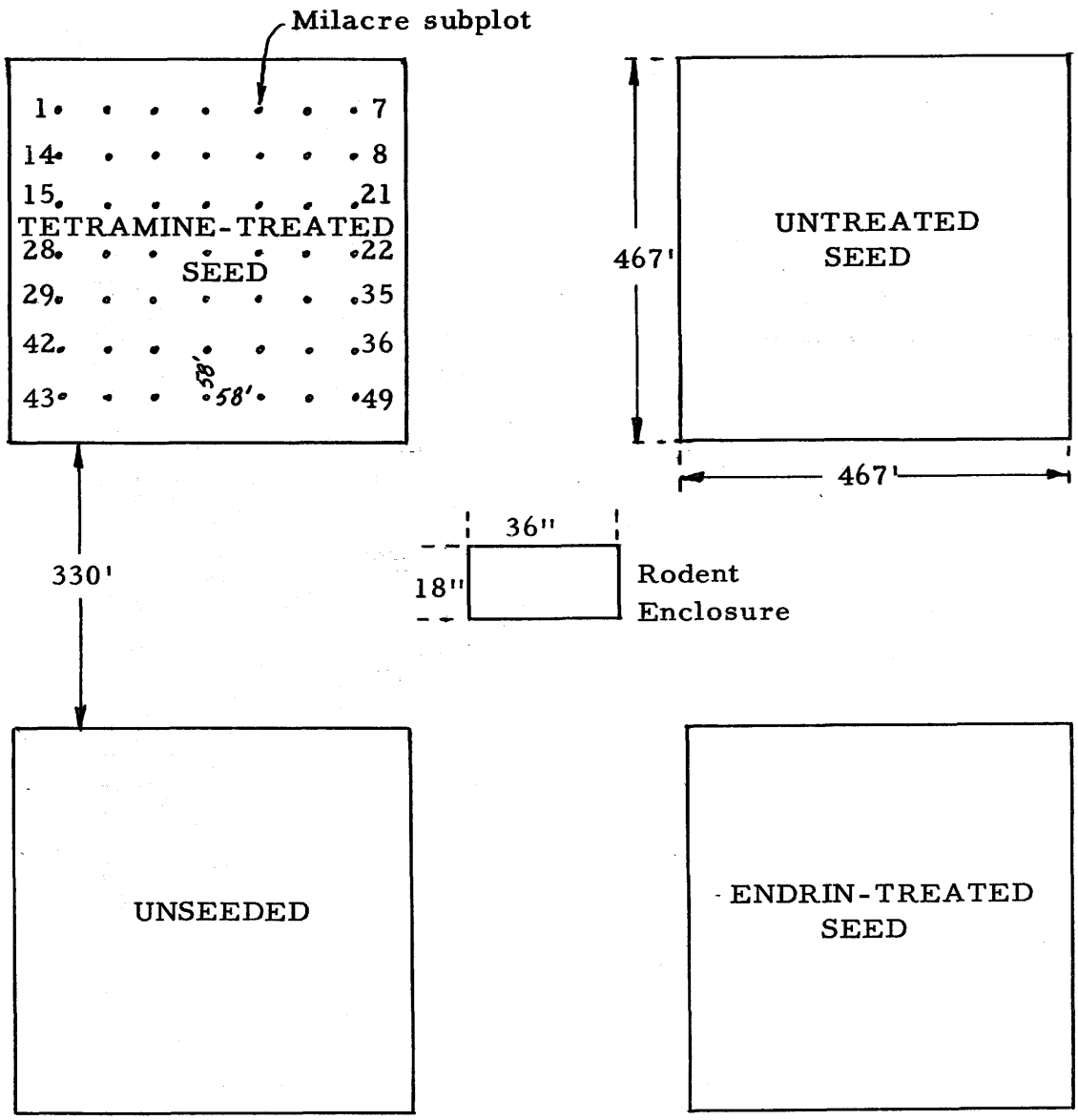

Figure 5. --Diagramatic sketch of preferred plot layout for each broadcast seeding test.

Figure 6. --Rodent exclosure located on top of small ridge in center of Austin Creek Unit B. Exclosure is 36" x 24" x 3" with 1/2" mesh hardware cloth top.

and are complicated by contradictory findings among different replications. Tetramine seed treatment gave clearly superior results on the two Siuslaw testing units; on the Olympic, best results were obtained with Endrin-treated seed. Consequently, it is impossible to rate either chemical treatment as universally superior. Unseeded check plots failed to produce seedlings in three out of four instances, and number of new seedlings on the remaining plot was negligible. This is reliable proof that natural seedfall did not seriously confound treatment effects, and, indirectly, that broadcast seeding can give positive results.

Table 1.--Laboratory germination tests of treated and untreated Douglas-fir seed

Seed lot	Tetramine-treated sample		Endrin-treated sample		Untreated sample [1]		
	#1	#2	#1	#2	#1	#2	#3
	Percent						
Siuslaw seed [2]	72	68	44	50	72	74	68
Olympic seed [3]							
Oven test	78	92	71	72	90	--	--
Table test	81	[4]47	91	[4]46	85	--	--

1/ Untreated Olympic seed was also tested by the Wind River Nursery at Carson, Washington. Germination percent was 78.

2/ All treated samples, 42 days, 20-30°C in sand. Sample #1, no chilling. Sample #2, chilled 2 weeks at 5°C. All untreated samples, 35 days, 20-30°C in sand. Sample #1, no chilling. Sample #2, chilled 2 weeks at 5°C. Sample #3, soaked 16 hours at 20°C, chilled 2 weeks at 5°C.

3/ All samples stratified in terralite for 8 weeks.

4/ Values out of line due to possible, though unexplained, mechanical failure, and are probably not representative.

The laboratory germination tests indicated with reasonable assurance that all seed was of good quality before treatment (Table 1). Reduction in germinative capacity from Endrin treatment was appreciable in the Siuslaw seedlot, but not consistently so in Olympic seed.

Field germination tests, which were screened against rodent interference, also showed that germinative capacity of Siuslaw seed was somewhat reduced by Endrin treatment (Table 2). Since repellent treatment almost invariably reduces germination, the instances that show greater germinative capacity for treated seed are difficult to interpret. Similarly, the low field germination obtained under exclosures on the Olympic is clearly out of line with laboratory germination and difficult to explain. However, second-year totals,

Table 2.--Field germination of treated and untreated Douglas-fir seed in rodent exclosures[1/]

Treatment	Time of Examination	Siuslaw N. F.		Olympic N. F.	
		Austin Creek B	Austin Creek D	Bear Creek	N. Fork Calawah
		Percent			
Tetramine	Summer '55	52.0	82.5	1.0	7.5
	Fall '55	52.5	83.0	2.0	12.0
	Fall '56	2/	2/	17.0	33.5
Endrin	Summer '55	40.0	59.0	2.5	24.5
	Fall '55	40.5	60.0	3.0	27.0
	Fall '56	2/	2/	21.0	47.5
Untreated	Summer '55	62.5	64.5	0	20.0
	Fall '55	63.5	67.0	.5	26.0
	Fall '56	2/	2/	13.5	31.5

1/ Cumulative ratings based on 200 seeds per test.

2/ Siuslaw exclosures were last rated on June 15, 1956; no new seedlings were noted at that time.

which were notably increased through delayed germination in 1956, to some extent parallel the superiority trends found on broadcast-seeded plots.

Small mammal populations were probably low on all test areas, though rodent pressure was presumably high enough to moderately limit seeding success. Rodent censusing was conducted on only one replication, Austin Creek Unit B. This census involved three separate live trappings: pre-seeding census, February 1955; post-seeding census, March 1955; and summer census, June 1955. Species of Peromyscus (deer mouse), Microtus (meadow mouse), Tamias (chipmunk), Sorex (shrew), Mustella (weasel), and Neotoma (woodrat) were included in the total catch. The latter two, however, are not considered seed eaters and have been eliminated from the census figures (Table 3). Since a small number of rodents can make serious inroads on broadcast seed, these trappings indicate some pressure. However, minimum population levels at which rodents significantly affect success of broadcast seeding have not been determined.

Table 3.--Rodent census data for Austin Creek Unit B[1/]

Treatment	Rodents trapped per 120 trap nights					
	February 1955		March 1955		June 1955	
	Number	Percent	Number	Percent	Number	Percent
Tetramine	6	5.0	[2/] 12	10.0	[3/] 19	15.8
Endrin	10	8.3	4	3.3	[2/] 16	13.3
Untreated	11	9.2	[4/] 24	20.0	[3/] 24	20.0
Unseeded[5/]	21	17.5				

1/ Values based on 40 live traps per treatment checked after each of 3 consecutive nights.

2/ Includes 2 recatches from previous census.

3/ " 4 " " " "

4/ " 5 " " " "

5/ March and June censuses were not made on this plot.

Based on cumulative number of seedlings which germinated over a 2-year period, treated seed gave better results than untreated seed in broadcast tests (Table 4). In brief, tetramine was best on the Siuslaw and Endrin on the Olympic. In all cases the second-best treatment was also one with a rodent repellent. The effectiveness of untreated seed approached that of Endrin on the Siuslaw and tetramine on the Olympic.

Table 4.--Cumulative totals of seedlings per acre and stocking at end of 1956 growing season

Replication	Treatment	Number of milacre subplots	Pre-dominant aspect	Number of seedlings per acre	Stocking percent
SIUSLAW					
Austin Creek Unit B	Tetramine	49	W+NW	1,326	63.3
	Endrin	49	NE+SW	1,041	53.1
	Untreated	49	Level	653	42.9
	Unseeded	49	NW+Level	82	6.1
Austin Creek Unit D	Tetramine	49	N	2,856	77.6
	Endrin	48	NW	1,083	50.0
	Untreated	49	W	959	57.1
	Unseeded	49	S	0	0
OLYMPIC					
Bear Creek	Tetramine	49	SE+Level	1,388	51.0
	Endrin	49	SE	15,775 1/	69.4
	Untreated	51	S	1,117	45.1
	Unseeded	49	SE	0	0
North Fork Calawah	Tetramine	49	SW	1,408	51.0
	Endrin	48	W	3,958	56.2
	Untreated	49	W	633	30.6
	Unseeded	49	W	0	0

1/ This value is highly influenced by 2 milacre subplots with 118 and 207 seedlings, respectively. A recalculated value, eliminating these subplots, is 9,532 seedlings per acre.

Endrin was apparently more phytotoxic to Siuslaw seed than to Olympic seed, though it is not clear whether differences in seed or in treatment were primarily responsible. Phytotoxicity of the Endrin treatment was also reflected in results of laboratory and field germination on the Siuslaw. A corresponding assumption that tetramine treatment was phytotoxic to Olympic seed, does not seem particularly valid since somewhat indifferent seedling success with tetramine was counterbalanced by good laboratory germination. Field germination in exclosures on the Olympic was too irregular to lend any real assistance in further assessing this inconsistency.

The erratic behavior of both chemical seed treatments in these tests indicates a complexity of interacting variables, which the present experiment cannot reconcile. Further refinement in testing technique and increased replication appear necessary to overcome such factors in an experiment of this type.

Analysis of variance was used in a statistical comparison of treatment effects for the Olympic and Siuslaw tests combined. Averages (Table 5) of seedling and stocking totals--both cumulative and net--were compared at the five percent probability level.[4/] Net figures are based on only those seedlings that were living at the time of the final examination. Cumulative figures include, in addition, seedlings that germinated but died prior to the final examination. Since these averages mask wide disparities in numerical count and reversed treatment superiorities on different replications, significant differences were found for only two out of twelve comparisons, as shown below:

Standard	Comparison	Difference
Cumulative number seedlings per acre	Tetramine vs. Endrin	3,720
	Tetramine vs. untreated	904
	Endrin vs. untreated	4,624
Net number seedlings per acre	Tetramine vs. Endrin	2,115
	Tetramine vs. untreated	548
	Endrin vs. untreated	2,663

4/ Since equal error variance for the unseeded treatment could not logically be anticipated, this treatment was not included in the analysis. However, the almost complete failure of the unseeded plots to produce seedlings is, in itself, indicative of seeding effectiveness.

Standard	Comparison	Difference
Cumulative stocking (percent)	Tetramine vs. Endrin	4
	Tetramine vs. untreated	17
	Endrin vs. untreated	13
Net stocking (percent)	Tetramine vs. Endrin	1
	Tetramine vs. untreated	18*
	Endrin vs. untreated	17*

The above gives some mathematical evidence that treated seed outproduced untreated seed, but differences between repellents cannot be attributed to more than chance occurrence.

Table 5.--Seedling and stocking totals after two growing seasons--average of all replications

Treatment	Seedlings per acre: Cumulative	Seedlings per acre: Net	Stocking: Cumulative	Stocking: Net
	number	number	percent	percent
Tetramine	1,744	1,030	61	44
Endrin	1/ 5,464	1/ 3,145	57	43
Untreated	840	482	44	26

1/ Statistical comparisons were also made that reflected elimination of extraordinary values on two milacre subplots. These comparisons were non-significant.

Re-examinations in 1955 and 1956 clearly showed that considerable delayed germination occurred on all replications following the initial seedling tally early in 1955 (table 6). Seedlings recruited after the first examination were 54.5 percent of the cumulative total number. Regional differences of 31.2 percent on the Siuslaw and

*Significant at 5 percent.

62.2 percent on the Olympic undoubtedly reflect local influences in addition to those of seeding and examination date.[5/] Stocking percentages also showed a considerably greater net increase on the Olympic than on the Siuslaw.

Increases in stocking and number of seedlings were particularly striking for Olympic tetramine-treated seed. Net results at the end of 1956 compared favorably to those attained with this repellent on the Siuslaw. Though mortality was generally heavier during 1956, it was not excessive in either study area during the two growing seasons. A number of live seedlings examined on the Olympic had tap roots up to 8 inches long at the end of 1955.

SUMMARY AND CONCLUSIONS

Test areas on the Olympic and Siuslaw National Forests were successfully restocked to Douglas-fir by broadcast seeding. Rodents were judged to be at least moderately limiting to the success of direct seeding on these areas. Results were best when seed was treated with a chemical rodent repellent, and poorest when seed was untreated. Endrin and tetramine, the two repellents under test, were inconsistent in superiority--the former ranked first on the Olympic; the latter gave best results on the Siuslaw. Unseeded check areas showed that these results were not complicated by regeneration resulting from natural seedfall, and further demonstrated that artificial broadcast seeding can be a successful means of securing initial regeneration. More refined experimental technique and increased replication are probably necessary in broadcast seeding tests.

5/ The large amount of second-year germination on the Olympic undoubtedly was highly influenced by late (early spring) seeding.

Table 6. --Net totals of seedlings per acre and stocking, 1955 and 1956 growing seasons

Replication	Treatment	Number of seedlings: Summer 1955	Died	Re-cruited	Fall 1955	Died	Re-cruited	Fall 1956	Stocking Percent: Summer 1955	Fall 1955	Fall 1956
SIUSLAW											
Austin Creek Unit B	Tetramine	898	326	367	939	592	61	408	49.0	49.0	24.5
	Endrin	816	245	225	796	531	0	265	40.8	42.9	20.4
	Untreated	531	143	102	490	245	20	265	34.7	34.7	16.3
	Unseeded	61	0	21	82	62	0	20	4.1	6.1	2.0
Austin Creek Unit D	Tetramine	1,898	571	734	2,061	816	224	1,469	63.3	61.2	57.1
	Endrin	667	125	229	771	229	187	729	35.4	41.7	41.7
	Untreated	633	122	203	714	306	123	531	40.8	40.8	34.7
	Unseeded	0	0	0	0	0	0	0	0	0	0
OLYMPIC											
Bear Creek	Tetramine	20	0	429	449	184	939	1,204	2.0	24.5	51.0
	Endrin	1/ 7,449	653	3,469	1/ 10,265	6,245	4,857	1/ 8,877	53.1	63.3	53.1
	Untreated	333	39	510	804	333	274	745	17.6	39.2	31.4
	Unseeded	0	0	0	0	0	0	0	0	0	0
North Fork Calawah	Tetramine	61	61	490	490	306	857	1,041	6.1	32.7	42.9
	Endrin	1,271	354	1,354	2,271	896	1,333	2,078	35.4	50.0	56.2
	Untreated	41	0	408	449	245	184	388	4.1	28.6	22.4
	Unseeded	0	0	0	0	0	0	0	0	0	0

1/ Recalculated to reflect elimination of 2 milacre subplots with extraordinary seedling counts, these values become 4,170; 5,681; and 4,851, respectively.

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