Candidates should be familiar with the theoretical principles and techniques underlying the full range of clinical biochemical analyses. Emphasis should be placed upon the factors which govern the choice of method and on the evaluation of instruments and methods.

The headings and subsections mentioned below indicate those methods, instruments, and analytical techniques, whose principles should be understood. Knowledge concerning the physico-chemical principles and rationale behind the basic design (rather than precise technical details) of the instruments, and methods of assessing their performance, may be expected. Candidates should be able to discuss the uses of the different classes of instruments and the relative merits which lead to their selection in various analytical situations.

The candidate must obtain adequate laboratory experience which is the basis of successful study. Practical experience is the main guide to understanding which topics should be known in detail or in outline only.

**General Physical Techniques**
- Fractional distillation, for example, preparation of solvents.
- Reverse osmosis.
- Solvent extraction, partition coefficients.
- Ultra-centrifugation.
- Freeze-drying.
- Mass spectrometry (including tandem).
- Preparation of high quality water

**Photometric methods**
- Absorptometry.
- Spectrophotometry.
- Fluorimetry.
- Flame spectrometry.
- Nephelometry.
- Turbidimetry.
- Atomic absorption spectrophotometry, flame and flameless.
- Flame emmision spectroscopy
- Inductively coupled plasma emission.
- ICPMS.

**Volumetric Methods**, including complexometric titration.

**Gasometric Methods**

**Electrometric Methods**
- pH, including CO₂-responsive systems.
- Other ion-sensitive electrodes, for example, Na⁺, K⁺, Ca²⁺
- Redox, including O₂-responsive systems.
- Titrations, poteniiometric, amperometric, and conductimetric.
- The use of such methods in dynamic situations such as enzyme rate determination and for semi-automatic analysis should be understood.
- Ion selective electrodes
- Biosensors

**Chromatographic and Electrophoretic Techniques**, both quantitative and qualitative applications.
- Various supporting media, for example, paper, membranes, gels, thin layer, ion exchange resins
- Molecular sieves, disc, column, continuous flow, and others.
- Immuno-electrophoresis, conventional, medium and high voltage electrophoresis.
- Isoelectric focusing.
- Gas and high-pressure liquid chromatography.
- Sample preparation, dialysis, desalting, concentration, preparation of derivates.
- Capillary electrophorsis.

**Imunochemical Techniques**
- Qualitative techniques for identification of proteins.
- Quantitative techniques for measuring concentration of specific proteins, radial immunodiffusion, nephelometry or turbidimetry.
- Laurell rocket, cross-immuno-electrophoresis.
- Competitive binding techniques, radioimmunoassay and immunoassay using non-isotopic labels, for example, enzymes.
- Enzyme immunoassay (see 3.10).
- Standardisation of these techniques and their use for various proteins in body fluids.
- Homogeneous assays
Isotope Techniques

Legal requirements for storage and disposal and permission to administer to humans. Use in quantitative and qualitative analysis, for example, single and double isotope dilution techniques, autoradiography. Radio-ligand assay. Use in radioimmunoassay. Problems of purity of labelled compounds, storage and specific activity. Immunoradiometric assay. Labels. Functional Sensitivity of an assay. Interference in assays – e.g. heterophile antibodies.

Osmometry
Methods of measurement, for example, depression of freezing point. Osmolar gaps

Enzymology

Mechanised Techniques and Work Simplification

Laboratory Data Processing and Computing
Use of computers for data collection, processing, and as management tools. Expert Systems

Analysis of Laboratory Error/Statistics
Concepts of reference range, analytical error, biological variation, and various simple parameters for describing data (mean, mode, standard deviation, confidence limits, variance, tests of significance). Applications to: assessment of inaccuracy and imprecision, errors of instruments, pipettes, and other equipment. Quality control methods.

Trace Elements.
Zinc, Copper, Aluminium, Lead, Arsenic, Chromium, Cadmium, Mercury.

Quantities and Units
SI Units - their advantages and disadvantages.

Specimen Collection, Preservation, and Preparation for Analysis
Constituent stability. Documentation and specimen flow systems. Interferences in the collection process.
CLINICAL BIOCHEMISTRY

The clinical biochemist should have an understanding of the major biochemical abnormalities found in disease and their methods of detection in the laboratory. They should have a general knowledge of the interpretative aspects of clinical biochemistry.

A Clinical Biochemist should understand the principles of testing the more important biochemical and physiological functions of organs or organ systems and be able to advise clinicians on their performance. They should take every opportunity to consult with clinicians to improve their understanding of the clinical manifestations of disease.

Water and Electrolytes
Distribution of water and electrolytes
Measurement of plasma volume, total body water, sodium and potassium spaces
Causes of hyper and hyponatraemia and hyper and hypokalaemia.
Osmolality. Hyperosmolar coma
Plasma and urine osmolality
Shock. Metabolic effects of trauma
Diagnosis and quantitative assessment of water and electrolyte loss
Diuresis; pharmacological - osmotic
Measurement of intracellular electrolytes
Distinction between diabetes insipidus and compulsive water drinking
Syndrome of inappropriate ADH.

Respiratory Function: H+ Metabolism
H+ (pH), pCO₂, pO₂, oxygen saturation
Lactate and pyruvate levels
Simple lung function tests
Assessment of body deficit or excess of H+
Understanding of acid/base disorders

Renal Function
Clearance tests: measurement of glomerular filtration rate and renal plasma flow.
Tubular function tests: concentration tests, ammonium chloride loading tests, amino acid chromatography, and renal glycosuria.
Normal and abnormal urine composition including abnormal pigments.
Proteinuria, differential protein clearance.
Renal failure. The nephrotic syndrome.
Renal Calculi.

Diabetes Mellitus and Hypoglycaemia
Substrate, neural, and endocrine regulation of insulin and glucagon secretion - the gut as an endocrine organ.
Diagnosis of diabetes mellitus and monitoring by glycated haemoglobin and urine albumin excretion.
Somatostatin. C-peptide.
Glucose tolerance, glycosuria. Plasma insulin and glucagon measurement.
Differential diagnosis of coma: ketoacidosis, lactate acidosis, hyperosmolar coma, and hyper-glycaemia.
Diagnosis of insulinoma and other causes of hypoglycaemia; use and dangers of provocative tests, for example, tolbutamide and glucagon.
Classification and diagnosis of DM
Value and limitations of plasma insulin assays in hypoglycaemia and hyperglycaemia.
Glycosylated haemoglobins and proteins.

Diagnostic Enzymology
Types of assays, enzyme determination in serum, urine, and cells.
Stability of enzymes.
Diagnostic use of enzymes.
Use of isoenzymes.

Proteins: The Serum Proteins in Health and Disease
Interpretation of electrophoretic protein patterns, recognition of paraproteins and their further investigation by immunological techniques.
Causes of hypoalbuminaemia. Assessment of protein-losing enteropathy and renal loss of protein. Effects that malnutrition and malabsorption can have on protein state. Disorders of immunoglobulins and changes in specific proteins, for example, alpha1-antitrypsin, transferrin, and their use in diagnostics.
Urine proteins, including Bence Jones protein. Protein selectivity as an indication of renal damage.

Gastric Function
Stimulation tests using pentagastrin and insulin.
Secretory levels in pernicious anaemia, peptic ulcer, neoplastic disease, and Zollinger-Ellison syndrome.
Urea breath tests.
Intestinal Function
Absorption tests, for example, glucose, xylose, fat, lactose, iron.
Pancreatic enzymes, secretin and pancreozymin test, Lundh test.
Disaccharidases.
Balance studies (associated with food and urine analyses).
Faecal analyses; fat, nitrogen, sugars (in children).
Electrolytes, including analyses of ileostomy fluid.
Sweat tests.
Hydrogen breath testing.

Liver Function
Metabolic disturbances in liver disease
Bilirubin and conjugated bilirubin
Urobilinogen
Urobilin
Enzymes in liver disease, for example, alkaline phosphatase, γ-glutamyl transferase
Aminotransferases
Protein synthesis, particularly albumin
Immunoglobulin changes in liver disease, alphafetoprotein
Cholesterol. Bile salt metabolism
Bromsulphalein excretion test
Differential diagnosis of disease producing jaundice
Diagnosis of non-icteric liver disease
Serological and PCR markers in diagnosis and monitoring of liver disease (Hep A,B,C)

Lipids
Biochemical basis and limitations of classifications of lipoprotein disorders
Genetic and acquired disorders of triglyceride, lipoprotein, and cholesterol metabolism
Hyper and hypolipoproteinemia
Theories of atherogenesis and coronary heart disease
Investigations and principles of treatment of hyperlipidaemias
Lipoprotein (a).

Calcium, Magnesium and Bone
Properties and actions of parathyroid hormone, calcitonin, and Vitamin D. Metabolism of Vitamin D to its hormonal form.
Regulation of secretion of hormonal Vitamin D
Parathormone
Calcitonin
Significance of the concentration of calcium, phosphate, and magnesium in plasma. Urinary excretion of calcium and renal tubular handling of calcium and phosphate.
Differential diagnosis of hypercalcaemia and hypocalcaemia.
Pathogenesis of renal stones
Metabolic bone disease
Magnesium metabolism; causes and effects of deficiency
Collagen crosslinks eg deoxypyridinoline.

Endocrine Function
The hypothalamic-pituitary-adrenal axis.
Investigation of infertility in males and females.
Investigation of hirsutism in females.
Monitoring of IVF therapy.
Diagnosis of thyrotoxicosis and myxoedema. In vitro and in vivo function tests. Use of TRH.
Growth hormone and prolactin.
Stimulation and suppression tests.
Somatostatin. Hypothalamic releasing and inhibitory hormones.
Steroid synthesis and metabolism. Congenital adrenal hyperplasia.
Mechanisms of hormone action, receptors, cyclic AMP, and cyclic GMP.
Transport of hormones
Renin-angiotensin-aldosterone system
Hormones of the gastro-intestinal tract

Nutrition
The digestion of proteins, carbohydrates, and lipids and the biological role of vitamins and the trace elements.
The nutritional concept of protein quality and its assessment by measurement of biological value and nitrogen balance; also the importance of these factors in patients on synthetic diets.
The laboratory methods of assessing vitamin status and measuring trace elements.
Inborn Errors of Metabolism
Possible defects in protein biosynthesis arising from genetic mutations. Quantitative and qualitative enzyme abnormalities occurring in genetic disorders. The biochemical consequences of a primary enzyme block in a metabolic pathway and the ways in which clinical and pathological signs may be produced. Methods of detecting metabolic disorders, with particular consideration of screening selected clinical groups, for example, the mentally subnormal and the newborn. Evaluation of detection programmes. Antenatal diagnosis. Methods of treatment, particularly by dietary restrictions and vitamin supplementation, and the biochemical monitoring of the treatment.

Haem and Porphyrins

Toxicology and Drugs
Detection and quantification of common drugs in therapy, for example, digoxin, lithium, and anticonvulsants. Overdosage, for example, salicylates, paracetamol, and barbiturates, and suspected addiction, for example alcohol, morphine, morphine derivatives, and amphetamines. Differential diagnosis of coma. Metabolic effects of ethanol. Environmental hazards, for example, lead, mercury.

Cerebrospinal Fluid

Amniotic Fluid
Bilirubin, creatinine content Presence and significance of alpha-fetoprotein. Lecithin/sphingomyelin ratio, palmitate, and other tests of foetal lung maturity. Screening for Down syndrome.

The Biochemical Effects of Neoplasia
Effects of tumors, both anatomical and pathological. Tumor markers, their biochemical and pathological significance and their use in management of benign and malignant tumors. Some examples of this are: alpha fetoprotein, hCG, CEA, ectopic production of hormones and the syndromes these cause.

Molecular Biology
Principles of PCR, Northern, Southern and Western Blots. Testing for common diseases (HFE, CF)

Cardiac Markers
Troponins, CK-MB, CK-isoforms, myoglobin. Homocysteine
LABORATORY MANAGEMENT

It is not expected that the candidate will have had the opportunity to become fully conversant with all details of laboratory management. The Clinical Biochemist should however, have a reasonable knowledge of the important aspects of the following:

Organisation of a clinical biochemistry laboratory, including routine and emergency services.
Screening and profiling.
Staff training, performance management, and work assignment.
Laboratory safety including chemical, radiation, physical and biological hazards.
Reagents and apparatus, their selection, sources of supply, and techniques for assessing the quality of equipment and reagents.
Budget preparation and monitoring.
Presentation of results of biochemical analysis, reports of results.
Laboratory design.
Quality control implementation, monitoring, performance evaluation.
Laboratory statistics. Use of mean, mode, median, standard deviation, variance, standard error of mean, analysis of variance, F-test, t-test, and non-parametric statistics. Regression analysis.
Method Comparison. Use of regression analysis, Bland-Altman plots. NCCLS and AACB guidelines for method comparison.
Determination of Reference Intervals Sample selection. Statistical analysis (detection of outliers, sample size consideration) Parametric and non-parametric methods including confirmation of Gaussian distribution.
Quality Management Systems ISOguide 25, ISO 9000 systems.

Efficiency of Laboratory Testing Strategies
Diagnostic sensitivity, specificity and efficiency of tests, ROC curves.
Evidence Based Medicine

SPECIAL PROJECTS
Short-term projects involving a considerable number of skills required of a laboratory supervisor (analytical, instrumental, evaluative, managerial, organisational) should be undertaken to encourage initiative and independence. The candidate should acquire an ability for clear report writing and should be encouraged to write assays on topics for discussion with his supervisor.

EDUCATIONAL ACTIVITIES
The candidate should plan his course of study in preparation for the Membership examination in consultation with their supervisor and the State Branch Education Representative.

The candidate should attend the following activities as an adjunct to higher own studies and practical experience.

(a) Regular seminars on clinical biochemistry run by the Association.
(b) Appropriate lectures, seminars, discussions or case presentations held in hospitals or other institutions.
(c) The annual Course in Chemical Pathology held jointly by the Association and the Royal College of Pathologists of Australasia.
(d) The Annual Scientific Meeting of the Association.
(e) Use of the Internet e.g. web sites of the AACB, AACC, ACB.
(f) Use of Medline and other searches.