

Growth Hormone (plasma, serum, saliva, urine)

1 Name and description of analyte

1.1 Name of analyte
Growth hormone

1.2 Alternative names
Somatotropin, somatotrophin, somatropin

1.3 NCLM code
To follow

1.4 Description of analyte
Growth hormone is a peptide hormone. Many forms are detectable in the circulation, predominantly a 22 kDa form, but 5 kDa, 9 kDa, 12 kDa, 16 kDa, 17 kDa, 20 kDa, 23 kDa, 27 kDa and 30 kDa forms have also been identified. GH exists in monomeric, dimeric or oligomeric (both non-covalent and disulphide linked, >45k Da) forms and it can be present in the plasma as a free hormone or bound to GH binding proteins. GH can undergo post-translational modifications, further increasing its heterogeneity, by glycosylation, de(s)amidation and acetylation. The lower molecular weight forms of GH generally show reduced or negligible biological activity and the oligomeric forms are also reported to have variably diminished bioactivity. Two growth hormone binding proteins exist in plasma. One is the extracellular domain of the growth hormone receptor, which binds GH with high affinity, and the other is a lower affinity binding protein with an α_2 -macroglobulin structure. The binding of GH to growth hormone binding proteins inhibits the interaction of GH with membrane receptors.

1.5 Function of analyte
Growth hormone is an anabolic hormone: it promotes somatic growth through the production of somatomedins in the liver and differentiation of mesenchymal-derived cell lineages (including adipocytes). It also has various metabolic activities: as a counter-regulatory hormone to insulin, in the retention of sodium, phosphorus and nitrogen, in the transport of amino acids into muscle and in stimulation of lipolysis, lactogenesis and immune function, although it could be argued that none of these actions is essential to life. Growth hormone is secreted in a pulsatile fashion with up to 6–12 discrete pulses per day giving an interpulse frequency of approximately 2h, with most secretion occurring during sleep. Women of reproductive age have higher pulse amplitude and higher baseline GH than men but both sexes have the same pulse frequency. The half-life of GH in plasma is 15-20 minutes.

2. Sample requirements and precautions

2.1 Medium in which measured
1. Plasma, serum or blood
2. Growth hormone can also be measured in saliva as a surrogate for free plasma concentration, but most recommendations for the use of growth hormone measurements relate to plasma/serum measurements. The

measurement of growth hormone in urine is largely confined to paediatric practice.

2.2 Precautions re sampling, handling etc.

Separation of serum from blood cells is recommended within 2 h of collection. GH is stable in serum for 8 h at 15–25 °C, for 1 day at 2–8 °C and for up to 2 months at -20 °C. Repeated thawing and freezing should be avoided.

3. Summary of clinical applications and limitations of measurements

3.1 Applications

1. Diagnosis and monitoring of treatment of acromegaly
2. Investigation and monitoring treatment of growth hormone deficiency
3. Investigation into the possibility that anabolic hormones have been taken to enhance physical performance.

3.2 Limitations

Physiological changes in growth hormone concentrations may be difficult to distinguish from pathological causes given the pulsatile secretion of the hormone and extent of intra-individual variation

4 Analytical considerations

4.1 Analytical methods

Growth hormone is routinely measured using immunoassay (immunometric, immunofluorometric, immunochemiluminometric assay or radioimmunoassay) using either polyclonal or monoclonal antibodies. Mass spectrometric assays are also available, primarily for research rather than routine use.

4.2 Reference method

No reference method has yet been explicitly defined. Most methods are traceable to mass spectroscopic techniques, for example involving tryptic digestion of the peptide hormone followed by chromatographic separation of the resulting peptides and either quantification by isotope dilution LC/MS-MS using the isotopically labelled forms of the peptides as internal standards or quantification of the peptide amino acid content after proteolysis and derivatisation by GC/MS-MS.

4.2 Reference materials

Recombinant DNA-derived human GH WHO NIBSC (National Institute for Biological Standards and Control: South Mimms, UK) 2nd IRP 98/574. Previous reference materials (on which methods used in some recently published papers are based) have included pituitary derived WHO IRP 66/217, which was replaced with purified pituitary derived WHO IRP 80/505 in 1990 and later by the biosynthetic WHO IRP 88/624 in 1995.

4.3 Interfering substances

The potential for interference is likely to be assay specific. However, it is commonly quoted that presence of haemoglobin in concentrations up to 0.5 g/dL (5 g/L), conjugated and unconjugated bilirubin in concentrations

up to 342 $\mu\text{mol/L}$ and the presence of triglycerides in concentrations up to 15 mmol/L have no effect on results within the precision of the assay. For assays using biotin, samples should not be collected from patients receiving therapy with high biotin doses ($>5 \text{ mg/day}$) until at least 8 h after the last biotin administration.

4.4 Sources of error

- Different assays have variable cross reactivity to the 20kDa and other isoforms of GH, placental GH, therapeutic GH analogues and pegvisomant.
- Interference due to GH binding proteins.
- The placenta produces human chorionic somatomammotropin (placental lactogen), which is a separate but structurally similar entity that may cross-react in GH assays.
- Prolactin has structural similarity to growth hormone and may cross-react in GH assays.
- The potential for a hook effect/prozone phenomenon should be considered with sandwich assays.
- General interferences in immunoassay are caused by heterophilic antibodies and rheumatoid factor.

5. Reference intervals and variance

5.1.1 Reference interval

The information below should be used as general guidance only. Reference intervals are assay dependent and typically in the region of 0.3–3.3 $\mu\text{g/L}$ for a random serum sample in an adult. Reference intervals for serum GH, where quoted, tend not differ between male and female adults. Gender differences in growth hormone secretion (with higher concentrations in women) may not be of clinical significance given the high biological variation in serum GH concentrations (see section 6.2). Urinary GH excretion in adults (mean \pm SD) has been reported as $1.38 \pm 1.24 \text{ ng/L}$ for men and $3.02 \pm 3.78 \text{ ng/L}$ for women on an overnight collection and a reference interval of $<15 \text{ ng/24h}$ has been quoted for a 24h urine collection for adults of both sexes.

Serum growth hormone concentrations in infants <3 months of age are reported to range from 2–35 $\mu\text{g/L}$ (median 16 $\mu\text{g/L}$) irrespective of glycaemic status, with a tendency to higher values in infants with a birth weight below the 10th centile for gestational age or those who were born prematurely. Reference intervals for serum GH through infancy and early childhood show a decrease in the upper limit of the reference interval with increasing age. The reference interval for overnight urinary GH excretion in prepubertal children >3 years of age has been reported in the range of 0.75–3.5 ng/night . There is a peak in urinary growth hormone excretion in children that occurs at Tanner stages 3 and 4, corresponding to an age interval of 11–18 y in boys and 9–15 y in girls. During pregnancy, pituitary GH expression in the mother is suppressed and a GH variant expressed by the placenta becomes the predominant form in plasma.

5.1.2 Extent of variation

5.1.2.1 Interindividual CV: 60–90%

5.1.2.2 Intraindividual CV: 40–61%

5.1.2.3 Index of individuality: 0.67

5.1.2.4 CV of method

It has been recommended that assays should be able to achieve a lower limit of quantification of 0.05 µg/L with a CV of <20%.

5.1.2.5 Critical difference: 178%

5.1.3 Sources of variation (effect on GH indicated by arrows)

- Time of sample collection, sleep/wake status
- Body mass (obesity ↓)
- Older adults ↓
- Food intake ↓
- Glycaemic status (low [glucose] ↑, high [glucose] ↓)
- Other sources of variation include steroids, renal function, liver disease, thyroid disease and hypogonadism (see section 7).

6 Clinical uses of measurement and interpretation of results

6.1 Uses of measurement

6.1.1 Diagnosis of acromegaly

The Endocrine Society guidelines (Katznelson L, Laws ER, Melmed S, *et al.* Acromegaly: An Endocrine Society Clinical Practice Guideline; J Clin Endocrinol Metab, 2014;99(11):3933–3951) suggest that a GH of <1 µg/L after a glucose load is sufficient for excluding the diagnosis, provided that hyperglycaemia is achieved. This test may be considered unnecessary if [IGF-1] is raised in conjunction with clinical features of growth hormone excess.

6.1.2 Monitoring treatment of acromegaly

Following surgery, a serum GH <0.14 µg/L suggests 'surgical remission'; a value of <1 µg/L indicates 'control' and normalization of the mortality risk (2014 guidelines). Treatment with somatostatin receptor ligands should be monitored using IGF-1 and GH concentrations. The utility of glucose-suppressed GH values is not clear. Treatment with growth hormone receptor antagonist (Pegvisomant), which blocks production of IGF-1, should not be monitored using GH concentrations, since GH hypersecretion persists and Pegvisomant interferes with GH assays.

6.1.3 Diagnosis of growth hormone deficiency

This may be dependent on the assay used for the measurement of growth hormone and the test used to stimulate growth hormone secretion. Severe GH deficiency is defined by a peak GH response of <3 µg/L. Generally, an increase in GH to a concentration >6.67 µg/L following a provocation test is considered to be an appropriate response in both adults and children. Higher cut-offs have higher specificity for normality (see section 8.1). Various stimuli can be used, with insulin-induced hypoglycaemia and glucagon considered to be the most reliable.

6.1.4 To aid the decision to give growth hormone replacement

The (UK) National Institute for Health and Care Excellence (NICE) has recommended in guidance TA64 (2003) that recombinant human GH should be used only for adults with severe deficiency that is severely affecting their quality of life. Suitability for GH replacement therapy is assessed by failure to show an increase in growth hormone of ≥3 µg/L in response to a reliable stimulation test in conjunction with an impaired quality of life assessed by a questionnaire that improves by at least seven points after initiation of therapy and concurrent prescription of other hormones for pituitary hormone insufficiency. Separate NICE guidance

(TA188 2010) states that GH may be prescribed for growth disturbance in children due to insufficient secretion of GH but the criteria used to make this diagnosis are not stated in the NICE guidance.

6.1.5 Diagnosis of GH insensitivity

A basal serum GH $>5 \mu\text{g/L}$ has been proposed as one of the criteria used to make a diagnosis of GH insensitivity syndrome in conjunction with measurements of basal and stimulated IGF-1, growth hormone binding protein (GHBP) and the patient's height.

6.2 Confounding factors

The release of GH is pulsatile and has a circadian rhythm, hence a low result on a random sample does not imply decreased pituitary function nor does a result within a reference interval on a random sample exclude growth hormone excess or deficiency, nor a raised result on a random sample imply growth hormone excess. Thus the value of isolated measurements of GH is limited. The peak GH response criteria is not applicable to pre-pubertal children in the absence of sex steroid priming; there is little published research regarding the need for sex steroid priming in adults in order to achieve an optimum GH response, but this may also be the case.

7 Causes of abnormal results

7.1 High concentrations

Causes include:

- acromegaly due to pituitary adenoma, growth hormone releasing hormone secreting tumours or ghrelin secreting tumours
- genetic disorders causing gigantism
- growth hormone resistance
- treatment with growth hormone or growth hormone receptor antagonists
- hyperthyroidism
- hypoglycaemia
- hyperandrogenism
- other causes.

7.1.1 Investigation

A random growth hormone $\geq 1 \mu\text{g/L}$ and/or a failure to suppress growth hormone to a concentration of $< 1 \mu\text{g/L}$ during an oral glucose tolerance test is consistent with growth hormone excess (see section 6.1). These cut-off values should be regarded as for guidance only, since many exceptions have been found in clinical practice.

7.2 Low values

Causes :

- congenital GH deficiency (may occur in isolation or combination with deficiencies in other pituitary hormones), GHRH receptor gene defects, GH secretagogue receptor gene defects, GH gene defects, brain structural defects and midline facial defects
- tumours of hypothalamus or pituitary
- infiltrative/granulomatous disease of the central nervous system
- infective disease of the central nervous system
- cranial irradiation or chemotherapy
- surgery to the pituitary or hypothalamus

- pituitary infarction
- brain damage/cranial trauma
- hypothyroidism
- hypogonadism
- glucocorticoid excess
- acquired growth hormone deficiency may be idiopathic
- other causes.

7.2.1 Investigation

In an adult, a failure to show an increase in [GH] following a provocation test (see section 6.1.4) is consistent with growth hormone deficiency. A random value of <9.7 µg/L is suggestive of growth hormone deficiency in term neonates aged 3–5 days. Patients with three or more pituitary hormone deficiencies and a low [IGF-1] are considered to be growth hormone deficient in the absence of provocative testing.

7.3 Notes

Physiological causes for high values include age (neonatal, puberty/adolescence), pregnancy, exposure to cold or heat and exercise. Physiological causes for low values include obesity, old age (somatopause) and critical illness. Concentrations of GH fluctuate in response to nutritional factors, being lower after food ingestion and higher during fasting. In addition to diurnal variation (higher during sleep), GH concentrations also show seasonal variation (higher in the autumn than the spring).

8 Performance

8.1 Sensitivity, specificity etc. for individual conditions

- Growth hormone <9.7 µg/L in a neonate at 3–5 days of age has a sensitivity of 100% and a specificity of 98% for growth hormone deficiency.
- The insulin stress test has 96% sensitivity and 92% specificity for the diagnosis of growth hormone deficiency at a GH cut-off of 5.1 µg/L.

9 Systematic reviews and guidelines

9.1 Systematic reviews

Numerous reviews are available on the use of growth hormone therapy but are outside the scope of this document.

9.2 Guidelines

1. NICE Technology Appraisal TA64. Growth hormone deficiency (adults) - human growth hormone, August 2003.
2. NICE Technology Appraisal TA188. Human growth hormone (somatropin) for the treatment of growth failure in children, May 2010.
3. Molitch ME, Clemmons DR, Malozowski S *et al.* Evaluation and treatment of adult growth hormone deficiency: An Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab* 2011;96:1587-1609.

9.3 Recommendations

1. Giustina A, Barkan A, Casanueva FF *et al*. Criteria for Cure of Acromegaly: A Consensus Statement. *J Clin Endocrinol Metab* 2000;85:526-529.
2. Giustina A, Chanson P, Bronstein MD *et al*. A Consensus on Criteria for Cure of Acromegaly. *J Clin Endocrinol Metab* 2010; 95:3141-3148.
3. Andersson A-M, Orskov H, Ranke MB *et al*. Interpretation of growth hormone provocative tests: comparison of cut-off values in four European laboratories. *Eur J Endocrinol* 1995;132: 340-343.
4. Trainer PJ, Barth J, Surgeon C *et al*. Consensus statement on the standardisation of GH assays. *Eur J Endocrinol* 2006;155:1-2.
5. Clemmons DR on behalf of the conference participants, Consensus statement on the standardisation and evaluation of growth hormone and insulin-like growth factor assays. *Clin Chem* 2011; 57: 555-559.

10. Links

10.1 Related analytes

Prolactin is related to growth hormone via the growth hormone-prolactin gene family and has a similar structure.

10.2 Related tests

Insulin-like growth factor 1 (somatomedin)

Author: Dr Jinny Jeffery

Date Completed: 1.2015

Date Revised: