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INTERNATIONAL UNION FOR THE PROTECTION OF NEW VARIETIES OF PLANTS

GENEVA

TECHNICAL COMMITTEE

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COMBINED OVER-YEARS (COY) CRITERION FOR DISTINCTNES IN DUS TRIALS

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SUMMARY OF THE USE OF THE COY CRITERION

Introduction

(i) Following advice from the Technical Working Party for Automation and Computer Programs (TWC), the Technical Committee at its meeting in November 1986 recommended that for grass species the combined over-years (COY) criterion be used to assess distinctness. This document describes this analysis in comparison with previously used criteria and explains a refinement of it which although not part of the criterion is under consideration by the TWC. The following paragraphs summarize the information provided in this paper.

Previously Used Criteria

(ii) The previous UPOV criterion for distinctness required that a difference between varieties should be statistically significant at the 1% probability level in at least 2 out of 3 years for one or more measured characteristics. This is called the 2x1% rule and the technical experts who accepted it recognized the importance of seeking repeatability of significant variety differences over years.

(iii) This rule was criticized because a difference between varieties which just fails to achieve the 1% significance level contributes no more to the assessment of distinctness than for instance, a zero difference in one year or even a non-significant difference of the opposite sign. If the differences between varieties are in the same direction for each of 3 years with one difference significant at the 1% level and 2 differences at the 5% level this would not, under the 2x1% rule, be sufficient evidence of distinctness. An intuitive interpretation of 3 such results suggests that the varieties are distinct and that information from all 3 years should be taken into account simultaneously when making distinctness decisions.

(iv) In an attempt to overcome this weakness a modification of the 2x1% criterion referred to as the t-score criterion was introduced. In this criterion distinctness is based on a calculation which uses the results from all 3 years, but does not allow one very extreme result to outweigh the evidence from the other 2 years. Since under this calculation all variety pairs distinct using the 2x1% criterion are also distinct using t-score, the t-score criterion is less strict than the 2x1% criterion though in practice the relaxation in standard is not large.

Combined Over-Years (COY) Criterion

(v) Although the previously used criteria included a requirement for repeatability over years they are based on the plot error within trials and hence do not take into account variety variation over years. It can be shown that some characteristics are much less consistent than others over years. Where decisions on distinctness are based on characteristics which are consistent from year to year, with the previously used criteria, there is relatively low risk that repeated tests in a later year would lead to a different decision. However using the same criteria decisions based on less consistent characteristics have a greater risk that they could not be reproduced. Hence with the 2x1% and t-score criteria the risks involved in making decisions are of varying magnitude depending on the over-year consistency of the characteristics on which distinctness is based. This is unsatisfactory for both testing authority and breeder. The COY criterion was introduced to overcome this difficulty. It provides probability levels of differences between variety means over years occurring by chance if no difference exists where the differences are compared with over-year variation. The choice of probability level to use with the COY criterion is still under consideration by the TWC and is being examined by member States over the next few years using circulated computer programs. In the meantime the Technical Committee has suggested that at least the 5% level of significance is used.

(vi) An F-ratio statistic, F_3, is included in the over-years criterion to identify any excessive variation in the differences between a candidate variety and a control compared with the general variety x years variation over the 3 years of test. The combined over-years criterion should be treated with caution where a significant value of F_3 occurs.

Modified Joint Regression Analysis (MJRA) Adjustment to the COY Criterion

(vii) At its meeting in Hanover, 1986, German experts proposed a refinement to the COY analysis to allow for an exceptional change in the spacing between variety means on a characteristic in one of the 3 test years due to environmental conditions e.g. the convergence of heading dates in a late spring. It was considered that this systematic variety x year variation should be excluded from the variation on which the significance of differences is based in the COY criterion. The refinement, known as Modified Joint Regression Analysis (MJRA) adjustment, has not yet been sufficiently evaluated to be included in the COY criterion. However, it has been added as an option in the COY computer program circulated to member States for their investigation.

Computer Programs

(viii) A magnetic tape containing computer programs to enable all the above criteria to be carried out has been circulated to member States represented at the fifth TWC session. Further copies can be provided on request from UPOV.

DESCRIPTION AND USE OF THE COY CRITERION

Introduction

1. The Technical Committee has decided to introduce the combined over-years criterion as the official UPOV method for determining distinctness for grass species. This note describes the COY criterion and a modification of it involving the modified joint regression analysis (MJRA). The previously used criteria are also defined to provide a background to the reasons for the change to the COY criterion. In addition computer programs to enable the criteria to be compared on actual data are described. These programs are being forwarded on magnetic tape to the member States who were represented at the fifth TWC session and can be supplied on request to other member States.

Previously Used Criteria

2. The original UPOV distinctness criterion was the 2x1% criterion. This was subsequently modified to the t-score criterion. The following is a brief description of these criteria.

2x1% Method

3. This criterion for distinctness of varieties was based on separate t-tests carried out in each year of test between a candidate variety and each other variety. These use the t-values defined by

$$t = \frac{\bar{x}_1 - \bar{x}_2}{\sqrt{2}\,SE(\bar{x})}$$

where $\bar{x}_1$, $\bar{x}_2$ are the means on a measured characteristic of the two varieties being compared and $SE(\bar{x})$ is the standard error of a variety mean estimated from the plot error mean square. The significance level of a calculated t-value is obtained from t-tables using the degrees of freedom of the plot error mean square. For each comparison between a candidate variety and some other variety one test is available in each of the years that the candidate variety was grown in the trials, either 2 or 3 years. Under this criterion the candidate variety is considered to be distinct from another variety if in at least 2 years the t-value is significant at the 1% level in the same direction. This criterion established that repeatability of differences over years is an important part of distinctness.

T-Score Method

4. A difficulty with the 2x1% method is that a within-year difference which just fails to achieve the 1% significance level contributes no more to the separation of a variety pair than a zero difference or even a non-significant difference of the opposite sign. For example, 3 differences in the same direction one of which is significant at the 1% level and the others at the 5% level would not be regarded as sufficient evidence for distinctness. The t-score method was introduced to overcome this weakness. In this method the t-value calculated according to formula (1) is converted to a t-score using

constants k_1 and k_2 where these are the tabulated t-scores at the 5% and 0.1% levels respectively. The conversion from t-values to t-scores is then defined by:

(i) t-score = 0 if $-k_1 < t < k_2$
(ii) t-score = t-value if $k_1 < t < k_2$ or $-k_2 < t < -k_1$
(iii) t-score = k if $t > k_2$
(iv) t-score = -k if $t < -k_2$

5. These conversion rules are illustrated in Figure 1. Using t-scores two varieties are distinct if the absolute sum of their t-scores over 3 years exceeds the critical value 5.2 which is equivalent to twice the tabulated t-value at the 1% significance level with a large number of degrees of freedom. As with the 2x1% criterion the t-score method requires more than a single large t-value for distinctness but the confirmatory evidence need not be so strong. Thus three 5% results, provided they are in the same direction are sufficient to ensure distinctness. With regard to the stringency of the t-score criterion compared with the 2x1% criterion since all variety pairs distinct using the 2x1% criterion are also distinct using the t-score criterion and some additional pairs are distinct using t-score, the t-score criterion is less strict though in practice the relaxation in standard is not large.

The Combined Over-Years Criterion (COY)

6. Although the previously used criteria included a requirement for repeatability over years they were based on the plot error mean square and hence did not take into account variety variation over years. Since variety measurements are less consistent on some characteristics over years than on others the use of these criteria resulted in acceptance standards varying between characteristics. Inconsistency is indicated by large values of λ where

$$\lambda^2 = \frac{\text{varieties x years mean square}}{\text{plot error mean square}}$$

and consistency by values of λ near to 1. Values of λ from a specimen set of trial data are given in Table 1 and show a range from 1.3 to 2.2 indicating different variation over years for different characteristics. The COY criterion meets the above criticism by using the varieties x years mean square to estimate the error instead of the plot error mean square. Using this criterion two varieties are distinct if the absolute value of

$$\frac{\bar{\bar{x}}_1 - \bar{\bar{x}}_2}{\sqrt{2}\, SE\,(\bar{\bar{x}})}$$

is larger than a specified critical percentage point in the distribution of t where $\bar{\bar{x}}_1$, $\bar{\bar{x}}_2$ are the means of the varieties over 3 years and $SE(\bar{\bar{x}})$ is the standard error of a variety mean calculated from the variety x years mean square.

7. Thus this criterion is based on a single analysis of variance for each characteristics. For m years and n varieties this analysis of variance breaks down the available degrees of freedom as follows:

Source	DF
Years	m-1
Varieties	n-1
Varieties x years	(m-1)(n-1)

Using this criterion the ration, F, defined as

$$F = \frac{\text{varieties mean square}}{\text{varieties x years mean square}}$$

provides a measure of the discriminating power of a characteristic. Thus Table 1 shows that characteristics 5 and 8 are the most discriminating characteristics and 4 and 14 are the least discriminating for this data set. The choice of the critical probability level to use with this criterion is still under consideration by the TWC. It has been determined from theoretical considerations that for a 3 year test the COY criterion applied at the 1% probability level is of approximately the same stringency as the 2x1% criterion on a characteristic with a λ value of 1.7 Compared with the 2x1% criterion the COY criterion applied at the 1% level is less stringent for characteristics with values less than 1.7 and more stringent for characteristics with values greater than 1.7. Similarly for a 2 year test it has been determined theoretically that the 2x1% criterion and the COY criterion applied at the 0.1% level have similar acceptance standards when λ =1.5. In the United Kingdom the λ values which occur are generally slightly smaller than these and hence it has been found in practice that the COY criterion operated at the 1% level for a 3 year test and at the 0.1% level for a 2 year test provides a slightly less strict standard than the 2x1% standard and is more similar to the t-score standard. However other member States have reported different results and a final decision on the probability levels to use with the COY criterion has yet to be taken. In the meantime the Technical Committee has agreed that a level of at least 5% is used until further information is obtained from member States.

Homogeneity of Varieties x Years Variance

8. From the previous paragraphs it is recommended that the combined over-years criterion be used in preference to the 2x1% or the t-score methods with one qualification. The varieties x years mean square used in the combined over-years criterion is a pooled value calculated from a large number of varietal comparisons and hence may not be appropriate to any particular comparison. Hence to ensure that the specific within pair varieties x years mean square with 2 degrees of freedom is not larger than the pooled varieties x years mean square the ratio, F_3 say, should be calculated and tested for significance. The combined over-years criterion should be treated with caution where a significant F_3 ratio occurs.

Modified Joint Regression Analysis (MJRA) Adjustment to the COY Analysis

9. As pointed out above the COY criterion uses the varieties x years variation on which to base the SE of a variety mean. When considering the varieties x years interaction two sources of variation can be identified. Firstly a systematic effect causing the occurrence of different slopes of the regression lines relating variety means in individual years to the average variety means

over all years. Such an effect can be noted for the heading date characteristic where in a year with a late spring the range of heading dates can be compressed compared with the normal leading to a reduction in the slope of the regression line for variety means in that year versus average variety means. Secondly a non-systematic effect represented by the variation about these regression lines. Where only non-systematic varieties x years variation occurs the slope of the regression lines have the constant value 1.0 in all years but when systematic variation is present slopes differing from 1.0 occur but with an average of 1.0. When the MJRA is used the SE of variety mean is based on the non-systematic part of the varieties x year variation.

10. The distinction between the total varieties x years variation and the varieties x years variation adjusted by MJRA is illustrated in Figure 2 where variety means in each of 3 years are plotted against average variety means over all years. The variation about 3 parallel lines fitted to the data, one for each year, provides the total varieties x years variation as used in the COY criterion described above. These regression lines have the common slope 1.0. This variation may be reduced by fitting separate regression lines to the data, one for each year. The resultant residual variation about the individual regression lines provides the MJRA adjusted varieties x years mean square. It can be seen that this adjustment is only effective where the slopes of the variety regression lines differ between years such as can occur in heading dates.

11. The use of this technique in determining distinctness was suggested by Dr. Laidig, Federal Republic of Germany, and has been included as an option in the distinctness computer programs for evaluation by member States.

Computer Programs

12. Three computer programs are available for evaluating varietal distinctness and the stringency of criteria. Relationships between them and their data files are shown in Figure 3. Operating instructions and test data with sample outputs are available and are being forwarded separately to member States represented at the fifth TWC session. The following is a brief description of each program.

13. ANAL: The function of ANAL is to provide variety means and other statistics from plot means in a single year. Plot means for a range of characteristics are held in an 'E' file with associated information on variety and characteristic names. Specified parameters are held in a file named ANAL.DAT and resulting variety means with related statistics are provided in an 'M' file. Missing plot values should be entered as -1 and these are fitted using the NAG subroutine F01ABF which is not included in the circulated program. The function of this subroutine is to invert a symmetric matrix and could be replaced by a similar subroutine if the NAG library is not available locally. However in practice missing values rarely occur and the program will operate without the NAG subroutine if none are present in the data being analyzed.

14. TVAL: This program provides comparisons between specified variety pairs using the 3 criteria, 2x1%, t-score and COY. The COY criterion can be used with or without the MJRA adjustment. In the latter case its use is clearly indicated on the outputs. The use of this program is also shown on Figure 3. Variety means and related statistics are read from 'M' files, one for each

year. The selection of data from these files for control and candidate varieties is achieved through a separate computer file which contains for each variety to be included in the analysis its code number in each year of test. Characteristics to be included and the 'M' files in which the data are to be found are also specified in this file together with a list of the varieties for which comparisons are required.

15. TSUM: This program enables the alternative criteria to be evaluated more easily by a re-arrangement of the output to provide a summary of the distinctness status of each variety nominated for comparison tests without the detail provided by TVAL. It operates in a similar way and reads the same parameter file. Its use is also specified in Figure 3. However to facilitate its operation in practice, 3 frequently changed parameters are held in a smaller file named TSUM.DAT. This avoids unnecessary alteration of the TVAL parameter file which is required for normal processing.

TABLE 1: Summary of over-year analyses of variance for a specimen set of trials (Early Perennial Ryegrass (Diploid) Crossnacreevy, UK, 1982-84)

Characteristics	Mean squares/plot					
	Years (Y)	Varieties (V)	Varieties x Years (VxY)	Plot Error (E)	$F_1 = V/(VxY)$	$\lambda = \sqrt{(VxY)/E}$
4 Angle year of sowing	749.3	237.5	29.30	15.00	8.10	1.40
5 Spring height	1919.2	393.0	12.05	5.55	32.6	1.47
8 Date of ear emergence	4506.4	307.1	9.52	4.13	32.3	1.52
10 Height at ear emergence	6111.2	620.7	24.47	9.44	25.4	1.61
11 Width at ear emergence	4134.3	332.3	22.34	13.33	15.9	1.30
14 Flag leaf length	1193.5	51.3	4.79	1.00	10.7	2.19
15 Flag leaf width	81.4	2.6	0.24	0.12	10.9	1.41
17 Stem length	10474.1	593.6	52.89	13.39	11.2	1.99
20 Height in aftermath	149.2	341.4	21.35	12.16	16.0	1.33
24 Ear length	1677.4	47.6	2.58	1.51	18.5	1.31

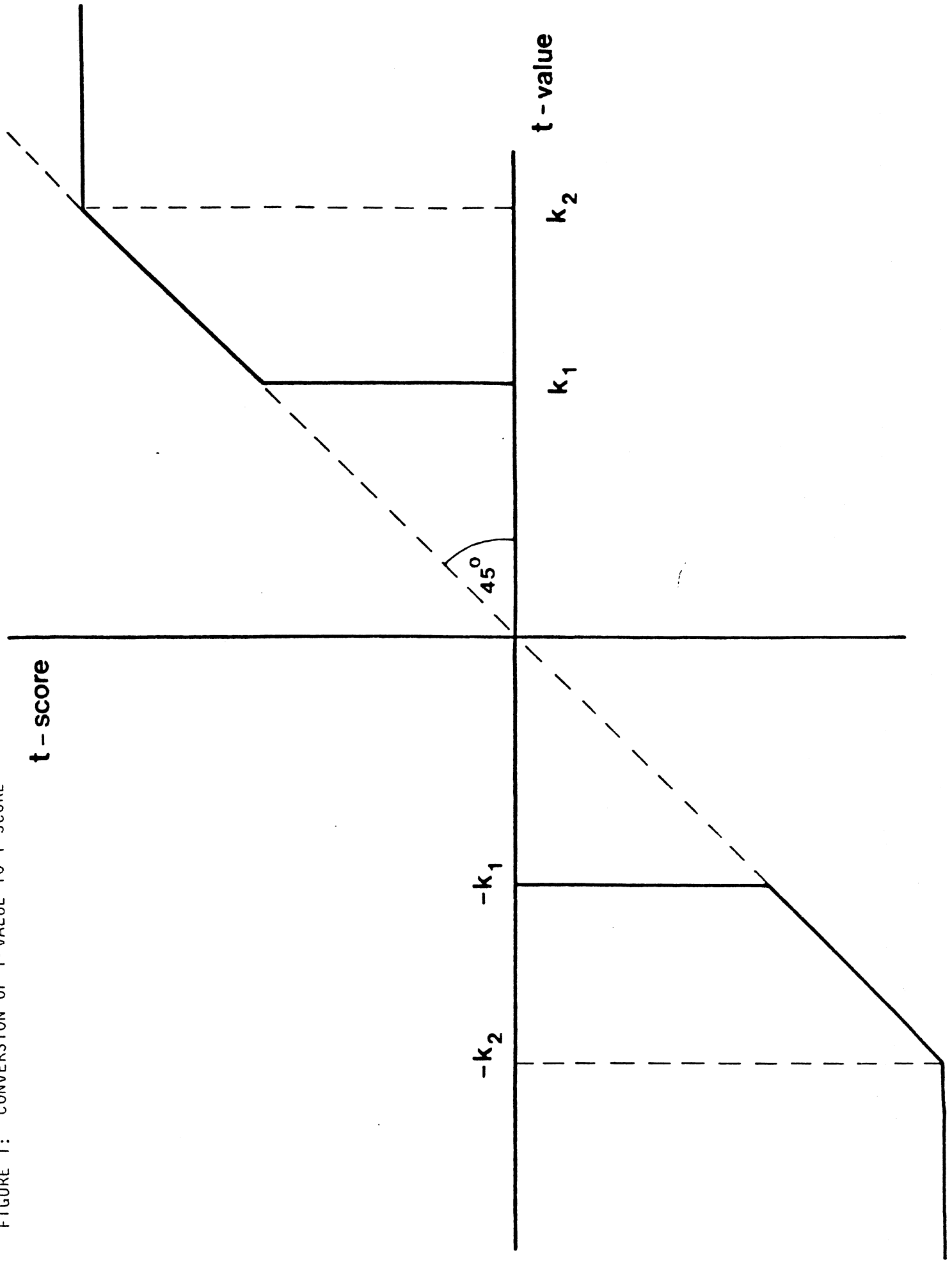


FIGURE 1: CONVERSION OF T-VALUE TO T-SCORE

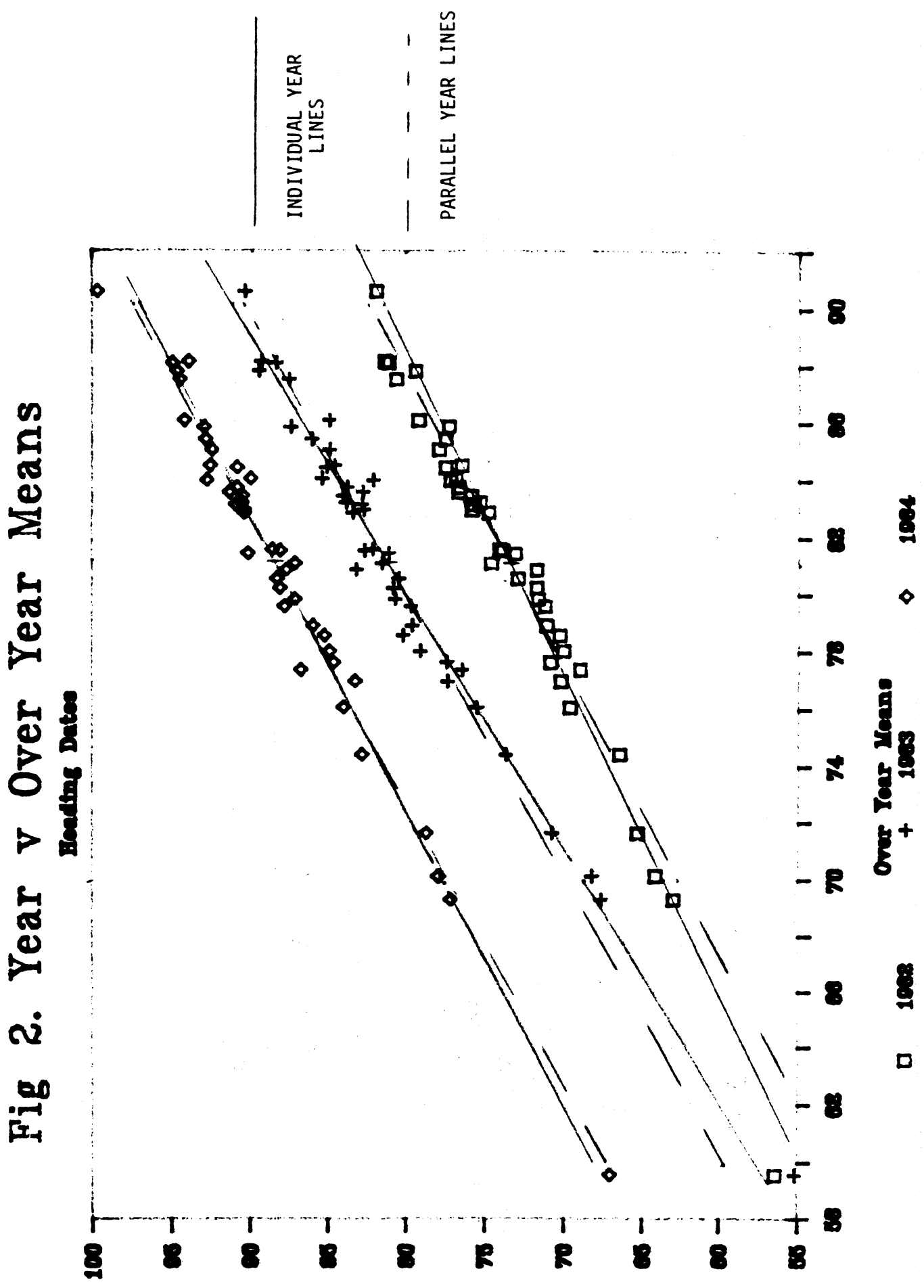
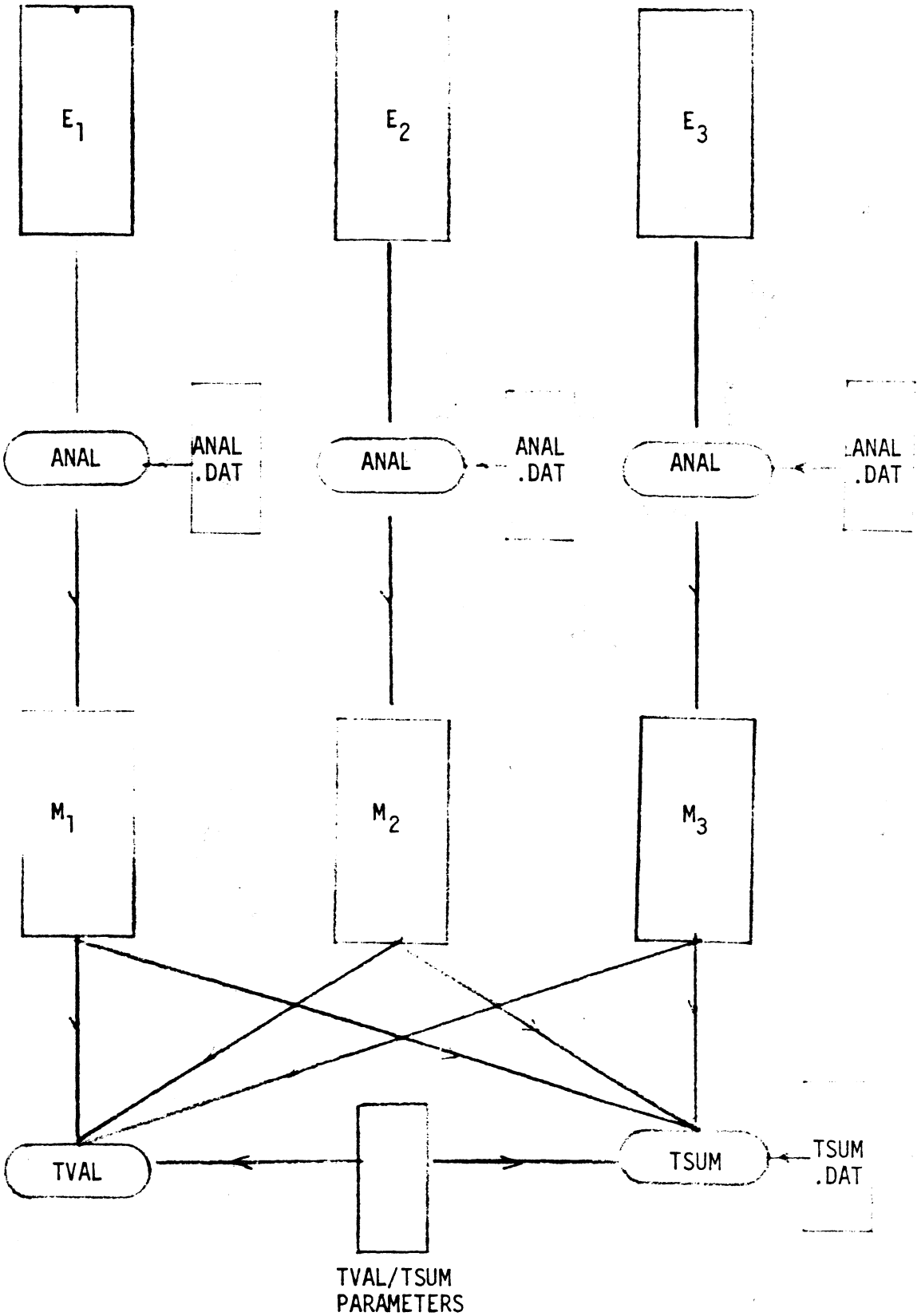


Fig 2. Year v Over Year Means

FIG 3: RELATIONSHIP BETWEEN DATA FILES AND DUS PROGRAMS



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