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STATISTICAL METHODS FOR VISUALLY OBSERVED CHARACTERISTICS

Document prepared by the Office of the Union

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EXECUTIVE SUMMARY

1.   The purpose of this document is to report on developments concerning "Statistical Methods for Visually Observed Characteristics".

2.   The TWC is invited to:

    (a)   note that China has been invited to make a presentation at the thirty-fourth session of the TWC to describe the statistical methods used in the DUSTC software package for the analysis of distinctness and uniformity;

    (b)   consider appropriate naming of the different methods to avoid confusion with other methods used in UPOV, such as COYD;

    (c)   note that a study on the development of software to implement the method developed by experts from Denmark and Poland would be reported to the TWC, at its thirty-fifth session by an expert from France; and

    (d)   consider the guidance presented in the Annex to this document as the basis for guidance to be proposed for inclusion in a future revision of document TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability".

3.   The following abbreviations are used in this document:

    TC: Technical Committee
    TC-EDC: Enlarged Editorial Committee
    TWA: Technical Working Party for Agricultural Crops
    TWC: Technical Working Party on Automation and Computer Programs
    TWF: Technical Working Party for Fruit Crops
    TWO: Technical Working Party for Ornamental Plants and Forest Trees
    TWPs: Technical Working Parties
    TWV: Technical Working Party for Vegetables

4.   The structure of this document is as follows:

ANNEX   New Statistical Method for Visually Observed Characteristics with Multinomial Distributed Data (in English only)

BACKGROUND

5.   The background to this matter is provided in documents TWC/33/26 and TC/52/23 "Statistical Methods for Visually Observed Characteristics".

DEVELOPMENTS IN 2015

6.   At their sessions in 2015, the TWV, TWC and TWA considered documents TWV/49/20, TWC/33/26, and TWA/44/20 "Statistical Methods for Visually Observed Characteristics", respectively.

7.   The TWV, TWC, TWA, TWF and TWO noted that the TC, at its fifty-first session, had agreed to remove the document "Statistical Methods for Visually Observed Characteristics" from the program for the revision of document TGP/8, and to consider the matter under a separate agenda item (see documents TWV/49/32 Rev. "Revised Report", paragraph 77, TWC/33/30 "Report", paragraph 70, TWA/44/23 "Report", paragraph 60, TWF/46/29 Rev. "Revised Report", paragraph 62 and TWO/48/26 "Report", paragraph 71, respectively).

8.   The TWV, TWA, TWF and TWO noted that the TWC had invited an expert from China to make a presentation at the thirty third session of the TWC on the analysis of visually observed characteristics using the DUST China (DUSTC) software package using the data set of meadow fescue provided by Finland (see documents TWV/49/32, paragraph 78, TWA/44/23, paragraph 61, TWF/46/29 Rev., paragraph 62 and TWO/48/26, paragraph 73, respectively).

9.   The TWF agreed that statistical methods were not routinely used for fruit crops (see document TWF/46/29 Rev. "Revised Report", paragraph 61). The TWO agreed that statistical methods were not used for the analysis of visually observed characteristics in DUS examination of ornamental plants (see document TWO/48/26 "Report", paragraph 70).

10.   The TWC noted the presentations by members of the Union on how they intended to use the new statistical method for visually observed characteristics in DUS examination, reproduced in the Annex to this document (in English only) (see document TWC/33/30, paragraphs 69 to 76).

11.   The TWC received a presentation by an expert from China on "Analysis of visually observed characteristics using the DUST China (DUSTC) software package" using the data set for growth habit in meadow fescue provided by Finland. A copy of the presentation is provided in document TWC/33/26 Add.1.Rev.

12.   The TWC agreed to invite China to make a presentation at the thirty-fourth session of the TWC to describe the statistical methods used in the DUSTC software package for the analysis of distinctness and uniformity.

13.   The TWC received a presentation on "Ways in which members of the Union intend to use the new statistical method for visually observed characteristics in DUS examination" by an expert from Finland. A copy of the presentation is provided as document TWC/33/26 Add.2.

14.   The TWC noted that Finland intended to use the new statistical method for the analysis of seven visually observed ordinal characteristics in Timothy, Meadow Fescue and Tall Fescue, White Clover and Red Clover.

15.   The TWC agreed that the naming of the different methods should be clarified to avoid confusion with other methods widely used in UPOV, such as COYD.

16.   The TWC welcomed the offer by an expert from France to study the development of software to implement the method developed by experts from Denmark and Poland (see document TWC/30/19 "Consequences of Decisions for DUS Examination when using Statistical Methods for Visually Observed Characteristics"), in collaboration with experts from Finland and the United Kingdom.

DEVELOPMENTS IN 2016

17.   The TC, at its fifty-second session, held in Geneva, from March 14 to 16, 2016, considered document TC/52/23 "Statistical Methods for Visually Observed Characteristics".

18.   The TC noted that the TWF had agreed that statistical methods were not routinely used for fruit crops, and that the TWO had agreed that statistical methods were not used for the analysis of visually observed characteristics in DUS examination of ornamental plants.

19.   The TC noted that China had been invited to make a presentation at the thirty-fourth session of the TWC to describe the statistical methods used in the DUSTC software package for the analysis of distinctness and uniformity.

20.   The TC noted that Finland intended to use the new statistical method described in the Annex to document TC/52/23 for the analysis of seven visually observed ordinal characteristics in Timothy, Meadow Fescue and Tall Fescue, White Clover and Red Clover.

21.   The TC agreed that the naming of the different methods should be clarified to avoid confusion with other methods used in UPOV, such as COYD.

22.   The TC noted that the TWC had welcomed the offer by an expert from France to study the development of software to implement the method developed by experts from Denmark and Poland, in collaboration with experts from Finland and the United Kingdom.

23.   On March 14, 2016, the expert of France informed the Chairperson of the TWC that the study on the development of software to implement the method developed by experts from Denmark and Poland would be reported to the TWC, at its thirty-fifth session.

24.   The TWC is invited to:

(a)   note that China has been invited to make a presentation at the thirty-fourth session of the TWC to describe the statistical methods used in the DUSTC software package for the analysis of distinctness and uniformity;

(b)   consider appropriate naming of the different methods to avoid confusion with other methods used in UPOV, such as COYD;

(c)   note that a study on the development of software to implement the method developed by experts from Denmark and Poland would be reported to the TWC, at its thirty-fifth session by an expert from France; and

(d)   consider the guidance presented in the Annex to this document as the basis for guidance to be proposed for inclusion in a future revision of document TGP/8 "Trial Design and Techniques Used in the Examination of Distinctness, Uniformity and Stability".

[Annex follows]

NEW STATISTICAL METHOD FOR VISUALLY OBSERVED CHARACTERISTICS
WITH MULTINOMIAL DISTRIBUTED DATA

I.   ORDINAL CHARACTERISTICS

Summary of requirements for application of the method

- The method is appropriate to use for assessing distinctness of varieties where:
- The characteristic is ordinal and recorded for individual plants (usually recorded visually)
- There are some differences between plants
- The observations are made over at least two years or growing cycles on a single location
- There should be at least 20 degrees of freedom for estimating the random variety-by-year interaction term.
- The distribution of the characteristic should be unimodal, i.e. notes with large number of plants should occur next to each other, zeros at one or both ends of the scale should not cause problems as long as most varieties have plants that fall in different notes
- The total number of plants for each variety should not be too low, at least 5 times the number of notes the variety covers

Summary

The method can be considered as an alternative to the χ^2-test for independence in a contingency table. The χ^2-test only takes the variation caused by random sampling into account and may thus be too liberal if additional sources of variation are present. Also the χ^2-test does not take the ordering of the notes into account. The combined over-years method for ordinal characteristics takes other sources of variation into account by including a random variety-by-year interaction term (as for the COYD method described in TGP/8/1 Part II: 3).It takes the ordering of notes into account by using a cumulative function over the ordered notes. The inclusion of the random effect is expected to decrease the number of distinct pairs of varieties compared to the χ^2-test for independence, but to better ensure that the decisions are consistent over coming years. Taking the ordering of notes into account is expected to increase the power of the test and thus to increase the number of distinct pairs.

The method is based on a generalisation of the traditional analyses of variance and regression methods for normally distributed data, which are called "generalized linear mixed models". A general description of the method may be found in Agresti (2002) and a more specific description – using other examples of data may be found in Kristensen (2011).

The combined over-years method for ordinal characteristics involves
- Calculating the number of plants for each note for each variety in each of the two or three years of trials, which results in a 3-way table (see the example)
- Analyse the data using appropriate software
- Compare each candidate to the reference varieties and the other candidates at the appropriate level of significance to see which varieties the candidate is distinct from
- Check if the variety-by-year interaction term for distinct pairs is considerably larger than the average for all variety pairs

Example

For demonstration a subset of varieties from a DUS experiment with Meadow fescue (*Festuca pratensis*) in Finland was chosen. The notes for Plant: growth habit at inflorescence emergence (Characteristic 9 of TG/39/8) in 2010, 2011 and 2012 were analysed (Table 4). In most cases 40-60 plants were recorded in each year. This characteristic is rather sensitive to the growing conditions. This is apparent from table 4 where it is seen that the note 1 was recorded only in 2012 while note 7 was recorded only in 2010. Also it is seen that the most common note (over all varieties) in the three years was note, 5, 3 and 3, respectively in 2010, 2011 and 2012. The applied analysis method takes this into account by calculating an additive effect of each year (as for the COYD method for normal distributed data).

The estimated percent of plants in each note for each variety are shown in Table 2.

Table 1. Number of individual plants with each note for each variety and year for the characteristic Plant: growth habit at inflorescence emergence in Meadow fescue *(Festuca pratensis)*

Variety	Note																				
	1			2			3			4			5			6			7		
	2010	2011	2012	2010	2011	2012	2010	2011	2012	2010	2011	2012	2010	2011	2012	2010	2011	2012	2010	2011	2012
A	0	0	2	0	2	20	4	27	23	1	23	5	32	2	8	4	0	1	0	0	0
B	0	0	0	0	1	20	1	12	21	9	5	11	29	0	5	8	0	0	0	0	0
C	0	0	0	0	4	24	3	21	21	1	21	7	30	7	6	8	1	1	0	0	0
D	0	0	2	0	6	17	7	35	23	6	11	14	31	1	3	3	0	0	0	0	0
E	0	0	1	1	9	22	9	30	28	13	12	6	31	1	1	0	0	0	0	0	0
F	0	0	0	0	1	11	0	13	14	6	22	15	27	14	18	10	4	1	0	0	0
G	0	0	0	0	3	29	8	34	25	10	18	4	25	3	1	4	0	0	0	0	0
H	0	0	5	0	6	28	7	48	21	19	6	4	19	0	1	1	0	0	0	0	0
I	0	0	1	0	2	20	5	29	21	6	23	8	29	5	9	6	0	0	0	0	0
J	0	0	0	0	0	15	1	35	27	0	16	12	35	5	6	4	0	0	2	0	0
K	0	0	0	0	0	16	2	24	14	4	17	13	29	17	13	9	0	2	2	0	0
L	0	0	3	0	3	20	4	34	26	7	17	8	28	5	3	2	0	0	0	0	0
M	0	0	0	0	1	18	5	24	22	7	27	13	30	7	6	5	0	0	2	0	0
N	0	0	0	0	2	10	3	18	24	2	15	9	25	16	14	11	1	1	1	0	0
O	0	0	0	0	5	19	9	39	29	9	8	10	23	2	1	3	0	0	0	0	0
P	0	0	2	0	9	23	13	30	32	7	4	3	19	0	0	2	0	0	0	0	0
Q	0	0	1	0	4	24	9	27	24	10	19	8	28	5	2	3	0	0	0	0	0
R	0	0	0	0	3	24	2	30	26	6	21	6	35	6	1	5	0	0	0	0	0
S	0	0	1	0	5	16	6	25	27	14	19	11	26	8	4	2	0	0	0	0	0
T	0	0	0	0	6	19	3	36	24	4	5	7	18	3	7	5	0	0	0	0	0
U	0	0	2	0	7	17	11	41	31	15	11	8	30	0	0	0	0	0	0	0	0
V	0	0	3	0	15	32	11	33	18	13	6	5	30	3	0	4	0	1	0	0	0
W	0	0	0	0	7	22	4	28	30	6	16	6	37	5	2	6	0	0	1	0	0
X	0	0	1	0	5	19	2	24	17	4	17	15	40	6	7	2	0	0	0	0	0
Y	0	0	1	0	3	12	2	8	24	4	6	5	24	0	13	6	0	0	0	0	0
Z	0	0	0	0	1	14	1	25	17	2	16	15	26	10	13	10	0	0	0	0	0
1	0	0	2	0	6	24	5	38	24	8	9	8	34	2	2	0	0	0	0	0	0
2	0	0	0	1	4	20	5	29	26	5	16	11	37	5	3	3	0	0	0	0	0
3	0	0	2	0	10	24	7	28	27	8	12	4	30	1	0	0	0	0	0	0	0
4	0	0	1	0	9	17	7	31	28	6	10	9	30	2	2	2	0	0	0	0	0
5	0	0	0	0	3	14	1	24	26	9	22	16	36	8	4	5	0	0	0	0	0

Table 2. Estimated percent of plants for each note of each variety

Variety	Note						
	1	2	3	4	5	6	7
A	0.2	5.7	34.8	33.7	24.5	1.1	0.1
B	0.2	5.9	35.4	33.5	23.9	1.0	0.0
C	0.1	4.8	31.2	34.4	28.1	1.3	0.1
D	0.2	8.2	41.8	30.8	18.2	0.7	0.0
E	0.4	12.4	48.7	25.7	12.4	0.5	0.0
F	0.0	1.7	14.6	28.9	51.0	3.6	0.2
G	0.3	10.3	45.8	28.2	14.9	0.6	0.0
H	0.6	17.0	52.3	20.9	8.9	0.3	0.0
I	0.2	5.6	34.1	33.9	25.1	1.1	0.1
J	0.1	4.3	29.2	34.6	30.3	1.4	0.1
K	0.1	2.5	19.6	32.5	42.8	2.5	0.1
L	0.2	7.8	40.8	31.4	19.1	0.8	0.0
M	0.1	4.6	30.2	34.5	29.1	1.3	0.1
N	0.1	2.2	18.1	31.6	45.1	2.8	0.1
O	0.3	10.1	45.5	28.4	15.1	0.6	0.0
P	0.5	16.0	51.8	21.8	9.5	0.3	0.0
Q	0.3	8.8	43.1	30.0	17.1	0.7	0.0
R	0.2	6.7	37.8	32.7	21.7	0.9	0.0
S	0.2	7.0	38.8	32.3	20.8	0.8	0.0
T	0.2	7.9	41.0	31.2	18.8	0.7	0.0
U	0.4	12.1	48.4	25.9	12.7	0.5	0.0
V	0.5	16.5	52.1	21.4	9.2	0.3	0.0
W	0.2	7.1	38.9	32.2	20.7	0.8	0.0
X	0.1	5.2	32.6	34.2	26.6	1.2	0.1
Y	0.1	4.4	29.7	34.6	29.7	1.4	0.1
Z	0.1	2.7	21.3	33.3	40.3	2.2	0.1
1	0.3	10.6	46.2	27.8	14.5	0.5	0.0
2	0.2	6.7	37.8	32.7	21.7	0.9	0.0
3	0.4	12.6	49.0	25.4	12.2	0.4	0.0
4	0.3	9.3	44.1	29.4	16.3	0.6	0.0
5	0.1	4.4	29.7	34.6	29.7	1.4	0.1

The candidates were variety *A* and *B* and the remaining varieties *C, D,…, 5* were reference varieties, a measure of the differences and the P-values for testing the hypothesis of no difference between candidate and reference varieties were calculated. The differences and the *P*-values are shown in Table 6. An F_3-value is calculated in a similar way as for COY-D for normally distributed characteristics and is used in order to ensure that the pair did not became distinct because of a very large difference in only of the years without being different in other years (TGP/8/1 Draft 13 Section 3.6.3). Therefore, a significant difference between two varieties with a high F_3-value should be examined carefully before the final decision is taken. The F_3-values and their significances are also shown in Table 6.

For the data shown here candidate *A* could be separated from 11 of the reference varieties when using a 1% level of significance while candidate B could be separated form 10 of the reference varieties. The two candidates could not be separated from each other. The largest F_3-*value,* 5.43, was found for variety pair *B-S* (the approximate threshold for the F_4 values to be significant is 4.98). This means that the interaction for this pair should have been considered if this pair had been distinct on this characteristic.

Table 3. Differences and F$_3$ values together with P-values for relevant pairs of varieties

Variety	Candidate A				Candidate B			
	Difference	P$_{Difference}$	F$_3$	P$_{F3}$	Difference	P$_{Difference}$	F$_3$	P$_{F3}$
A	-	-	-	-	0.03	0.9011	0.22	0.4051
B	-0.03	0.9011	0.21	0.6566	-	-	-	-
C	0.19	0.4507	0.02	0.8782	0.22	0.4051	0.09	0.7694
D	-0.39	0.1243	0.04	0.8522	-0.35	0.1856	0.07	0.7947
E	-0.84	0.0011	0.73	0.4154	-0.81	0.0030	1.73	0.2215
F	1.26	<.0001	0.56	0.4743	1.29	<.0001	1.46	0.2584
G	-0.63	0.0125	1.66	0.2298	-0.60	0.0255	3.06	0.1144
H	-1.22	<.0001	1.17	0.3080	-1.19	<.0001	2.37	0.1579
I	0.03	0.8922	0.29	0.6041	0.07	0.8004	0.99	0.3448
J	0.30	0.2267	1.13	0.3146	0.34	0.2081	0.37	0.5600
K	0.88	0.0007	0.00	0.9669	0.91	0.0010	0.25	0.6274
L	-0.33	0.1879	0.52	0.4895	-0.30	0.2651	1.39	0.2681
M	0.24	0.3255	0.82	0.3878	0.28	0.2949	1.87	0.2047
N	0.99	0.0002	0.00	0.9734	1.02	0.0003	0.18	0.6805
O	-0.61	0.0162	0.27	0.6151	-0.58	0.0317	0.96	0.3525
P	-1.15	<.0001	0.24	0.6350	-1.11	0.0001	0.90	0.3664
Q	-0.47	0.0630	2.59	0.1421	-0.43	0.1039	4.28	0.0685
R	-0.17	0.5056	0.06	0.8115	-0.13	0.6174	0.50	0.4984
S	-0.22	0.3813	3.50	0.0943	-0.18	0.4858	5.43	0.0448
T	-0.34	0.1848	0.82	0.3879	-0.31	0.2578	0.20	0.6650
U	-0.82	0.0013	1.04	0.3352	-0.79	0.0035	2.18	0.1735
V	-1.18	<.0001	0.03	0.8674	-1.15	<.0001	0.08	0.7799
W	-0.23	0.3621	0.17	0.6870	-0.19	0.4653	0.00	0.9662
X	0.12	0.6441	0.00	0.9863	0.15	0.5764	0.23	0.6444
Y	0.27	0.3246	0.19	0.6753	0.30	0.2936	0.00	0.9791
Z	0.77	0.0032	0.64	0.4435	0.80	0.0038	0.12	0.7404
1	-0.66	0.0093	0.00	0.9861	-0.63	0.0196	0.23	0.6443
2	-0.17	0.5049	0.15	0.7116	-0.13	0.6165	0.71	0.4219
3	-0.87	0.0009	0.07	0.8017	-0.83	0.0026	0.52	0.4907
4	-0.53	0.0393	0.03	0.8714	-0.49	0.0684	0.09	0.7760
5	0.27	0.2712	0.31	0.5938	0.31	0.2471	1.03	0.3376

In order to examine whether one or more varieties have a different variety by year interaction than the main part of the varieties, the actual contribution to the interaction was calculated for each variety and compared to the average contribution from all varieties. This was done using an F- value, F_4.

The F_4 values for each variety in the analysis are shown in Figure 2. The largest F_4-value, 2.78, was found for variety S (the approximate threshold for the F_4-values to be significant is 4.98). This value was not significantly larger than 1. The F_4-value is calculated as the quotients between the each varieties contribution to the overall interaction and the average interaction over all varieties. As the contribution for the actual variety enters in both the numerator and denominator of the F_4-value this test is approximate.

It is also seen that some varieties, e.g. *I, K, N, X, 1, 2, 3* and *5* have a very low interaction with year indicating that their response to year is very close to the mean reaction for all varieties.
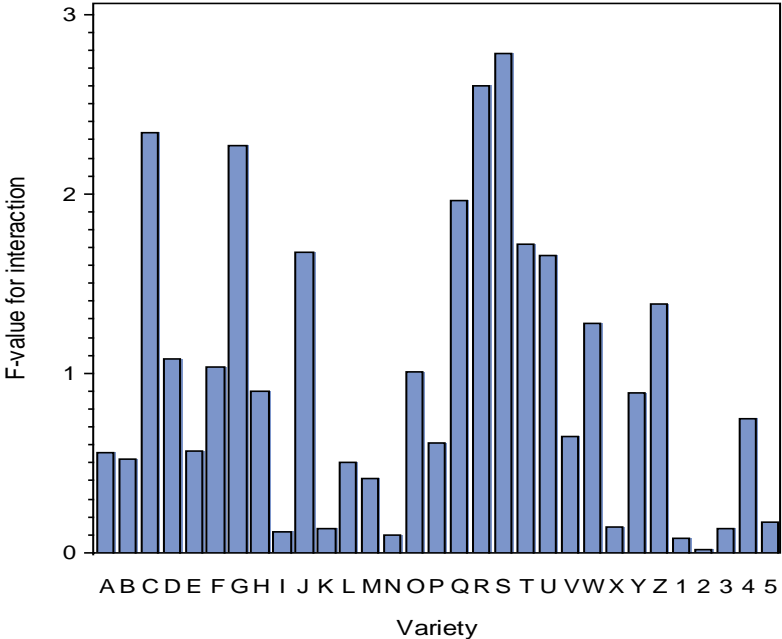
Figure 1. F_4-values for each variety's contribution to the interaction for ordinal characteristic growth habit

II. NOMINAL CHARACTERISTICS

Summary of requirements for application of the method

The method is appropriate to use for assessing distinctness of varieties where:
- The characteristic is nominal and recorded for individual plants (usually recorded visually)
- There are some differences between plants
- The observations are made over at least two years or growing cycles on a single location
- There should be at least 20 degrees of freedom for estimating the random variety-by-year interaction term.
- The expected number of plants for each combination of variety and note should be at least one – and for most of the combinations the number should be at least 5.

Summary

The method can be considered as an alternative to the χ^2-test for independence in a contingency table. The χ^2-test only takes the variation caused by random sampling into account and may thus be too liberal if additional sources of variation are present. The combined over-years method for nominal characteristics takes other sources of variation into account by including a random variety-by-year interaction term (as for the COYD method described in TGP/8/1 Part II: 3). The inclusion of the random effect is expected to decrease the number of distinct pairs of varieties compared to the χ^2-test for independence, but to better ensure that the decisions are consistent over coming years. The method is based on a generalisation of the traditional analyses of variance and regression methods for normally distributed data, which are called "generalized linear mixed models". A detailed description of the method – using other examples of data may be found in Agresti (2002) or Kristensen (2011).

The combined over-years method for nominal characteristics involves
- Calculating the number of plants for each note for each variety in each of the two or three years of trials, which results in a 3-way table (see the example)
- Analyse the data using appropriate software
- Compare each candidate to the reference varieties and the other candidates at the appropriate level of significance to see which varieties the candidate is distinct from
- Check if the variety-by-year interaction term for distinct pairs is considerably larger than the average for all variety pairs

Example

No example shown at present.

III.   BINOMIAL CHARACTERISTICS

Summary of requirements for application of the method

The method is appropriate to use for assessing distinctness of varieties where:
- The characteristic is recorded for individual plants (usually recorded visually) using a scale with only 2 levels (such as present/absent or similar)
- There are some differences between plants
- The observations are made over at least two years or growing cycles on a single location
- There should be at least 20 degrees of freedom for estimating the random variety-by-year interaction term.
- The expected number of plants for each combination of variety and note should be at least one – and for most of the combinations the number should be at least 5.

Summary

The method can be considered as an alternative to the χ^2-test for independence in a contingency table. The χ^2-test only takes the variation caused by random sampling into account and may thus be too liberal if additional sources of variation are present. The combined over-years method for binomial characteristics take other sources of variation into account by including a random variety-by-year interaction term (as for the COYD method described in TGP/8/1 Part II: 3). The inclusion of the random effect is expected to decrease the number of distinct pairs of varieties compared to the χ^2-test for independence, but to better ensure that the decisions are consistent over coming years.

The method is based on generalisation of the traditional analyses of variance and regression methods for normally distributed data, which are called "generalized linear mixed models".

The combined over-years method for binomial characteristics involves
- Calculating the number of plants for each note for each variety in each of the two or three years of trials, which results in a 3-way table
- Analyse the data using appropriate software
- Compare each candidate to the reference varieties and the other candidates at the appropriate level of significance to see which varieties the candidate is distinct from
- Check if the variety-by-year interaction term for distinct pairs is considerably larger than the average for all variety pairs

Example

The proportion of plants with cyanid glucoside (Characteristic 4 in TG/38/7) was measured for some white clover varieties in Northern Ireland in each of 3 years. The variable was recorded as absent or present. In this example only 20 varieties are used and variety 1 and 2 are considered as candidates, while the remaining varieties are considered as references. The data are shown in Table 7.

Table 4. Number of plants without and with cyanid glucoside in 20 white clover varieties in each of 3 years

Variety	Year 1		Year 2		Year 3	
	Absent	Present	Absent	Present	Absent	Present
1	31	29	22	38	17	43
2	40	20	42	18	41	19
3	50	10	52	8	55	5
4	42	18	40	20	34	26
5	37	23	42	18	37	23
6	51	9	49	11	52	8
7	30	30	25	35	26	34
8	37	23	31	29	30	30
9	27	33	27	33	25	35
10	48	12	47	13	43	17
11	40	20	40	20	32	28
12	18	42	13	47	12	48
13	10	50	12	48	5	55
14	41	19	46	14	45	15
15	58	2	55	5	58	2
16	7	53	10	50	11	49
17	25	35	22	38	20	40
18	48	12	54	6	52	8
19	20	40	20	40	23	37
20	57	3	54	6	55	5

The analysis showed that for these data there was no interaction between variety and year, which means that the variance component for year by variety was estimated to be zero and thus all variation in the data could be explained by sampling variation. The F-test for comparing the varieties was 36.67 with a P-value less than 0.01%, so there were clearly some differences among the varieties.

More specifically the analysis showed that candidate variety 1 was significantly different from 12 of the reference varieties at the 1% level (Table 8) whereas candidate variety 2 was significantly different from 11 of the reference varieties. Also the two candidate varieties were significantly different at the 1% level (Table 8).

As there was no interaction between variety and year, all F_3 and F_4 values are estimated to be zero for these data. Therefore, they are not shown here.

Table 5. Estimated percent of plants with cyanid glucoside for each variety and comparison of each variety with the candidate varieties 1 and 2 using F-tests

Variety	Estimated percent	Candidate 1		Candidate 2	
		F	P	F	P
1	61.1			30.45	<.0001
2	31.6	30.45	<.0001		
3	12.7	77.01	<.0001	17.58	0.0002
4	35.5	23.05	<.0001	0.61	0.4395
5	35.5	23.05	<.0001	0.61	0.4395
6	15.5	70.09	<.0001	12.54	0.0011
7	55.0	1.38	0.2473	19.58	<.0001
8	45.5	8.69	0.0054	7.27	0.0104
9	56.1	0.93	0.3414	21.39	<.0001
10	23.3	49.59	<.0001	3.12	0.0853
11	37.8	19.27	<.0001	1.48	0.2309
12	76.1	9.28	0.0042	66.21	<.0001
13	85.0	24.61	<.0001	90.68	<.0001
14	26.6	41.43	<.0001	1.09	0.3034
15	5.0	82.34	<.0001	33.21	<.0001
16	84.5	23.44	<.0001	89.25	<.0001
17	62.8	0.11	0.7463	33.81	<.0001
18	14.4	72.95	<.0001	14.45	0.0005
19	65.0	0.58	0.4492	38.53	<.0001
20	7.8	84.99	<.0001	28.18	<.0001

IV.   COMMON TO ALL THREE METHODS

Software

The procedure *GLIMMIX* of *SAS* (SAS Institute Inc., 2010) can be used to estimate the parameters of the generalised linear mixed model, and the data-step facilities (and/or the procedure *IML*) of the same package can be used for the remaining calculations. However, similar facilities may be found in other statistical packages, thus the *glmer*() function of the package *lme4* of R can do the binomial analysis provided that there are more than one observation for each combination of variety and year.

Final note

In the case where there are only two notes, the methods for nominal and ordinal scaled characteristics both become identical as they reduce to the same binomial method: meaning that both methods can be applied to binomially distributed data.

References and literature

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