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METHODOLOGY: DEVELOPMENTS AND TRENDS

by
Dr N. Montalbetti
Director, Laboratory of Clinical Chemistry
St Anna Hospital
22100 Como, Italy



Background

Traditional analytical methods used in clinical endocrinology (bioassays, paper, column, thin-layer chromatography; colorimetry, fluorometry) often permitted only an indirect evaluation of hormones from their urinary catabolites and suffered from a lack of specificity, sensitivity and/or practicability.

During the last two decades, methods have been developed that make it possible to evaluate hormones in blood and other biological fluids directly, to separate their free-active fraction, and to study their transport in blood, their metabolism and their action on target cells. They include radioisotopic methods, gas-liquid chromatography, mass spectrometry, high performance liquid chromatography, competitive protein binding, radioimmunoassays and radioenzymatic assays.

Recent developments and trends

- Immunochemical methods

New markers are used as in enzymeimmunoassays (commercially available and used particularly for thyroid hormone) and in fluoroenzymeimmunoassays (less advanced, but very promising for steroid hormones); optimization of RIA increases practicability (solid-phase technique, automation) reduces costs (regenerable immobilized antibodies), and produces specific antibodies in large quantities, with constant characteristics, by means of hybrid cells.

- High performance liquid chromatography

The application of this technique, already used in the pharmaceutical industry for pure compounds, is rapidly growing in the endocrinological laboratory because of its versatility, simplicity and relatively low cost. Progress in instrumentation, with possible automation, will further promote its application.

- Radioreceptor assays

Methods using receptors for the determination of hormones are increasing: ACTH, LH, HPL, HCG, H.G.H. prolactin, insulin.

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- Receptor determination

Methods are available for determining membrane and cytoplasmatic receptors and nuclear acceptors of hormone-receptor complexes.

- Isoelectric focusing

This technique is used for the purification of proteic hormones.

- Chemiluminescence analysis

Procedures using this simple technique have been published for thyroxine, cortisol, progesterone and oestriol. More widespread application can be forecast when reagents and instruments are commercially available³. Development of sensors, eventually using an immobilized protein (enzyme or antibody) to recognize the hormone and an electrochemical or other physical detector, might greatly extend our knowledge and permit continuous monitoring of hormones in vivo.

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