**THE FEDERAL UNIVERSITY, KASHERE**

 **FACULTY OF AGRICULTURE**

 **DEPARTMENT OF ANIMAL SCIENCE**

ANS: 5209

Course Code: ANS 5209

Course Title: Animal experimentation and Research Techniques

No of unit: Two

Course Duration: Two hours

Status: Compulsory

Prerequisite: Nil

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**Course description**

This course is very important for successful Research in animal experimentation. This stems from the fact that Good understanding and judicious use of knowledge acquired from this course would

lead to yield good result.

**GRADING SYSTEM FOR THE COURSE**

This course will be graded as follows:

Class Attendance In form of random quizzes 10%

Assignments 10%

Test(s) 20%

Final Examination 60%

**TOTAL 100%**

***Attendance:*** It is expected that every student will be in class for lectures and also participate in all practical exercises. Attendance records will be in the form of random quizzes to determine each person’s qualification to sit for the final examination. In case of illness or other unavoidable cause of absence, the student must communicate as soon as possible with the instructors, indicating the reason for the absence.

***Academic Integrity:*** Violations of academic integrity, including dishonesty in assignments, Examinations or other academic performances are prohibited. You are not allowed to make Copies of another person’s work and submit it as your own; that is plagiarism. All cases of Academic dishonesty will be reported to the University Management for appropriate sanctions in Accordance with the guidelines for handling students’ misconduct as spelt out in the Students’ Handbook.

***Assignments and Group Work:*** Students are expected to submit assignments as scheduled. Failure to submit an assignment by certain student as at when due will earn such student zero for that assignment. Only under extenuating circumstances, for which a student has notified the instructor in advance, will late submission of assignments be permitted.

***Code of Conduct in Lecture Rooms:*** Students should turn off their cell phones during lectures. Students are prohibited from engaging in other activities (such as texting, watching videos, *etc*.) during lectures.

**Principle Statistics**

**Introduction**

Statistics: Mean qualitative figures for example, the number of bull born in a year, the number of schools and college in a state, number of unemployed youths in the country, the rainfall over a period of time, production of calves in a country, etc are referred as statistics. The numerical figures are termed the as **data,** data form only an aspect of the subject of statistics.

Statistics: Is the body of scientific principles and techniques, it also refers to constants, such as the mean and standard deviation, calculated from sample observations. Depending on the first two meanings, a number of definitions have been given for statistics. Hence

Statistics can be defined as the body of concepts, principle and method dealing with collection, summarization, analysis and interpretation of data.

Statistic is used in research for efficient planning of experiments, and for interpreting of experimental data.

The process of data collection may involve field trails, laboratory experiments and sample surveys. There are Two branches of statistics:-

1. **Sampling Design:** Deals with data collection, the data are recorded without interfering the process that is being observed.
2. **Experimental Design:** The subjects constituting the population are put into controlled environment, the salient feature and significance of the collected data must then be assessed and interpreted through careful analysis, for this purpose the data must be organized and summarized.

**Descriptive Statistics:** The branch of statistic dealing with summarization of the data e.g Average (Mean, mode, median) and measures of dispersion (standard deviation, variance).

**Inferential Statistics:** The branch of statistics dealing with interpretation of data e.g T-test, F-test, Chi-square-test etc.

**Some Basic Concepts**

* **Sample and Population:** In statistical sense, a population is a set of measurements or count of a single variable taken on all the individuals specified to be in a population. The population may be relatively small, the seed production per hectare of the *Sorghum almum* field in a specified area in a specific years, or large for example, the height of all bull over one year of age in Nigeria, or the yield that would result from all possible plots of a given shape that could be arranged on an experimental area.
* **A Sample** is a set of measurements, which constitute part of a population or rather proportion of the population, is knows as sample. We obtain information and make inferences about population from a sample. For this reason it is important that the sample be representative of the population. To obtain a representative sample we use the principle of randomness. A random sample is one in which any individual measurement is as likely to be individual as any other.

**Variable and Attributes**

Variability is a common characteristic in animal science, a quantitative and qualitative characteristic that varies from observation in a same group is called a variable. In case of qualitative variables, observations are made using interval scale whereas in the case of qualitative variables, observations are made using nominal scale (a name in given to each observation).

 Conventionally, the quantitative variables are termed **variables** and the qualitative variables are termed as **attributes** thus milk yield, availability of nitrogen in the soil, number of leaves or tiller per pasture plant and number of eggs laid by hen are all variables. The breed of animals, sex of animal and attributes.

**Measure of Central Tendency**

 It is the central value around which other values are concentrated. It is a representative item of a distribution e.g. mean, mode, median, harmonic mean, and geometric mean. Among all the measures of central tendency,

* **Mean:** Is the most widely used measure of central tendency in presenting average.
* **Mode:** Is used for qualitative data.
* **Median and Mode**: When frequency distributing is skewed (not symmetrical).
* **Median and Mode**: When extreme values are present in the raw data, mean median and mode can be used in case of symmetrical distribution.

**Measure of Dispersion:** It is a measure of spread, variation or scatterings of observations from the mean, means are the representative of a frequency distribution they do not tell anything about the scatterings of observation within the distribution suppose that we have the distribution of the yields (Kg/head) or (Kg/plot) of cow or pasture plant from five plots each. The distribution may be.

 Mean

Bread I 48, 42, 42, 41, 40 42

 Bread II 48, 42, 33, 57, 30 42

 It can be seen the mean yield for both breed varieties is 42kg. But we cannot say that the performance of the two breed are the same. There are greater uniformly of yields in the first breed, where as there is more variability in the yields of the second breed. The first breed may be preferred since it is more consistent in yield performance from the above example; it is obvious that central tendency alone is not sufficient to describe a frequency distribution. In addition to it we should have a measure of scatterness of observation. There are different measures of dispersion like the range, mean, deviation, standard deviation and variance the most common measure of dispersion, and the best for most purpose, is the variance or its square root, the standard deviation.

**Co-efficient of Variation:** The variability among experimental units involving different units of measurements and plot sizes can be composed by co-efficient of variability (CV). The co-efficient of variability expresses the standard deviation per experimental unit as a percent of the general mean of the experiment.

CV = S/X (100)

 For example, in a pasture Biomass experiment, the mean of all the plots in the experiment was 30.5t/ha and the standard deviation per plot was 1.18t/ha.

 LV = 1.8/30.5 x 100 = 3.9%

**Techniques and Procedures in Animal Experimentation**

**What is Research:** Research is the search for knowledge it is an integral part of science which is all about gathering and classifying information, discovery of new ways and ideas leading to a breakthrough and the genesis of a new scientific theory. Gathering and use of information in science fallow the steps of scientific enquiring.

1. Identification of problem
2. Definition of the problem
3. Analysis of the problem
4. Deduction based on the analysis including recommendation of solution to the problem
5. Implementation of the recommended solution. The last step is not required in theoretical research when implemental are not generally necessary.

Research is therefore any organised enquiring that aim at providing information for solving identified problems.

**Types of Research**

1. **Applied research:** explain a situation in order to enable the researcher understand it better. Such research can be regarded as an explanatory research it concern itself mainly with the question why? Applied research tries to discover why certain events, situation or phenomenon occur e.g. a herd of cattle that have been performing well with regards to milk production suddenly drops. A research may be carried out to find out how a phenomenon occurs. This explains the mechanism of the situation, in order to answer the questions of what, why and how therefore, both basis and applied research are necessary using different skills.
2. **Survey Research:** Survey research focuses on the populations of specific areas or even the universe as a whole. Data are collected from the population for intensive study and analysis. Carefully selected samples are used to represent the entire population. This is in order to minimize the use of resources and save time. The manner of selection of a representative sample enables the researcher to generalize or localize his findings for population. Random sampling have been used in order to generalize findings for the entire population from which the sample was drawn several method have been involved in survey research and they include:-

a. **Mail Questionnaire:** This contain worded questions that will be self administered by the researcher, such questionnaires are answered in writing; therefore only people who can read and write should be the respondents. The questionnaires are usually accompanied by a covering letter of introduction and instruction and how to answer the question such method is cheap to carry out and can cover a wide area, it also enables researcher reach respondents who are otherwise in accessible. A major demerit of the mail questionnaire is the rate of response is always low some respondents may be reluctant to settle down and answer the question because they simply don’t have the time.

b. **Personal Interview:** It is a survey method involving the person needing information and that providing it (the respondent). The interviewer administers the questionnaire himself by asking the respondent question and filling the form like questionnaire himself. The research has the opportunity to ask relevant questions that may arise due to a particular response. Clarification and explanation can be gained by the researcher during the course of interview. For a successful interview, the researcher must establish a good rapport with the respondent. This is achieved by giving the respondent the feeling that the information he is giving is pleasant and satisfying, the respondent must have the impressions that the researcher is responsible, reliable, and trustworthy,the respondent must be convinced that his contribution are valuable and worth, while. Response should be recorded as quickly as possible and they can be read to the respondent for confirmation or correction. The advantage is of personal interview are that the researcher has control over the types of response he receives.

c. **Observation:** In this method, the researcher can gather information only by watching and recording data. Observation includes monitoring behavioural and non behavioural activities and conditions of samples animals. The techniques of observation in which the observer is involved in a particular activity to be observed, structured observation in which the observation is controlled or not controlled by the observer, and direct and indirect observation. In animal research, the use of structured observation is prevalent. Treatments that cause certain behaviours in animals are administrated and controlled e.g. feeding feed with different concentrate.

**Experimentation:** This type of research explore whether relationship exist among some identifiable variables and nature of relationship is .A contrived situation may occur where certain variables existing in the situation seem to be either controlled or possibly eliminated. This gives the researcher the chance of performing the experiment the way it is desired. A control is a set standard against which certain variables or treatments are compared, e.g. experiment to see the effect of fish meal on the performance of layers. the diets without the fish meal become the control.

**Type of experimentation**

**Laboratory experiment:** In this type of research, all independent variables which can influence or affect the dependent variable are controlled. This done by minimizing .their effect on the dependent variables. Such research situation are isolated from what is ordinarily normal so that there is greater examination of interplay among desired variables. sample subjects to be experimented are divided into two group: a control and experimental group in order to determine if any between the administration and non administration of treatments on the samples. The control group is the group that receive no experimental treatments. Laboratory experiment is precise and replicable, it enables the researcher to test hypothesis, verify existing theories and discoveries. it allow for in depth study and understanding of the interaction that occur between variables.

 **Field experiment:** These are laboratory experiments performed live on farm sites. Rather than keep animal in the laboratory, they are grouped statistically on the field inform for manipulations of situations for necessary effects of the treatment administered on them. Field experiments therefore operate with less control than lab-experiments and are closer to reality than lab experiment. Field experiments are not common in Animal science. It also use control and experimental groups to properly measure the effects of experimental treatments on the sample subjects which are the experimental animals it has the advantage of replication suited for testing hypotheesis and verifying theories. Other are:-

* Field **studies** or on farm studies.
* Expost-facto research (after, the fact).

Assignment writes on these two type experiment mention above

**Sampling Techniques**

 A sampling is a part of the population; sampling is the procedure for drawing samples that will exhibit a phenomenon that is of interest to the researcher. Population may be finite that is countable or infinite that is uncountable. In both cases to reduce the cumbersomeness of research, representatives of the population are selected for study.

 The reason for sampling in research studies includes:-

1. There are similarities among elements that make up a population therefore the study of a few will provide enough knowledge to cover for the entire population.
2. It may be practically impossible to take a comprehensive study of the population because of the nature and pattern of distribution and dispersion.
3. It is cheaper to study a sample than the entire population.
4. It enables the researcher to be more thorough with better supervision.
5. The study of a sample enables us to get quicker result.

A good sample should bear certain characteristic such as being true representative of the entire population this enables the researcher to make accurate estimate regarding population. Good representativeness also relates to precision which ensures that random fluctuation or error variance or sampling error is minimal. It ensures that there is absence of systematic variance or sampling bias.

**Sampling Method**

There are two major method of sampling:-

1. Probability method
2. Non-probability method
3. **Probability Method:** In the probability method of sampling every subject is given the chance of being chosen at random. Equal and independent change of being included is the sample. The methods of probability selection include:-
	1. Random sampling
	2. Systematic sampling
	3. Stratified sampling
	4. Area sampling

1. **Random Sampling:** This is the most fundamental method of probability sampling. Its principle is randomness and is applied in probability sampling methods; it uses the principle or randomization, which is a procedure of giving every subject in a population an equal chance it appearing in a selection. This could be done by writing number of subjects on pieces of paper so that a paper is picked, shuffled and picked again until the required number is met. A table of random numbers could also be used. This however could only be used in a finite population where all subject or items can be listed serially. It eliminates bias.

2. **Systematic:** It involves selecting the nth subject form serially listed population of subjects. N is determined by dividing the population by the required sample size e.g. in a population of 7000 subject, 1000 needs to be selected. Then we say N = 7000/1000 = 7 we then start selecting from any number, the seventh subject so that is started from 7th then we have 14th,21st 28th, 35th, 42th, 49th, 56th, etc.

3. **Stratified Sampling:** This is an applied random sampling method in which the population is group into some definite characteristics called strata. Each stratum is then applied random selection of the subject. The number of subjects to be selected must be proportional to the stratum share of total population. It is a more superior method because it uses certain characteristic to group the population before applying random techniques.

4. **Area or Cluster Sampling:** This method is used mainly in geographically distributed populations. The cluster in which people with particular characteristic live are identified and proportionate selection of the subjects is made from the clusters.

**Non-Probability Sampling:** does not guarantee randomness, which means that the elements of population or subjects do not have the privilege of having equal chance or knowa Probability of being selected in the sampling process, if randomness occur, it does so by chance example of non-probability sampling method include:-

* Accidental or convenience sampling
* Multi-stage sampling
* Panel sampling
* Double sampling

Assignment write short note on the above mention non probability sampling

**Experiment Design:**

**Introduction:** The design of an experiment is more important than analysis. There are (3) basic principle in conducting experiment which must be adhere for many data to be generated

1. **Randomization:** This is basic principles of fundamental design and is defined as unbiased allocation of experimental material on plot. Randomization is achieved through the use of random number or table of random permutation.

**Reason for Randomization**

* It is essential to avoid any element of biasness.
* It is essential for valid estimate of experimental error.
1. **Replication:** This is defining as repetition as set treatments, none of which may appear more than one in the block or replication; replication provide an estimate of experimental error against which significant of treatment are tested.
2. **Local Control:** This is the controlling of variance or variability locality by effect in such a way. There is minimum different within the block and maximum different between the block.

**Basic Technology and Concept**

1. **Experimental Unit:** is the smallest unit receiving a certain treatment, the information or data for comparison are from such single units. Example a plot, pot, a single animal or group of animal receiving the feed from the same source, a small plot having the same type of pasture. An experimental unit could be a single pasture plant or a leaf to which a treatment is imposed.
2. **Treatment:** Is the material being forced on the subject (unit) and whose effect is to be monitored. The treatment can be either qualitative (e.g. pasture species, feed, fertilizer types) or quantitative (e.g. time, period, amount of fertilizer, amount of feed).
3. **Experimental Error:** is a measure of the sum of variation between plots or units receiving same treatments (variations in the treatment are treated alike). Inherent variability in the subject, uncontrolled external influences, and lack of uniformly in the application of treatments are possible causes of experiment error. Experimental error should be controlled so that we can estimate the treatment effects properly and compare effects of various treatments effectively.

**What is Experimental Design?**

Experimental Design of an experiment is a plan used in experimentation taking into consideration the nature and number of treatments and the experimental materials. Essentially, it refers to the rules regulating the assignment of treatments to the experimental units (plot, animals, a single plant, a leaf etc.). The purpose of experimental design is to provide a maximum amount if information relevant to the problem in the most efficient way. The choice of design is influenced by several considerations, notably the objectives, the amount of resources and the term available. In all cases, however, the emphasis is on the reduction of unknown error and the elimination of systematic bias.

Principle of Experimental Design:

 A proper excremental design must include or rather follow the three principles.

1. Replication
2. Randomisation
3. Local control (blocking)
4. **Replication:** When treatments appear more than once in an experiment, it is said to be replicated (repetition of treatments). It provides an estimate of the experiment error and increases precision of the experiment.
5. **Randomisation:** Every treatment should have an equal chance of being assigned to any experimental unit be it unfavourable or favourable. It allows elimination of bias and ensures a valid measurement of experimental error.
6. **Local Control** (Blocking): Blocking refers to the assignment of group of plots or treatments to a block of land with relatively homogenous soil. This principle allows certain restrictions on randomisation to reduce experimental error. For example blocking in a heterogeneous soil condition, the randomisation of treatments is now restricted to block only. Blocking is another way of improving the estimate of the error term, but if the blocking is justified.

**Types of Experimental Design**

Experimental Designs: Single factor experiments, knowledge of experimental design is necessary for selection of simple designs that give control of variability and enable the researcher to attain the required precision. The three factors that are important in selecting an experimental design are:-

* Types and number of treatments to be tested.
* Degree of precision
* Magnitude of heterogeneity (soil, environment for example) in the experimental material.

The experimental designs commonly used for single factor experimental are classified as complete block designs

1. Complete randomised design (CRD)
2. Randomised complete block design
3. Latin square design (LS)

In a complete block design, each block contains all the treatments while in an incomplete block design not all treatments may be present. The complete block design is suited for small number of treatment in large.

**Complete Block Designs**

1. **Completely Randomised Design (CRD):** A design which is based on the principles of replication and randomisation. In this design, treatments are assigned completely at random so that each experimental unit has the same chance of receiving any one treatment, for CRD any difference among experimental unit receiving the same treatments is considered as experiment with homogenous experimental units, such as laboratory and greenhouse experiments, such as effects are relatively easy to control (see layout below). For field experiments, where there is generally large variation among experimental plots in such environmental factors as soil, the CRD is rarely used.

Layout of CRD with four treatments (A, B, C and D) replicated four times.

|  |  |  |  |
| --- | --- | --- | --- |
| A4 | C2 | B3 | A1 |
| B1 | B2 | A2 | D3 |
| C4 | A3 | D1 | C3 |
| C2 | D2 | C4 | D4 |

**Advantage of CRD**

1. Flexibility is allowed that is any number of treatment and replication can be used.
2. The number of replication can be vary from treatment to treatment there by allow the experimental material to be utilizes.
3. The statistical analysis is simple.
4. The relative losses of information due to missing data are smaller than any other design.

The statistical analysis of data from CRD is very simple in same experiments it is possible that some units are likely to be destroyed or likely to fail to respond. Even when the results are missing like this or rejected to suspected quality, the analysis remain that there can be unique number of replication for different treatments.

**Disadvantage**

1. It is less accurate than other designs.
2. The assumption homogeneity of experimental material is misleading the practice and the consequences use of unrestricted randomization in treatment allocation usually gives a wrong estimate of error variance. For this reason other design are usually capable in estimating the standard error per experimental unit.
3. Outline of the ANOVA Table for CRD (using four treatment, replicate (4) four time).

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Sources of Variation** | **Degree of Freedom (df)** | **Sum of square (SS)** | **Mean square MS** | **Computed F** | **Table F** |
| **5%** | **1%** |
| Treatment  | t - 1 = 3 |  |  |  |  |  |
| Error | t(r-1) = 12 |  |  |  |  |  |
| Total | (rt-1) = 15 |  |  |  |  |  |

Note that MS= SS

 DF

 F = MS

 MSErrss

**Statistical Analysis of CRD**

1. The following table give the weight gain in gram/day of 15 birds fed on five different feed. Analysis the data to arrive it suitable conclusion about the feeds.

Feeds Replication

1 2 3 ΣX X

A 3 3 4 10 3.3

B 4 6 5 15 5.0

C 9 9 12 30 10

D 5 7 8 10 6.67

F 4 5 6 15 5.0

 Grand Total 90

n = T(R) in CRD

**Solution:**

1. CF = (GT)2  = (90)2 = 540

 n 5(3)

1. TSS = Total sum square =

Tss = Σ X21+ Σ X2 + Σ X2................... ΣXn – CF

 = 32 + 32 + 42 + 42..........................62

= 632 – 540 = 92.0

1. Trtss = (Σtrt)21 + (Σtrt)22 + ...............( Σtrt)n – CF

 r

= 102 + 152 + 302 + 202 + 152 – 540

 3

 = 76.67

1. Erss = Tss – all other = 92.0 – 76.67 = 15.33

ANOVA TABLE

Source DF SS MS F Tabular F

Trt (t-1) 4 76.67 19.17 12.50\*\* 5% 1%

Err t(r-1) 10 15.33 1.533 3.48 5.99

If computed F value is higher than tabulated F value at 5% and even at 1% hence concluded in highly significant.

LSD = t0.05 $\sqrt{2MSE}$ = 2.288 $\sqrt{2x1.533}$ = 2.25

 r 3

Rank mean in order of high

10, 6.62, 5.0, 5.0, 3.3

10 – 6.67 = 3.33

All mean carrying the same letters are statistically similar

 10 – 6.67 5.0 5.0 3.33

 3.33

 a 1.67c 1.67c

 3.24

 b

3.33c

5.0bc

10.0a

6.67b

5.0bc

Conclusion: Feed C produce more weight gain than all feed, there was no significant different between fed B;E and D and there no significant different between A,B and E. Feed D produce more weight gain this feed D. Feed C and D are superior to feed A.

Assignment

Treatment Replication

1 2 3 ΣX X

A 6 6 7 19 63.3

B 7 9 8 24 8.0

C 12 12 15 39 13

D 8 10 11 29 9.66

E 7 8 8 24 8.0

1. **Randomised Complete Block Design (RCBD):** It is one of the most commonly used experimental designs, particularly because of its flexibility and robustness. The design is especially salted for field experiments where the total number of treatments is not large and the experimental area has a predictable fertility gradient (heterogeneous experimental materials). The primary distinguishing feature of RCBD is the presence of blocks of equal size, each of which contains all the treatments (see diagram below) layout of RCBD with four treatments (A, B,C and D) replicated four times.

**Block 1 Block 2 Block 3 Block 4**

D3

A3

C3

B3

|  |
| --- |
| A1 |
| C1 |
| D1 |
| B |

|  |
| --- |
| B2 |
| A2 |
| D2 |
| C2 |

|  |
| --- |
|  A4 |
|  D4 |
|  B4 |
|  C4 |

Blocking Technique: The primary purpose of blocking is to reduce experimental error by eliminating the contribution known sources of variation among experimental units. This that variability within each block is and variability among block is maximised.

**Blocking Technique:** The primary purpose of blocking is to reduce experimental error by eliminating the contribution known sources of variation among experimental units. This that variability within each block is and variability among block is maximised. It increases in precision of the experiment due to the reduction of experimental error by adoption of local control (blocking.) Any number of replications can be included in RCBD. If large number of homogenous unit is available, large number of treatments can be included in this design. Statistical analysis is simple and easy.

Outline of ANOVA table for RCBD (using four (4) treatments replicated four (4) times).

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Sources of Variation** | **Degree of Freedom (DF)** | **Sum of square (SS)** | **Mean square MS** | **Computed F** | **Table F** |
| **5%** | **1%** |
| Block (rep) | r-1=3 |  |  |  |  |  |
| Treatment  | t-1=3 |  |  |  |  |  |
| Error | (t-1) (r-1)= 9 |  |  |  |  |  |
| Total  | rt-1 = 15 |  |  |  |  |  |

Example: An experiment was concluded to determine the effect of different feed on weight gain ability of 15 birds which are of different group, one to three and 10 weeks old.

Treatment I II III Σtrt X

A 4 4 5 13 4.23

B 5 7 6 18 6.0

C 7 10 10 27 9.0

D 5 8 7 20 6.67

E 4 6 7 17 5.67

Σ block 25 35 35 95 = GT

X 5 7 7

Solution

1). CF = (GT)2 = (95)2 = 601.67

 n 15

2.) Total sum square = Tss = Σtr21+tr22+............+tr2 – CF

 42+42+52+52+72+................72 – 60.67

 = 655 – 601.67 = 53.33

3.) Block sum square = Σblc21+ Σblc2+ Σblc3 – CF

 t

 = 252+352+352 -601.67 = 13.32

 5

4.) trtss = ΣtrtA2+ ΣtrtB+................... ΣtrtD – CF

 Block

 = 132+182+272+202+172 – 601.67

3

= 35.33

5.) Σrss = Total ss – all other

 = 53.33 – (13.33+35.33)

 = 53.33 – 48.66 = 4.67

ANOVA TABLE

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Sources**  | **Degree of Freedom (DF)** | **Sum of square (SS)** | **Mean square MS** | **Computed F** | **Table F** |
| **5%** | **1%** |
| Block (b-1) | 2 | 13.33 | 6.67 | 11.50 | 4.46 | 8.65 |
| Trt (t-1) | 4 | 35.33 | 8.83 | 15.13 | 3.84 | 7.01 |
|  Err (t-1)(b-1) | 8 | 4.67 | 0.58 |  |  |  |
| Total bt-1 | 14 |  |  |  |  |  |

Ms = SS

 DF

F = MS

 MSErrss

1. **Latin square Design (LS):** The randomised complete block design is useful for eliminating the contribution of one source of variation only. In contrast, the major feature of Latin square design, its capacity to simultaneously handle two known sources of variation among experimental units. Thus, in this design blocking is in two directions, the two directional blocking in is design, commonly referred to as row-block and column-blocking is accomplished by ensuring that every treatment occurs only once in each column block. This procedure makes it possible to estimate variation among row- blocks as well as among column-blocks and to remove them from experimental error. In this design number of treatment is always equal to number of columns and number of rows. Therefore, the layout is always a perfect square for example in assessing the moisture content of pasture plant, the rows may be individuals plants and the column may be leaf size like small, medium and large, in animal experiment, the age of animal may form rows, and weight of the animal may form column. Example of Layout of Latin square showing blocking in two direction handling variation due to age and weight of animal.

 Treatments are A,B,C and C.

 Column 1 to 4

 Weight of Animal

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| ROW 1 | A | C | B | D |
| ROW 2 | C | A | D | B |
| ROW 3 | B | D | A | C |
| ROW 4 | D | B | C | A |

Age of

 Animal

Outline of the ANOVA table for Latin Square (using 4 treatments)

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Sources of Variation**  | **Degree of Freedom (DF)** | **Sum of square (SS)** | **Mean square MS** | **Computed F** | **Table F** |
| **5%** | **1%** |
| Row | t-1 =3 |  |  |  |  |  |
| Column | t-1 =3 |  |  |  |  |  |
| Treatment  | t-1 =3 |  |  |  |  |  |
| Error | (t-1) (t-2) = 6 |  |  |  |  |  |
| Total | t2 -1 = 15 |  |  |  |  |  |

**Advantage of LSD**

* The Design control variation in two direction
* The error mean square is small that make the design very efficient
* The analysis of data is simple
* Analysis of data remain even in the case of missing plot techniques

**Disadvantage of LSD**

* The number of treatment is associated with number of row and Colum
* The design is more efficient when treatment number ranging from 5-12, if the treatment number is less than (5) five, error degree of freedom is too small to put any confidence in the result. On the other hand. If treatment numbers exceed 12 experimental size becomes too big to handle.

**Comparison Between three designs**

1. Experimental materials in LSD vary in two directions and in CRD experimental material are homogenous.
2. Number of treatments equal to number of row in LSD.
3. Number of block or replication is not associated with number of treatment, but in CRD are unrestricted randomisations.
4. Analysis of data is not flexible, but in CRD analysis of data is flexible.

**Example:** an experiment was carried out to investigate the effect of winter feeding on milk production. Four diets were fed to four cows over four periods of three weeks.

 Cow

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Period  | 1 | 2 | 3 | 4 | Total Row  |
| 1 | A:192 | B:195 | C:292 | D:249 | 928821761 |
| 2 | B:190 | D:203 | A:218 | C:210 |
| 3 | C:214 | A:139 | D:245 | B:163 |
| 4 | D:221 | C:152 | D:204 | A:139 | 7113221 Grand Total |
| Total column | 817 | 689 | 959 | 756 |

Treatment total A = 683, B = 752, C = 866, D = 918

Correction factor = (ΣX)2  = (3221)2  = 698427.56

 N 13

Un corrected sum of square = (ΣX2) = 674315

1. Total sum of square = ΣX2 – (ΣX)2  = 674315 – 648427.5

 N

 Total DF = 16 – 1 = 15 = 25887.44

1. Treatment SS = Σ$^{T2}/\_{n}$ - (ΣX)2

 N

 = (683)2 + (752)2 + (868)2 + (918)2 – CF

 4 4 4 4

 = 657935.25 – 648427.65 = 8607.687

 Treatment DF = 4 – 1 = 3

1. Row SS = Σ(R2) – CF

 N

 = 9282 + 8212 + 7612 + 7112 – CF

 4 4 4 4

 = 654966.75 – 648427.56 = 6539.19

 Row DF = 4 – 1 = 3

1. Colum SS = Σ(C2) – CF

 N

 = 8172 + 6892 + 9592 + 7562 – 6484427.56

 4 4 4 4

 = 658356.75 – 648427.56 = 9929.19

Residual SS = Total SS – (Trtss + Row ss + Colum ss)

 = 25887.44 – (8607.687 + 6539.19 + 9929.19)

 = 811.373

Residual DF = Total DF – (TrtDF + Row DF + Colum DF)

 = 15 – (3+3+3) = 6

ANOVA

Source DF SS MS Fcal Tabulated F Treatment 3 8607.687 2869.23 21.2187 DF=3, 6 =4.76 at 0.05

Row 3 6539.19 2179.73 16.12 DF=3, 6 =9.78 at 0.01

Column 3 9929.19 3309.73 24.47

Error or Residual 6 811.373 135.23

Total 15 25889.44

All our calculated F values are higher than the F Tabulated at 1% therefore we compare treatment mean at 1% level of significance. Row and column are ignored unless we want to know which row or columns are significantly different.

LSD = $t $x SED

 SED = $\sqrt{2 }$s2

 n

 = 2 x 135.23 = $\sqrt{270.46 }$ $\sqrt{67.615 }$ = 8.22

 4 4

 t = 3.707

 LSD = t x SED

 3.707 x 8.22 = 30.47

A = 170.75, B = 188, C= 217, D = 229.5.

Comparison Difference LSD Significance

AVs B 12.25 30.47 No

AVs C 46.25 30.47 Yes

AVs D 58.75 30.47 Yes

BVs C 20.00 30.47 No

BVs D 41.50 30.47 Yes

CVs D 12.50 30.47 No

A B C D

170.75c 188bc 217ab 229.5a

**Test of Significant:** There are three (3) types of test of significant. Z-test, t-test and f-test, plus chi-square.

1. **Z-test:** is used where population is large and the mean and standard deviation is known.

Z = X – U

 δ

 $\sqrt{n }$

 Using Z-test, you can text whether sample is belonging to a particular group or not.

 Example: In Addi integrated farm Kumo considerable experience has shown the breakage strength of egg is 75kg and standard deviation (Sd) is 1.2kg, a sample of egg gave a mean strength breakage of 65kg. Test whether the sample is representation in Addi farm or not

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 70 | 73 | 60 | 55 | 65 | 51 | 74 | 62 | 72 |

 Sample

 Breakage Strength

Solution:

* Identify the population mean U, Standard deviation = δ
* Identify the sample of mean X,
* Remember when the different is not significant it mean they are not belong to the same group and when it is significant a true representation or highly significant it mean they are from different group, you should also remember that the Z variance are absolute that, they don’t carry a sign that is they are – ve or + ve.

Solution

 X = 65

U = 75

Sample no. = 9 Z = 65-75

Sd = 1.2kg 1.2

 $\sqrt{9 }$

 Z = -10 = - 25

 1.2

 3

Z = Value at 5% = 1.96

 at 1% = 2.58

**conclusion:** as the calculate Z-value is 25 and is greater than table Z-value a 5% (1.96) and at 1% (2.58) therefore the is different between calculate and table Z-value, hence it concluded as the calculate Z-value is higher than table Z-value at 5% (1.96) and at 1% (2.55) hence there is significant difference, therefore it is a true representation of the farm.

Example 2

Considerable experiment has show that normal pulse rate of a Bull is 73 which and sd of 2.7, the pulse rate recorded for nine calve of animal science department farm of Federal University, Kashere are.

Calve: 1 2 3 4 5 6 7 8 9

Pulse rate: 68 74 73 70 71 67 71 70 75

 X = 71 Z = X - U

 U = 72 б = 71 – 72 = -1 = -1

Sd = 2.7 $\sqrt{n }$ 2.7 2.7 0.9

n = 9 $\sqrt{9 }$ 3

 = 1.11

 Z – Table value at 5% = 1.96

 at 1% = 2.58

**Conclusion:** Since the calculated Z-value is less than the tabulated value at 5% and 1% hence concluded that there is no significant different hence they not below to same group.

**T – Test:** This test of significant is use only when there are two treatments in an experimental. In the first place t-test unlike Z-test is used only when samples and not all population are studied, t-test is an approach involving the difference of means, and there are two type of t-test.

1. Paired t-test (equal number of observation)
2. Independent t-test

In the paired t-test there is one common inking, common factor between the treatment of experiment material while in the independent t-test there is no common linking factor.

tp = XA – XB

 SE XA = Mean A

pi = XA - XB  XB = Mean B

pooled SE SE = Standard Error

Paired t-test how to get standard error.

SE $\sqrt{ss}$

 n(n-1)

SS = CSS – CF

 б

 = Σ X2 – (ΣX) 2 or SE = $\sqrt{n }$

б = $\sqrt{Σ X2 –\left(ΣX\right)}$

 n-1

Question:

 Six animals were fed two different feed, feed, 1 and feed 2 after 8 weeks, the following weight in animal were recorded. Analysis this data statistically and recommended the feed to be used.

* Feed is the common linking factor between the animal.

Animal 1 2 3 4 5 6 ΣX

Feed (1) 7 8 7 9 8 9 48

Feed (2) 6 6 7 8 8 7 42

 d = 1 2 0 1 0 2 6= Σd

Deviation = d = (f1 – f2)

 Σ Feed1 = 48

 Σ Feed2 = 42

 X1 = ΣX1 = X1= 48 = 8

 Σd 6

 X2 = ΣX2 = X2 = 46 = 7

 Σd 6

Deviation: d = 1, 2, 0, 1, 0, 2 = 6 = Σd

 d2 = 1 4 0 1 0 4 = 10 = Σd2

SS = Css – CF = SE= $\sqrt{SS }$

 = Σd2 – (d)2 n(n-1)

 n

 = 10 – (6)2 = 4 SE = $\sqrt{4 }$ = $\sqrt{4 }$ = 0.365

 6 6(6-1) 30

 = t= XA - XB = 8 – 7 = 2.74

SE 0.365

t – Value calculated = 2.74\*

t – Value at 5% = 2.571, at 1% = 4.032.

Hence the calculated t value is higher than the table t value it is concluded there is significant different between the feed on weight gain between the animals. Hence feed one is superior to feed two hence it recommended to farmer.

Example (2) An animal scientist recorded the following weight gain (kg) from his cattle that were fed 2 difference type of feed analysis the data statistically and drawn suitable conclusion.

Animal 1 2 3 4 5 6 7 8 9 10 ΣX X

Feed A 4 8 5 2 9 3 3 6 7 2 49 4.9

Feed B 9 9 7 8 10 4 8 6 7 9 72 7.7

Deviation-5, -1, -2, -6, -1, -1, -5, 0, 0, -7,

d = FA - FB

d2 = 28, 1, 4, 36, 1, 1, 25, 0, 0, 49

SS = CSS – CF = Σd2 – (d)2

 n

CF = (25)2 = 784 = 78.4

 10 10

SS = 142 – 78.4 = 63.6

 SE = $\sqrt{ss } $ = $\sqrt{63.6}$

 n(n-1) 90

 = SE = 0.84

tp = XA - XB = 4.9 – 7.2 = -3.33

 SE 0.84

 Degree of freedom = 10 – 1 = 9

 Table value at 5% 1%

 -2.202 -3.25

 As the calculated value (- 3.33) is high than the table t-value at 9df at 5% level of probability (-2.263) and even at 1% level of probability (-3.25). Therefore there is different between feed A and B. So it is recommended to the farmer to fed feed B because is superior.

Assignment

 Eight animals (8) of the same age and breed they feed two type of feed after about 8 weeks, the animal record the following weight gain. Analyses the data and draw suitable conclusion.

Sheep 1 2 3 4 5 6 7 8

Feed A 20 20 18 17 9 8 10 7

Feed B 18 17 14 11 10 7 8 6

**Independent t-test:** This is used if there equal number of observation between two treatments or when the two treatments were test differently.

How to get pooled SE: tI = XA - XB

pooled SE

 Pooled SE = $\sqrt{SE }$21 + SE22

Example:

 Twelve animals were fed two different feed X1 and X2 after 4 weeks, the following weight (kg) were recorded. Analysis the data statistically and draw suitable conclusion which type of feed will recommended to the farmer.

 Weight Weight

Animal A Feed X1 X12 Animal B Feed X2 X22

1 7 49 7 6 36

2 8 64 8 6 36

3 7 49 9 7 49

4 9 81 10 8 64

5 8 64 11 8 64

6 9 388 12 7 298

 Σ = 48 42

X1 = 8 X2 7

 SE 1 = $\sqrt{ss1}$

 n1(n-1)

SE 1 = CSS – CF = ΣX2 = (ΣX2)2

 n

 388 – (48)2

 6

 388 – 384 = 4

 SE1 =$\sqrt{4}$ = 0.365

 30

 SE2 =$ \sqrt{SS}$1

 n(n-1)

SS2 = $\sqrt{SS}$1

 n(n-1)

SS2 = CSS2 – CF2 = ΣX2 – (ΣX)2

 n

= 298 – (C12)2

 6

= 298 – 294 = 4 = 0.368

 6 6

S E2 = 0.365

Pooled SE =Pooled SE = $\sqrt{SE }$21 + SE22

 = $\sqrt{0.365 }$2+0.3652

$ $ = $\sqrt{0.1331+0.1331} $

= Pooled SE = 0.516

 tI = X1 – X2

 pooled SE

tI = 8-7 = 1.74

 0.516

df = (n1-1) + (n2-1) =

 5 + 5 = 10

At table 5% 1%

 2.288 3.16

Conclusion: As the calculated value (1.74) is less than the table value at 10df at 5% level of probability (2.288). Hence there is no significant difference. The feed contributed same to weight gain.

**t1 Un equal number of series or un equal number of treatment**

Example: The following weight gain was recorded after feeding high protein and low protein to ten chicken. Analysis the data statistically to determine which feed give higher weight gain.

 High protein High Protein low protein low protein

Chicks 1 Weight gain Square Chick 2 Weight gain square

1 12 144 5 11 121

2 12 144 6 11 121

3 13 169 7 12 144

4 14 169 8 13 169

 51 653 9 13 169

 10 12 144 LP = low production 72 868

HP= High protein

ΣHP = 51 ΣLP = 72

Mean HP = 51 = 12.75 mean LP = 22 = 12

 4 6

ΣHP2 = 653 ΣLP2 = 868

= SEHP = $\sqrt{SS}$HP

 nHP (nHP-1)

SSHP = CSSHP - CFHP

 = CSSHP – CFHP

= ΣX2 – (ΣX)2

 n

 = 653 – (S1)2

 4

SSHP = 653 – 650.25 = 2.75

SΣHP = $\sqrt{2.75}$

 4(4-1) = 0.479

= SEHP = $\sqrt{SS}$LP

 nLP (nLP-1)

 SSLP = CSSLP – CFLP = ΣX2 = (LP2) – (ΣX)2= (LP)2

 n

 = 868 – (72)2 = 4

 6

 = 868 – 864 = 4

 SELP = $\sqrt{\begin{array}{c}4\\ 6(5)\end{array}}$ = 0.365

Pooled SE = $\sqrt{SE }$HP +SELP

 $=\sqrt{0.479 }$2+ 0.3652

 = 0.602

tI  = XA - XB = 12.75 – 12.0 = 1.246

 pooled SE 0.602

 df = (n1-1) + (n2 -1)

 df = (4-1) + (6-1)

 3 + 5 = 8

 T -value table at 8 df = 5% 1%

 2.306 3.358

Conclusion: As the calculated t-value (1.246) is less than, the t-value at 8 df 5% level probability the difference is not significant statistically, hence it concludated increase protein in the diet does not contributed to weight gain.

**T-test using paired test.**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Example: | blocksfeedX1feedX2Σb | 1 | 2 | 3 | 4 | 5 | 6 | ΣX484290= GT |
| 7 | 8 | 7 | 9 | 8 | 9 |
| 6 | 6 | 7 | 8 | 8 | 7 |
| 13 | 14 | 14 | 17 | 16 | 16 |

 Solution

Procedure calculated the following

1. CF
2. Total SS
3. Block SS
4. Treatment SS (feed)
5. Error SS

1. CF = (GT)2 = (48 + 42)2  = 90 = 675

n 6 x 2 12

2. Total SS CSS - CF

 = ΣX2 – CF

 = 72 + 82 + 72 + 92 + 82...............72 – 675

 = 686 – 678 = 11

3. Block SS (Bss) = Σ bl21 + Σ bl32 + ......... Σbl6 – CF

 Number of treatment (2)

 = 132 + 142 + 142 +.............162  - 675

 2

 = 681 – 675 = 6

4. Trtss (feed)

 = (Σ F1)2 + (Σ F2)2 – CF

 Number of block

 = 678 – 615 = 3

5. Error ss = Total – (all other ss)

 = 11 – (3+6) = 2

ANOVA TABLE

Sources DF SS MS F\*

Block SS (b-1)=5 6 1.2 3

Treatment (t-1)= 1 3 3 7.5

Error SS (b-1) (+-1) = 5 2 0.4 -

Total SS = bt – 1 = 11

Note that MS = SS , F = MS

 df MS Erss

t- value table at 5% and 1% level of probability

 Blocks Feed

5% 1% 5% 1%

5.08 10.97 6.61 16.16

Conclusion: The tabulate F value is higher than F-value calculated at 5% and 1% in block, meaning there is no significant different hence the block are statistically similar, but the feed F-value is higher than tabulated at 5% (6.61), hence, there is significant difference between feeds.

**F- test using independent.**

Example

 1 2 3 4 5 6 ΣX

Feed1 7 8 7 9 8 9 48

Feed2 6 6 7 8 8 7 42

Solution:

1) CF = (GT)2 = (48 + 42)2 = 675

 n 12

2) Total SS = CSS – CF

 = 72 + 72 + 92 +...................72 – 675

 = 686 – 675 = 11

3) TrtSS (feedss)

 ΣF12 + F22 – CF = (48)2 + (42)2 – 675

 No. of trts 6

6 78 – 675 = 3

4) Errss = Total SS – all other SS

 = 11 – 3= 8

ANOVA TABLE

Sources DF SS MS F

Feed (t-1) 1 3 3 3.75

Err SS t(t-1) 10 8 0.8

 2(6-1)

Total (tr-1) = 11

 F = MS

 MSErr

 MS = SS

 df

Table value

5% 1%

4.96 10.01

Conclusion: The F-value calculated is less than the table F-value at 5% and 1% this show there is no significant different between feed.

**Paired Plot Design**

In a field experimental where only two treatment i.e A&B and there is only one direction of variability we used paired plot design.

High low

A B A B A B

 Low High

**Assumption of Paired Plot Design**

1. A data generated from the experiment most be normally and independent distributed.
2. Variability in the experiment material is in one direction only.
3. The variance are homogeneous

Example.

 A poultry farmer tested two new poultry feed A&B for higher egg production and obtain the following number of egg per week, analyses the following data statistically and draw conclusion.

 Sample A B B A A B B A A B

 8 5 6 9 7 4 4 6 6 5

Deviation 3 - 3 3 - 2 1

d2 9 9 9 4 1

Σd = 12

Σd2 = 32

Solution

 tp = X1 – X2

 SE

 SE = $ \sqrt{SS}$ , SS = CSS - CF

 n(n-1)

1. CF = (Σd)2  = (12)2  - 144 = 28.8

 n 5 5

2. SS = CSS – CF

 CSS = Σ­d2 – CF = 32 – 28.8 = 3.2

3. SE = $\sqrt{\begin{array}{c}SS \\ \end{array}}$ , = $\sqrt{\begin{array}{c}3.2 \\S(5-1)\end{array}}$ = $\sqrt{\begin{array}{c}3.2 \\20 \end{array}}$ = 0.4

 n(n-1)

4. Mean A = 36 = 7.2

 5

 Mean B = 24 = 4.8

 5

 tp = XA – XB = 7.2 – 4.8 = 2.4= 6

 SE 0.4 0.4

 tp = 6

Assignment

Pasture agronomies tested two different forage variety and obtained the following varieties X and Y. Analysis the data statistically and draw suitable conclusion.

Y X X Y Y X X Y Y X X Y

 9 6 6 9 8 7 8 10 9 7 8 16

**Chi-square Test**

Uses

* It is used in testing significant between observed and theoretical values of events, therefore we tested weather they are normal distributed or not.
* Chi-square is use to find out whether there is linkage between two character or not, meaning that we test whether the character are independent or not.
* Chi-square is use in testing ration.

$\in $ 2 = (O – E)2

 E

O = Observed value

E = Expected value

Observed is practical value, while expected is theoretical value.

Example A Pasture Agronomies want to test weather starching and sugary seed with dark colour and light green colour segregated in the ration 9:3:3:1.

 Ration Observed Expected

Starching dark green 9 99 90

Starching light green 3 30 30

Sugary dark green 1 22 30

Sugary light green 1 9 10

 16 160 160

Expected value: Ration x GT Observed

 Total ration

 9 x 160 = 90

 16

**O – E (O – E)2 (θ – E)2**

 **E**

 9 81 0.9

 0 0 0

 -8 64 2.13

 -1 1 0.1

 **Σ = 3.13**

$\in $2 = 3.13 DF = n-1 = 4 -1 = 3

 At 5% = 7.815 at 1% = 11.341

The calculated $\in $2 – value is (3.13) is lower than the table chi-square value at 5% and 1%, hence concluded there is no significant difference between observed and expected value. The character actual segregated from 9:3:3:1.