QUALITY ISSUES IN HEMATOLOGY ANALYZERS

Dr. P.G.Subramanian
Hematopathology laboratory
Tata Memorial Hospital

Laboratory test results

Clinical diagnosis

Patient management

QUALITY

- A subjective term for which each person has his or her own definition.
- In technical usage, quality can have two meanings:
  - The characteristics of a product or service that bear on its ability to satisfy stated or implied needs
  - A product or service free of deficiencies."[5]

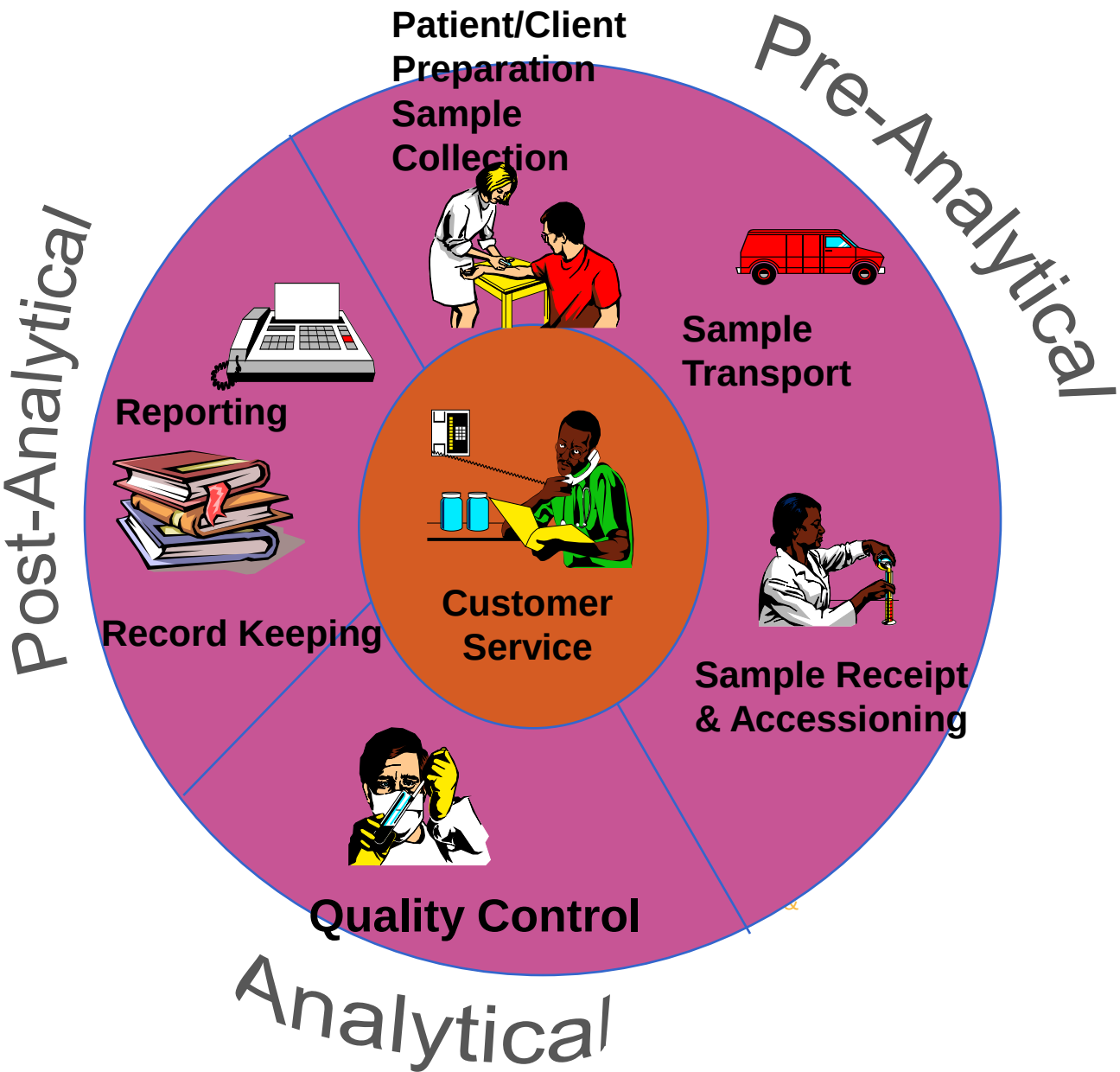
QUALITY

- Quality Assuarance
  - Prevention of defects/errors
- Quality Control
  - Detection of errors/defects

GOOD LABORATORY PRACTICE

- Quality is assured at
  - Pre-analytical phase
  - Analytical phase
  - Post analytical phase

The Quality Assurance Cycle



QUALITY MANAGEMENT AT THE PRE-ANALYTICAL PHASE

Request form shall contain … ISO15189 Cl. 5.4

XYZ Laboratory

Address ----------

Tel: ----------, Fax: ----------

Examination Request Form

Patient' name: _____

Date of Birth/Age:_____

Sex: _____ **Unique ID of patient**_____

Referring Doctor' name:_____

Address:_____

_____

Primary Sample Type: Blood/Fluid/Sputum/Stool/Microbiological Specimen/Slides/Tissue/_____ (Any other specify)

Date and Time of Primary Sample Collection: _____

Date and Time of Receipt of Primary Sample by the Laboratory: _____

Clinical History of the patient:

_____

_____

Treatment History of Patient:

_____

_____

Examinations Requested :_____

_____

_____

_____

Signature _____

BLOOD COLLECTION

- Verify the identity of the patient
- Do venipuncture in a proper manner
- Collect appropriate amount in the appropriate anticoagulant
- Proper recording of the identity of the person collecting the sample

EXAMPLE

- A serious, and potentially fatal, cause of mishap is collection from the wrong patient or subsequent specimen mix-up or transcription error. These can occur at any stage. It is essential to have a cross-check procedure.

SAMPLE TRANSPORTATION

- The samples should be transported into trays having protective covering and providing suitable environment required preventing deterioration of the sample. The person carrying the sample should be following the universal safety norms.

SAMPLE REJECTION CRITERIA

- Clotted sample
- Inadequate sample
- Hemolysed sample
- Mismatch in patient and sample identity
- Improper anticoagulant or additive use

- *Analysis of various reasons for sample rejections can generate a set of data which might reflect the highest probability of chance of pre-analytical errors due to which the samples were rejected.*

- *A feedback of this kind to the concerned ward of the hospital may enhance a positive attitude towards quality improvement at pre-analytical stage.*

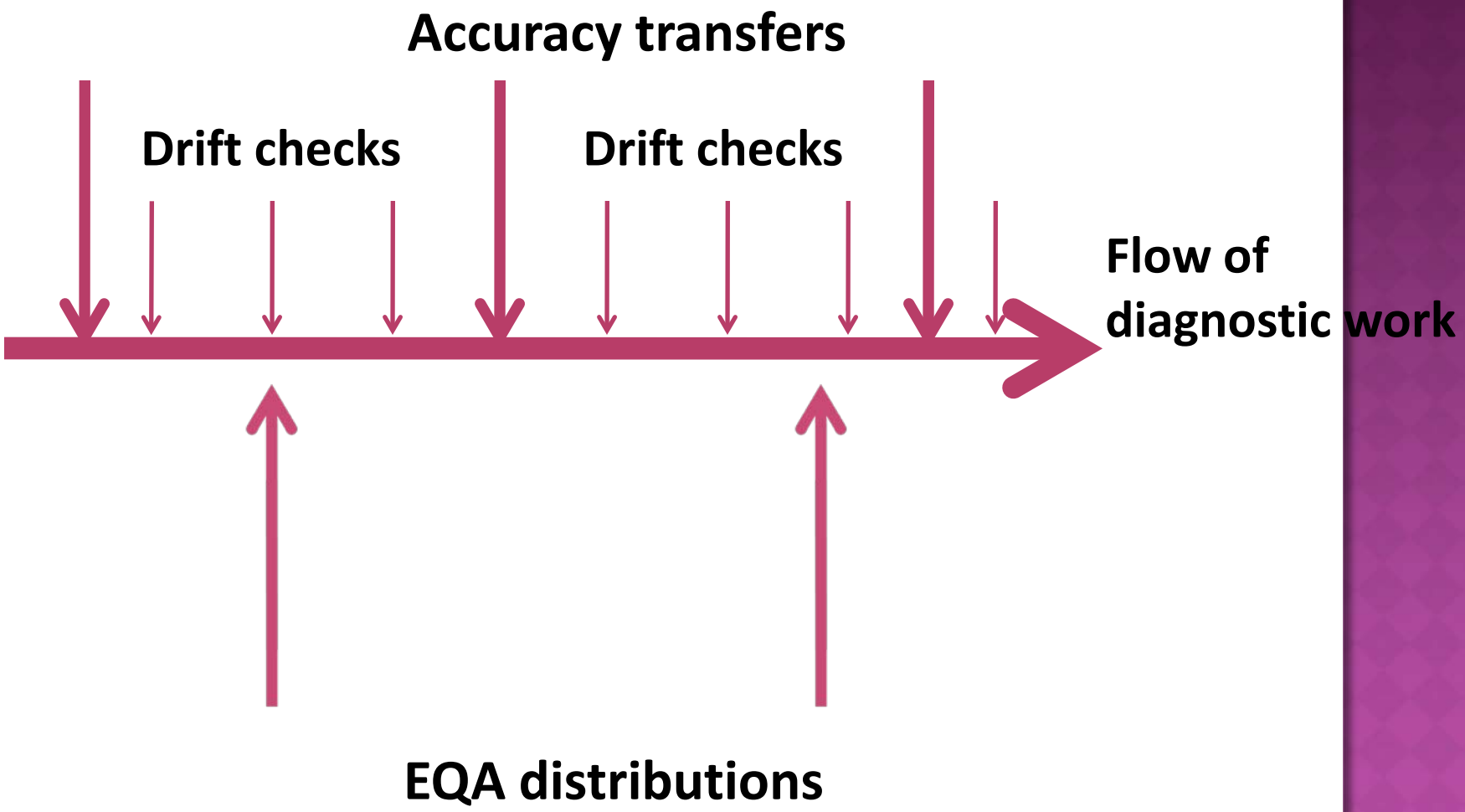
Quality Indicator – "Feedback shall be monitored on periodic basis for continual improvement"

QUALITY MANAGEMENT OF THE ANALYTIC PHASE

- Internal quality control (IQC),
- External quality assessment scheme (EQAS) and
- Standardization. [6]

(6) International Council for Standardization in Hematology

QUALITY CONTROL OF ANALYTIC PHASE



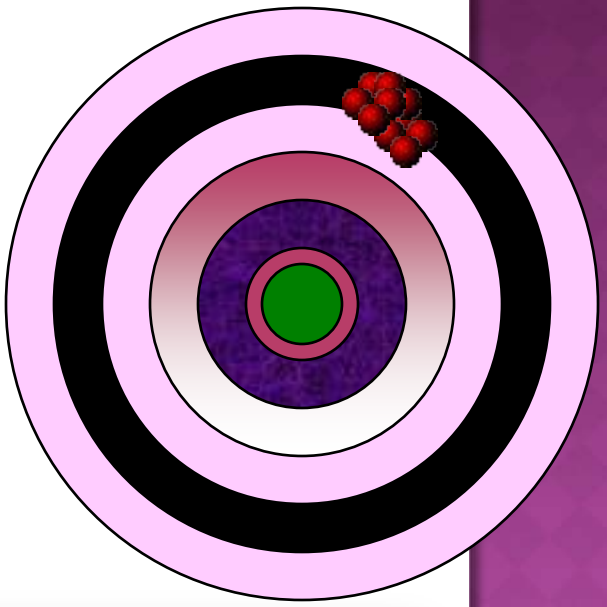
LET’S UNDERSTAND FEW IMPORTANT TERMS NOW…

PRECISION (REPRODUCIBILITY)

- **Definition**

  Precision refers to the reproducibility of a result.

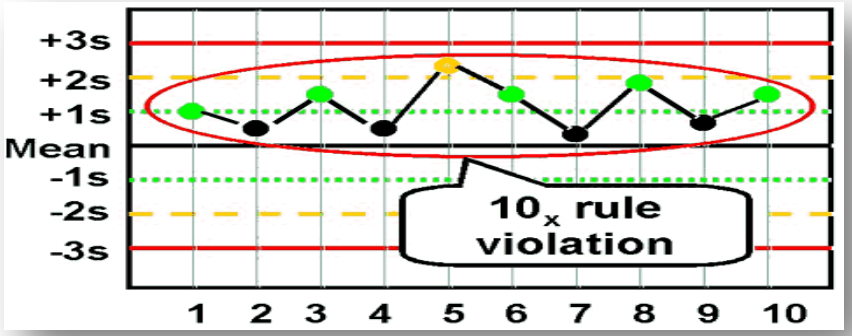
- **Comparing QC terms to a target** Figure illustrates that the results are precise (close together) but not accurate (they are not in the bull's-eye).



- **Checking precision is required while**

  -calibration

  -troubleshooting



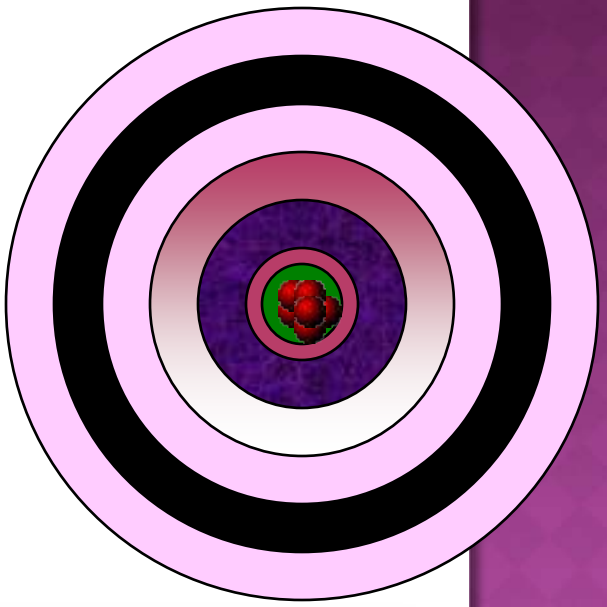
ACCURACY

- **Definition**

  Closeness of a result to the true (accepted) value.

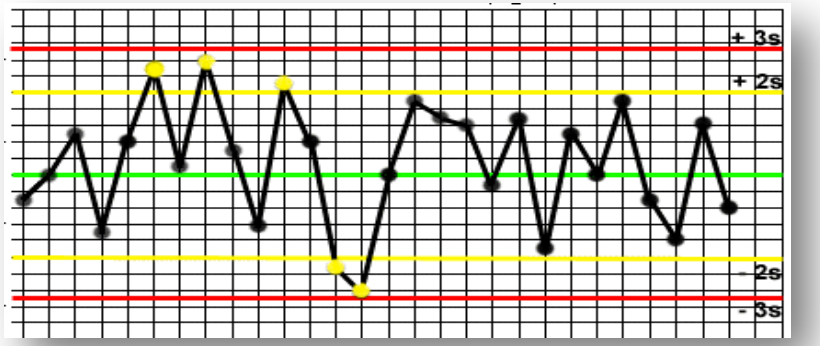
  NOTE: Before determining accuracy, first determine precision.

- **Comparing QC terms to a target** Figure illustrates that the results are accurate (in the bull's-eye) and precise (close together).



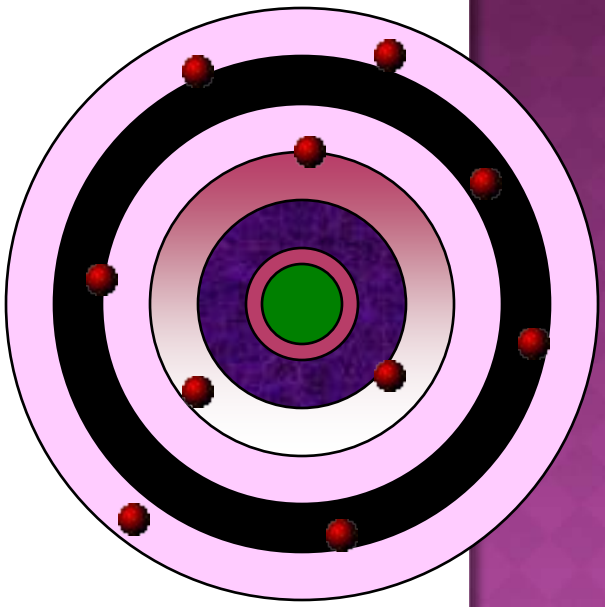
NOTE

- You cannot have accuracy without precision.
- However, you can have precision without accuracy.



NEITHER ACCURACY NOT PRECISION

- This figure illustrates that the results are neither accurate nor precise.
- None of the results are close together, and none of them are in the bull's-eye.



ACCURACY PROCEDURES AT THE TIME OF INSTALLATION

- Calibration
- Carry over check
- Linearity check

SETTING ACCURACY

Calibration

Is done to standardize the instrument for accuracy.

Calibrator

Certified Reference Material (CRM) used to calibrate a measurement on an analyzer.

Cal-Factors

If any deviation from calibration references is observed necessary calibration correction factors are applied to set the accuracy of the instrument.

CARRYOVER

- Carryover is defined as a number of cells remaining behind following the cycling of a blood sample.
- This test is performed to determine if one sample interferes with the accurate analysis of the next sample.
- Ideally, carryover Shall be very low.

Measure a specimen with a high concentration in triplicate, immediately followed by a specimen with a low concentration in triplicate.

$$\text{Carry over (\%)} = \frac{l_1 - l_3}{h_3 - l_3} \times 100$$

Where l_1 and l_3 are the results of the first and third measurements of the samples with a low concentration and h_3 is the third measurement of the sample with a high concentration.

LINEARITY CHECK

- Establishes the range for reporting of values

  if the total WBC count is 200 x 10^9/L

  If the upper limit of linearity of the analyzer is 100 x 10^9/L

Then the sample has to be diluted and re run to get accurate values of total WBC count

when diluted 1in 4 and run the analyzer will give a value of 75 x 10^9/L which might indicate that true value is 300 x 10^9/L

CONTROLS

- How many? -1, 2, 3
- Which levels? – low, normal, high
- How frequently – Daily, every 8hr, every hour
- What should we look for in a control

CONTROLS

- Atleast 2 levels
- CBC - low and normal
- Coagulation (PT and APTT) – normal and high
- Coagulation (Factor Assays) – low and normal
- Frequency – at least 8th hourly (for 24 hr service) or once a day

=

STATISTICAL APPROACH

=

TARGET VALUE

- **Mean** ($\overline{x}$) is the sum of all the measurements (Σ) divided by the number of measurements (n)

- Formula $\overline{X} = \sum x_i / n$

Where

x_i = each data point

n = the number of data points in the set

- Mean describes the “central tendency” of the data set.

- In clinical lab, the mean identifies the “target value” of a set of data points, usually QC or patient data.

DISPERSION FROM TARGET VALUE

Standard deviation (SD)

- SD quantifies the degree of dispersion of data points about the mean.
- SD is used to set limits upon which control result acceptability is determined.

Formula of standard deviation

$$S.D. = \sqrt{\frac{\sum \left(x - \bar{x} \right)^2}{n-1}}$$

$\sum$ = sum of

x = any single observed value

$\bar{x}$ = average value

n = total number of observed values

Normal Distribution Curve

or

Gaussian curve

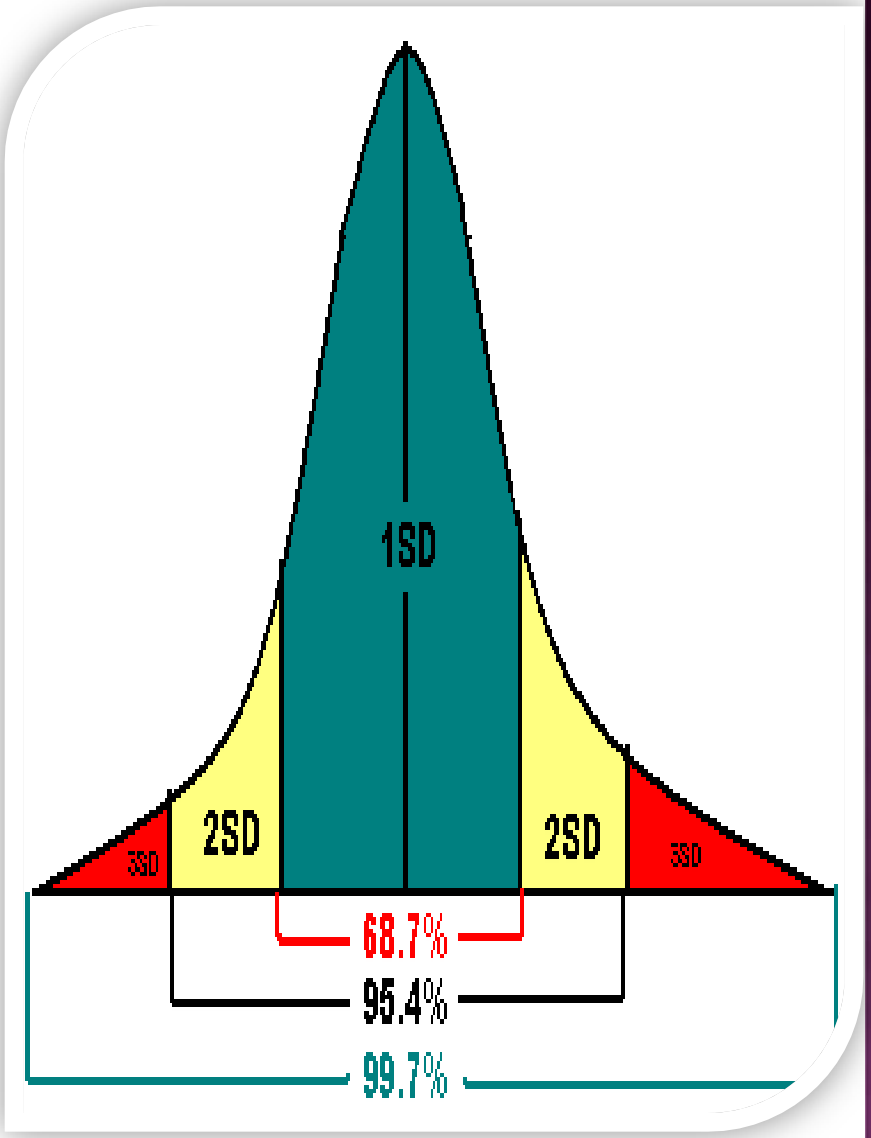
Describes events or data that occur symmetrically about the mean.

<u>Out of 100 events</u>

68.7 will fall within 1 SD

95.4 will fall within 2 SD

99.7 will fall within 3 SD



CONTROL LIMITS

How to calculate +1 SD, +2 SD, +3 SD & -1 SD, -2 SD, -3 SD

Mean + (1 x SD) = + 1SD

Mean + (2 x SD) = + 2SD

Mean + (3 x SD) = + 3SD

Mean - (1 x SD) = - 1SD

Mean - (2 x SD) = - 2SD

Mean - (3 x SD) = - 3SD

DISPERSION SIMPLIFIED

Coefficient of variation (CV)

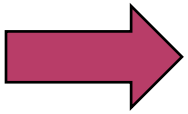
- CV is another way of indicating standard deviation, related to the actual measurement, so that variation at different levels can be compared.

- Formula $$C.V. = \frac{S.D.}{\bar{X}} \times 100$$

- It is expressed as a percentage (%CV).

LAB CAN ESTABLISH THEIR OWN 'CUT OFFS'

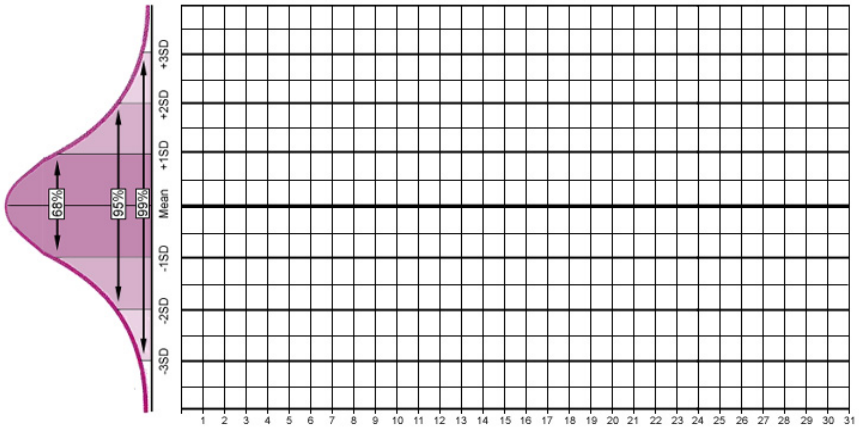
CBC parameters	Acceptable %CV	Improved %CV
WBC (White blood cell count)	4.0 %	3%
RBC (Red blood cell count)	3.0 %	2.5%
Hemoglobin	3.0 %	2.5%
MCV (Mean corpuscular cell volume)	2.0 %	1.5%
Platelet	9 %	7%

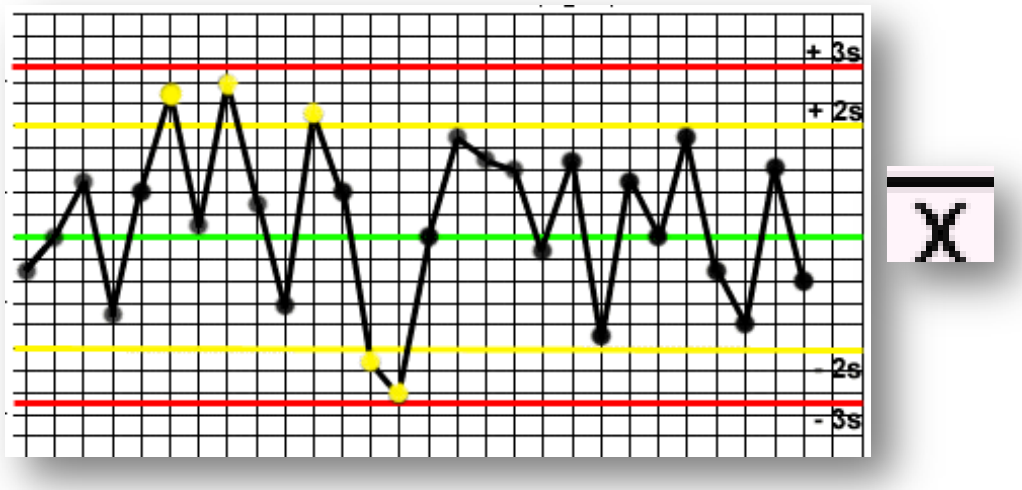


Quality Indicator – "%CV shall be monitored on periodic basis for continual improvement"

LEVEY-JENNING (L-J) CHART[7]

- Manually by using arithmetic graph paper.
- MS Excel Software in computer.
- Software like
  - MedLab QC
-
- Inbuilt QC program in the analyzers.





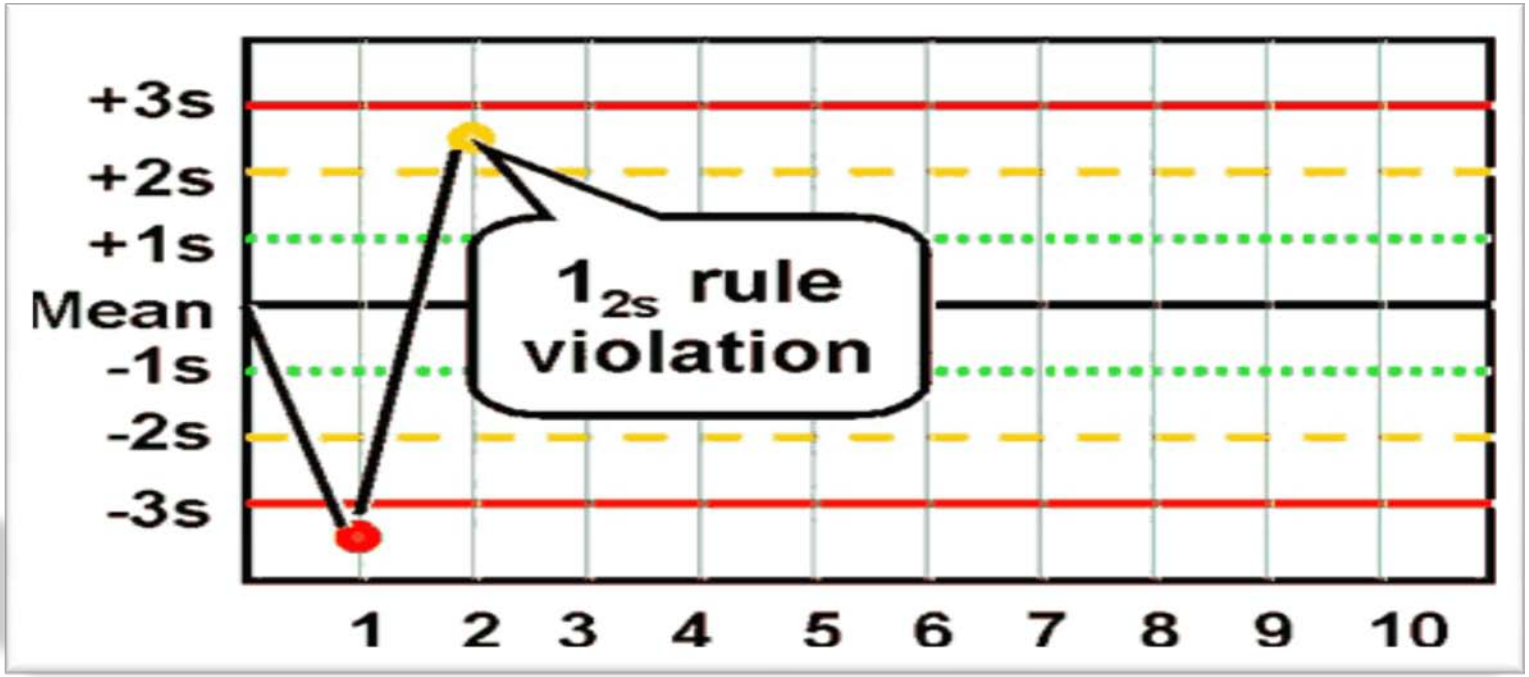
(7) Levey S, Jennings ER: The use of control charts in the clinical laboratory. *American Journal of Clinical Pathology* 1950; 20:1059-1066.

INTERPRETATION OF CONTROL CHART

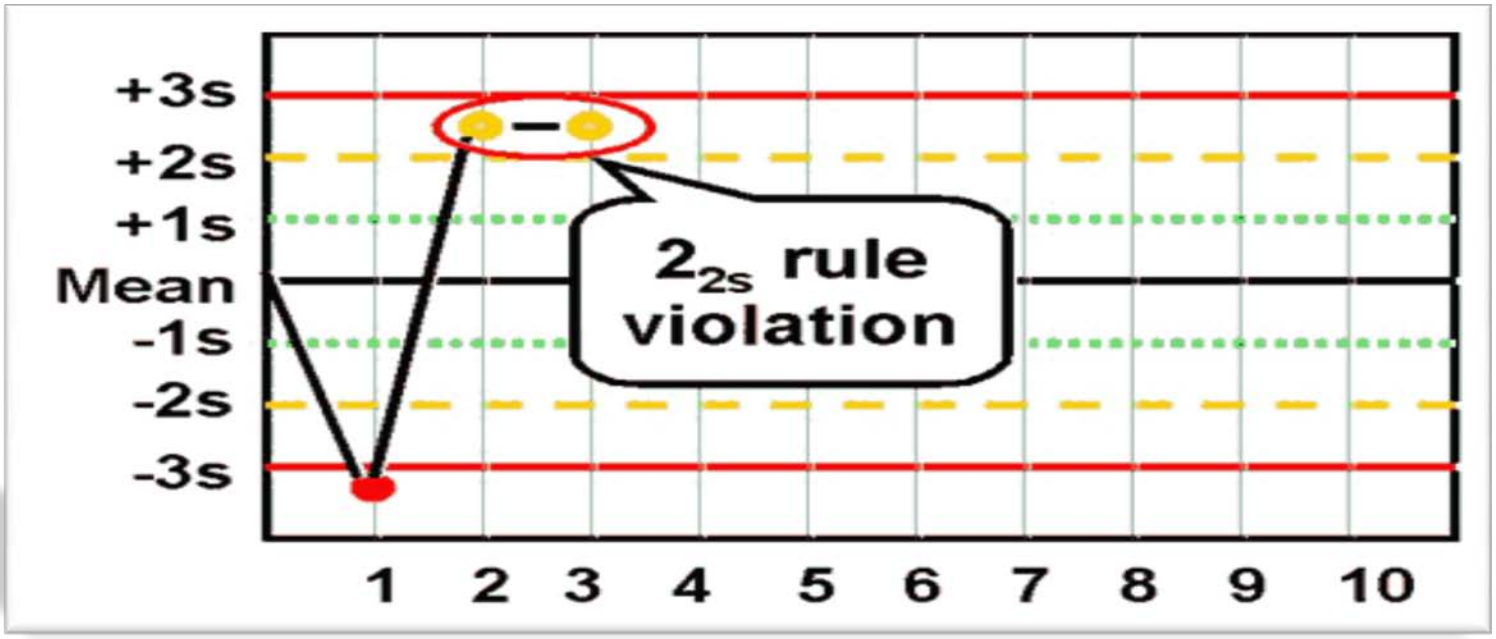
- Westgard’ s Rules
  - Control rules decide whether an analytical run is in-control or out-of-control.
  - "Westgard rules" are used when one control material is analyzed.

A run is rejected when a single control measurement exceeds the mean plus 3SD or the mean minus 3SD control limit.

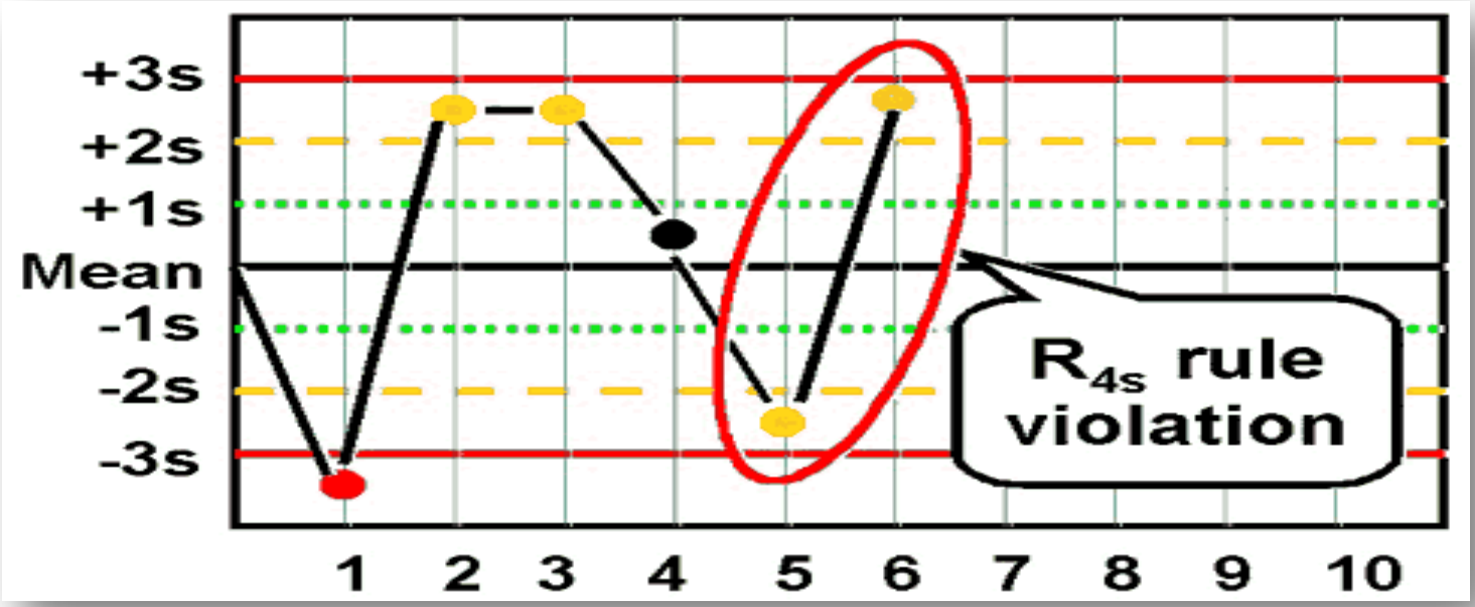
This rule is used as a warning rule to trigger careful inspection of the control data by the following rejection rules.



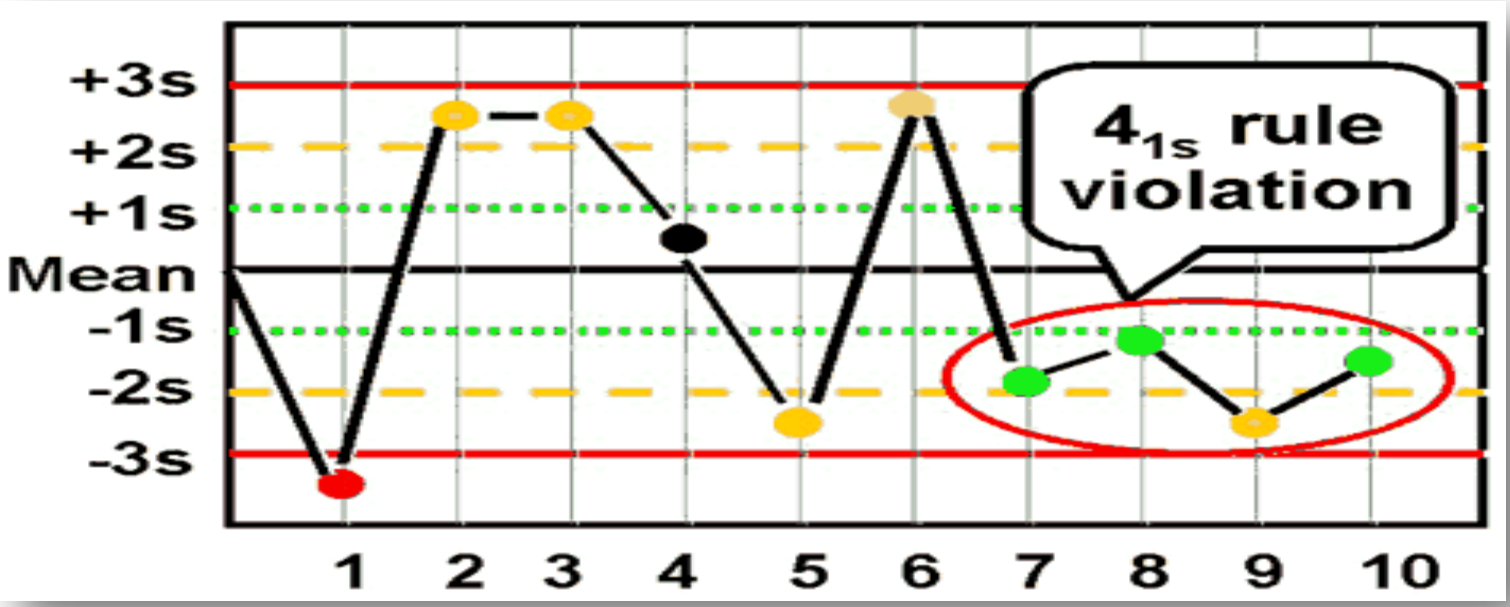
2_{2s} - reject when 2 consecutive control measurements exceed the same mean plus 2SD or the same mean minus 2SD control limit.



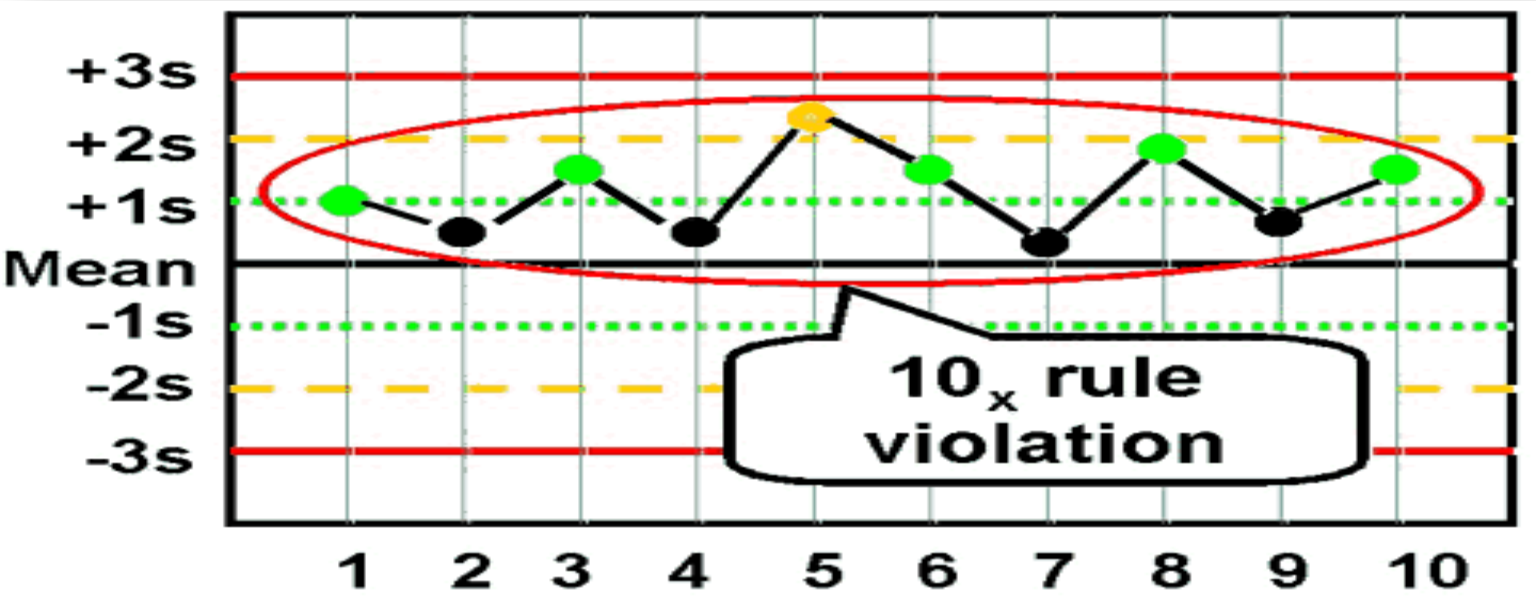
R_{4s} - reject when 1 control measurement in a group exceeds the mean plus 2SD and another exceeds the mean minus 2SD.



4_{1s} - reject when 4 consecutive control measurements exceed the same mean plus 1SD or the same mean minus 1SD control limit.



10_x - reject when 10 consecutive control measurements fall on one side of the mean.

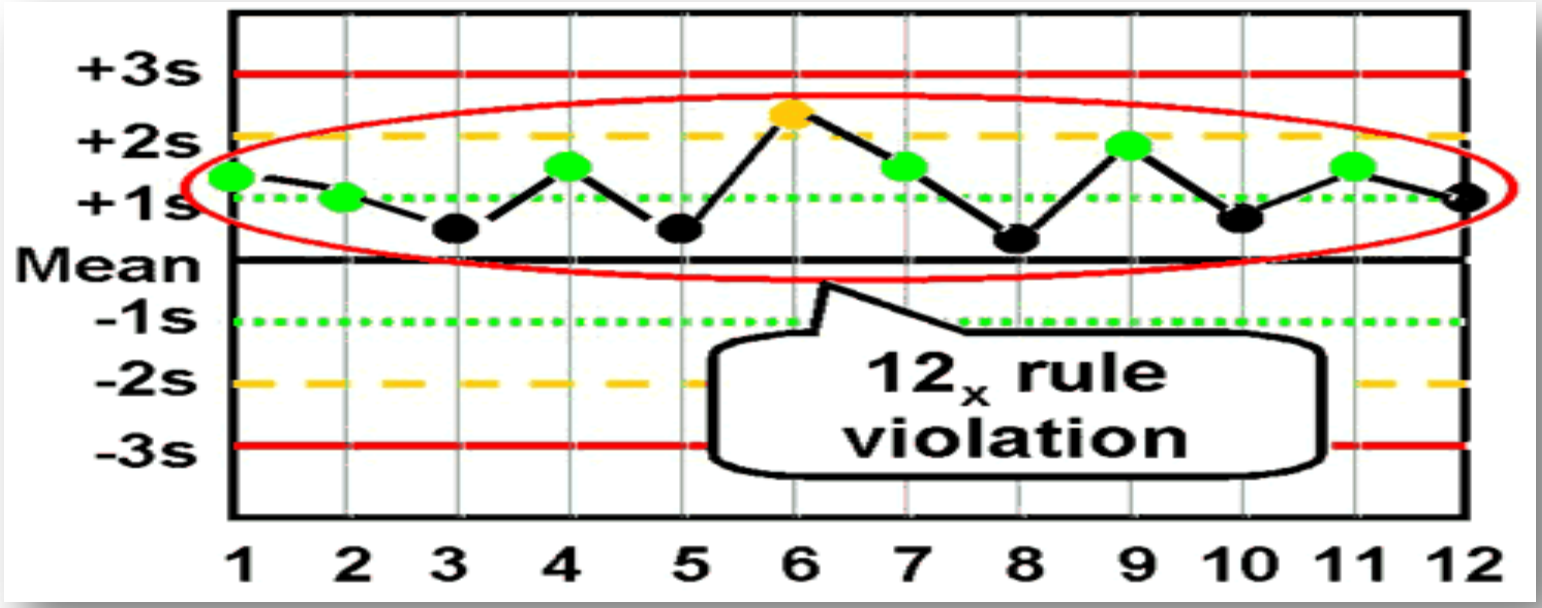


Modification of 10_x

8_x - reject when 8 consecutive control measurements fall on one side of the mean.

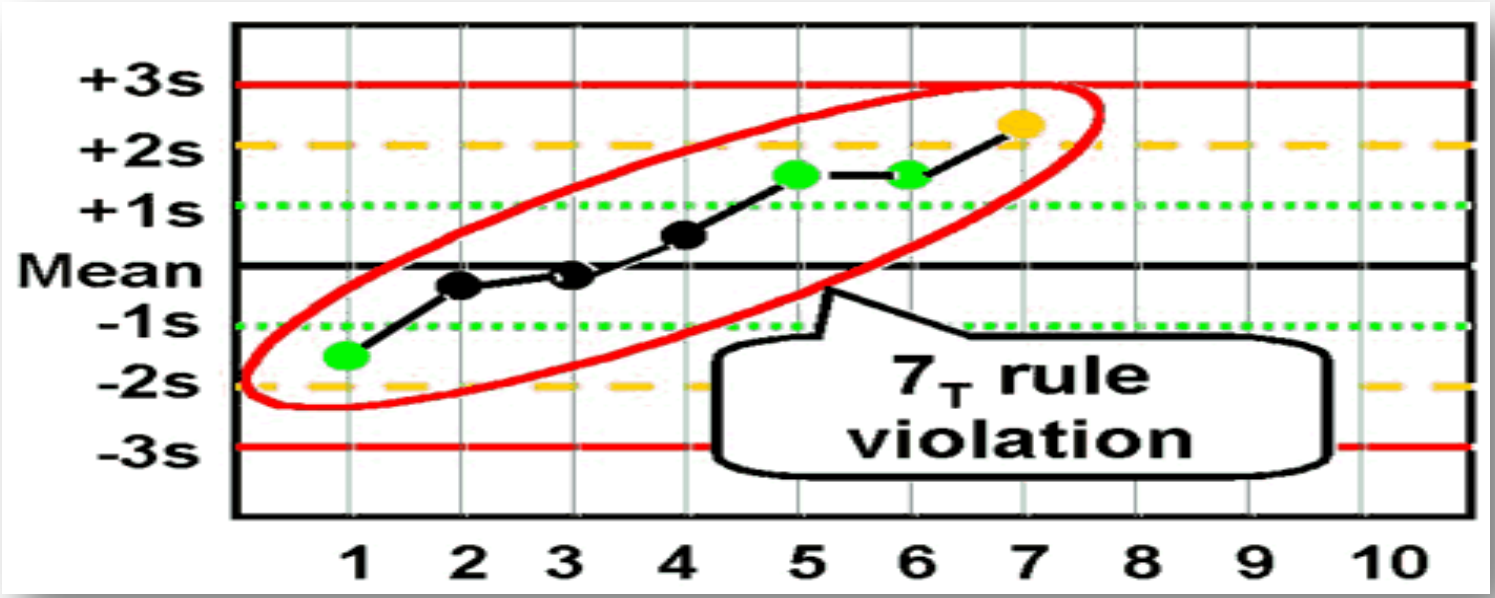
Modification of 10_x

12_x - reject when 12 consecutive control measurements fall on one side of the mean.

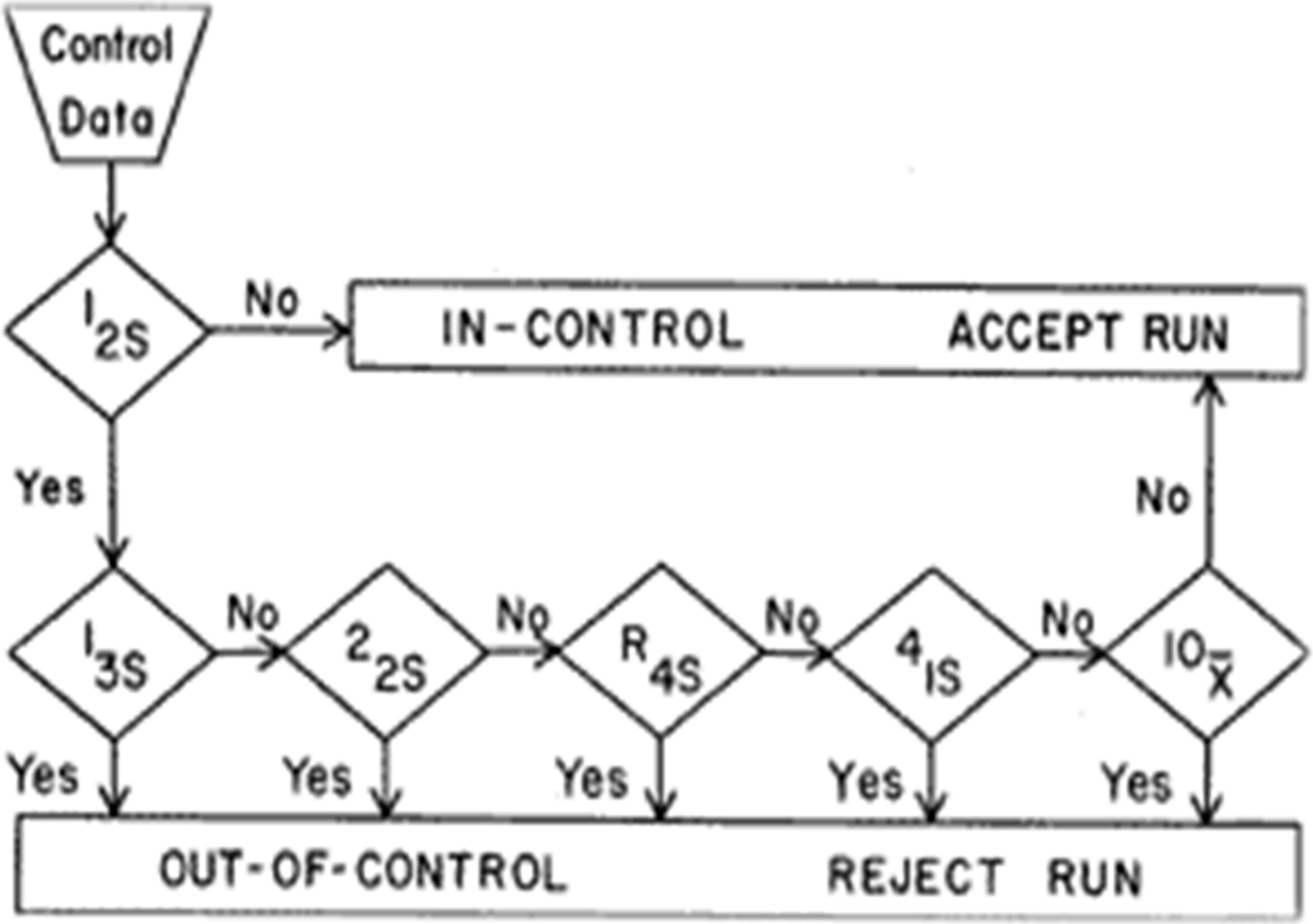


Look for a "trend"

7_T - reject when seven control measurements trend in the same direction, i.e., get progressively higher or progressively lower.



Summary of L-J Interpretation



DAILY IQC
HEMATOLOGY ANALYZERS

PROTOCOL FOR QUALITY CHECKS (IQC) OF HEMATOLOGY CELL COUNTER

First step

- **Start up status and background count check**
  (Monitor for acceptable background…..if required take corrective action)

Done once

- **QC monitoring using multilevel controls (Low level, normal level and high level)** (Monitor L-J chart, apply Westergard's interpretation, monitor %CV…..if required take corrective action)

Periodic in b/w check till shut down

- **QC monitoring using retained sample**
  (Monitor %CV…..if required take corrective action)

PROBABLE CORRECTIVE MEASURES WHEN CONTROL IS OUT

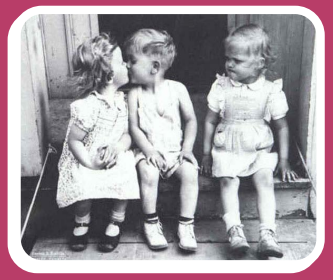
- Inform the supervisory staff.
- Check the QC material open vial expiry date, discoloration, expiry date.
- Check the temperature of the refrigerator in which the control material was kept.
- Check the reagent (expiry date, opening date etc.)
- Check the instrument status. (check priming, dispensing etc.)
- Rerun the controls.
- Run the normal retained sample.
- If problem persists run the fresh controls.
- If problem still persists call the engineer and also refer manufacturers troubleshooting guide.

  - And many more can be included / excluded depending on the laboratory set up.

OTHER ADDITIVES TO QUALITY CHECKS (IQC)

Comparative study (Z-Score)

Monitoring of Population mean (Bulls algorithm)



Rerun on same analyzer in special mode Rerun on another analyzer

Correlation with peripheral smears

Delta checks

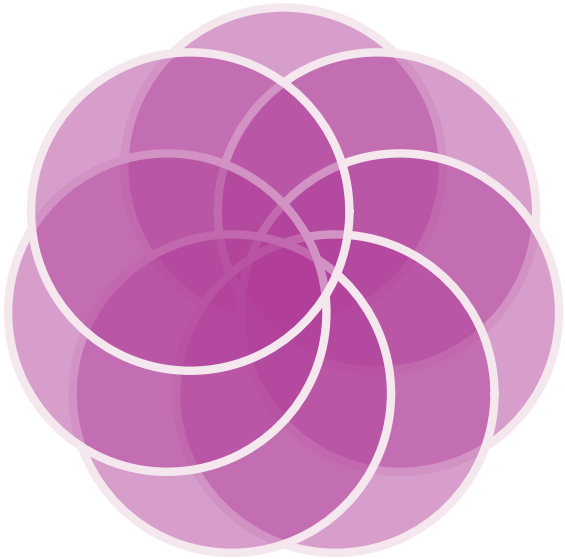
PERIPHERAL SMEAR STUDY NOT REQUIRED FOR ALL CBCS

Probable criteria when peripheral smear is made

Absolute lymphocytosis in any new case > 5 x 10^9/L

Absolute monocytosis in any new case > 1 x 10^9/L

Absolute basophilia in any new case > 0.1 x 10^9/L



Blast flag in the scatter plot in any new case.
Platelet clump flag and PIC/POC (Plt count <100).
Malaria like picture in the scatter plot.

All new cases of leukemia.

New cases of Bicytopenia / pancytopenia :

- Hemoglobin < 10 gm%
- Total Leukocyte Count <4 x 10^9/L
- Platelet Count < 100 x 10^9/L

Total Leukocyte Count or Platelet Count vote out even after rerun.

EXTERNAL QUALITY ASSESSMENT (PROFICIENCY TESTING)

COMPARATIVE PERFORMANCE EVALUATION

Z-Score

	A - 1	A - 2	A - 3	A - 4	A - 5	A - 6	A - 7	A - 8	A - 9	A-10	A-11	A-12	MEAN	SD
WBC	5.9	6.3	6.2	5.2	6.3	6.2	5.9	6.3	6.2	5.9	6.3	6.2	6.1	0.3
HGB	13.1	12.9	12.7	13.1	12.9	12.7	13.1	12.9	12.7	13.1	12.9	12.7	12.9	0.2
PLT	262	246	255	262	246	255	262	246	300	262	246	255	258.1	14.9

Z-SCORE	A - 1	A - 2	A - 3	A - 4	A - 5	A - 6	A - 7	A - 8	A - 9	A-10	A-11	A-12
WBC	-0.55	0.70	0.39	-2.74	0.70	0.39	-0.55	0.70	0.39	-0.55	0.70	0.39
HGB	1.17	0.00	-1.17	1.17	0.00	-1.17	1.17	0.00	-1.17	1.17	0.00	-1.17
PLT	0.26	-0.81	-0.21	0.26	-0.81	-0.21	0.26	-0.81	2.82	0.26	-0.81	-0.21

Z SCORE SCALING :-

< 0.5 - Excellent performance

0.5 to 1.0 - Satisfactory

1 to 2 - Acceptable

> 2 - Defect requiring attention

$$\text{Z Score} = \frac{\text{Observed value - Weighted mean}}{\text{SD}}$$

EQAS (NATIONAL)

CBC/
peripheral smears/
reticulocyte count

Nodal org. - AIIMS, New Delhi

- Contact person - Dr. Renu Saxena

Coagulation studies

Nodal org. - CMC, Vellore

- Contact person - Dr. Alok Srivastav

IF EQAS RESULTS ARE OUT

Check for the IQC results during the period when the EQAS sample was analyzed.

Follow the instructions from the nodal organization.

QUALITY MANAGEMENT OF THE POST - ANALYTIC PHASE

REPORT

ISO 15189, Clause 5.8 says -

Laboratory results shall be

"Unambiguous & Meaningful relevant to the clinical problem"

REPORT FORMAT

The report shall contains at least the following information: ISO 15189:2007 Clause 5.8.3 (a to q)

- clear, unambiguous identification of the examination including, where appropriate, the measurement procedure
- the identification of the laboratory that issued the report
- unique identification and location of the patient, where possible, and destination of the report
- name or other unique identifier of the requester and the requester's address
- date and time of primary sample collection, when available and relevant to patient care, and time of receipt by the laboratory
- date and time of release of report which if not on the report shall be ready accessible when needed
- source and system (or primary sample type)
- results and examinations reported in SI units or units traceable to SI units where applicable
- biological reference interval, where applicable
- interpretation of results, where appropriate
- other comments (e.g. quality or adequacy of samples which may have compromised with the result, etc)
- identification of the person authorizing the release of results
- if relevant, original and corrected results
- signature of authorization of the person checking or releasing the report is received.

BEFORE REPORT DISPATCH … DELTA CHECK

A formal way of testing for aberrant results is known as `delta check`. The blood count parameters should not differ from recent tests in the previous 2-3 weeks by more than a certain amount.

The difference should generally be not more than:

For Hb and RBC	For WBC	For Platelet count
10 %	20-25 %	50 %

Assuming that the patient's clinical condition has not altered significantly

Delta check must be done before dispatch of the report.
This is only possible if the previous report of the patient and the clinical condition is provided.

BEFORE REPORT DISPATCH… AUTHENTICATE

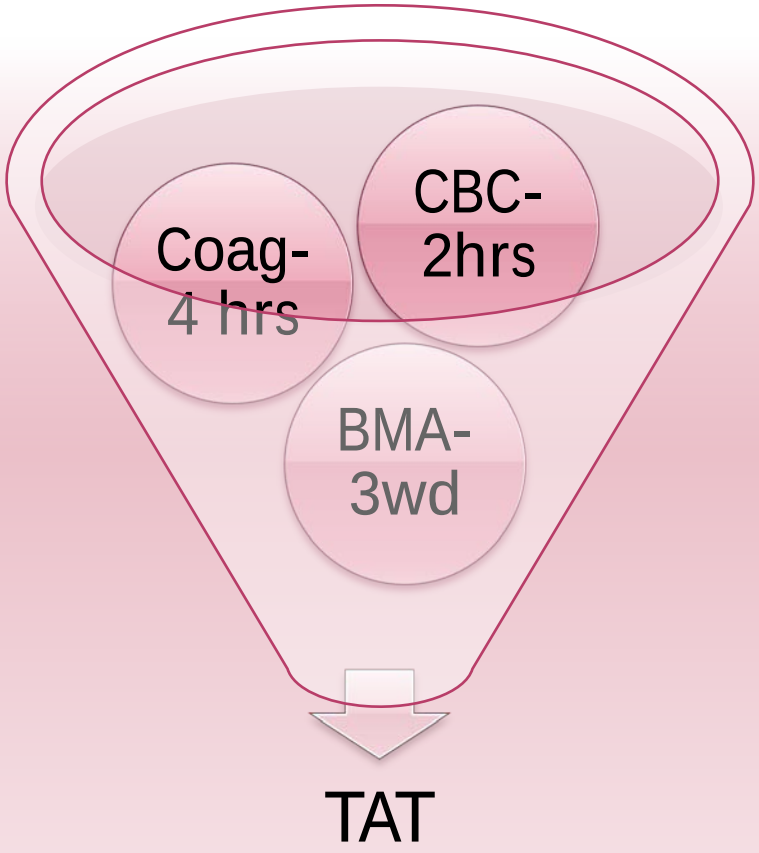
Each and every result generated is scrutinized and authenticated by the staff running the sample.

DURING REPORT DELIVERY

- The person at the dispatch counter delivers the reports to the identified individual.
- The receiver's identity, name and signature are taken and the time and date of dispatch is recorded in the register.

TURN AROUND TIME (TAT)

The laboratory takes responsibility for reporting the results within the specified turn around time.



The requester i.e. the clinician is notified in case of delay in examination only in such cases where the delay can compromise patients care.

Quality Indicator – "TAT shall be monitored on periodic basis for continual improvement"

Laboratory test results

Conclusion

- ✓ Reliable
- ✓ Timely
- ✓ Cost effective

Quality

Query's and suggestions

are welcome

THANK YOU