

PRODUCT SPECIFICATION

Anti-Human IGFBP-3 Antiserum, Product Code IGF63

(for in-vitro use only)

SOURCE:	Derived from rabbit serum raised against highly purified human IGFBP-3 (acid-stable binding subunit).
APPEARANCE / STATE:	Lyophilized material.
SPECIFICITY:	Detects both the complete 150 kD IGFBP-3 complex as well as smaller IGFBP-3 fragments. Cross-reactivity with hIGFBP-1 and hIGFBP-2 \ll 0.05 %.
SPECIES SPECIFICITY:	Specific for human and primate IGFBP-3.
STORAGE:	Store at 4°C (or -20°C respectively, after reconstitution). For storage after reconstitution, we recommend the addition of an appropriate preservative (e.g. 0.05% sodium azide). Avoid repeated freezing and thawing.
APPLICATION:	Radioimmunoassay (recomm. final dilution: 1 : 21 000)
AVAILABLE QUANTITIES:	2.5 µl (sufficient for 250 RIA tubes) 10 µl (sufficient for 1 000 RIA tubes)
RECONSTITUTION:	It is suggested to reconstitute the antiserum in PBS (e.g., 250 µl or 1 000 µl respectively, dilution factor 1:100). It is recommended to keep reconstituted reagents at room temperature for half an hour and to shake them vigorously on a vortex mixer before use.

Suggestions for Establishing an IGFBP-3 RIA

Reagents

Assay Buffer (AB):	0.05 M Sodium phosphate (PBS), pH 7.4 0.1 M Sodium chloride 0.25% BSA 0.05% Sodium azide 0.1% Triton X 100
Non-specific binding [NSB]:	Rabbit immunoglobulin: 50 µg/ml in AB
1st Antibody:	- Anti-hIGFBP-3 1:7 000 - Rabbit immunoglobulin: 50 µg/ml mix and dissolve in AB
Tracer:	¹²⁵ I-IGFBP-3: 200 000 cpm/ml in AB
Standards:	Geometric dilutions of hIGFBP-3 in AB (0.625 ng/ml ≤ x ≤ 40 ng/ml)
2nd Antibody:	Appropriate dilution of anti-rabbit IgG (e.g. 1:250) in 4% polyethyleneglycol 6 000.
Samples:	Serum or plasma samples should be diluted 1:500 or 1:600 with AB. Cell culture media may be used undiluted or diluted up to 1:30, depending on the expected IGFBP-3 concentration.

Assay Procedure:

- Mix 100 µl standard or sample with 100 µl of the 1st antibody solution and 100 µl of the tracer.
- Incubate for 2 days at 4°C.
- Add 500 µl of the cold 2nd antibody solution, mix and incubate for 1 hour at 4°C.
- Centrifuge and wash the precipitate with 1 ml of cold water.
- Count the precipitate in a gamma-counter.