

**Anti-human Insulin-like Growth Factor-1
Monoclonal Antibody 6C9-2E5
IgG Fraction of Ascites Fluid**

MC-2717

Lot # 8803

This IgG fraction contains mouse monoclonal antibody 6C9-2E5 raised against recombinant hIGF-1. This monoclonal antibody has been found to bind specifically to hIGF-1 by indirect ELISA, cytochemistry and western blots. This monoclonal antibody was tested for recognition of insulin, IGF-2, and other growth factors by ELISA techniques. It has been found to be a mouse IgG1 kappa by isotyping. Each vial contains 50 µg of highly purified IgG which was lyophilized from a volatile buffer solution.

Monoclonal Antibody Specificity

Polypeptide	% Cross Reactivity
hIGF-1	100
hIGF-2	0
Insulin (human)	0
EGF (human)	0
TGF α	0

Indirect ELISA

This IgG fraction has been found by indirect ELISA to bind to hIGF-1 at concentrations of 100 ng/well or less.

Western Immunoblot

Western immunoblots resulted in a single band being detected at ~ 7.5 kDa using 50 ng/lane of this IgG fraction.

Western Blotting Protocol

1. After SDS-PAGE (on either 4-15% gradient gels or single percentage gels, such as 12% gels) and electrophoretic transfer to PVDF membrane, block the membrane overnight with 4% normal goat serum in TBS/Tween-20 buffer.
2. Wash x 2 with TBS/Tween-20.
3. Apply the monoclonal antibody after dilution at least 50 ng/ml. (Note: higher dilutions may be needed). Use 1% normal goat serum in TBS/Tween-20 as buffer. Let the primary antibody bind for 2-4 hours.
4. Wash x 3 with TBS/Tween-20.
5. Apply affinity purified HRP-goat anti-mouse IgG antiserum diluted 1:2500 (Note: dilution may vary depending upon supplier) in 1% normal goat serum in TBS/Tween-20. Incubate 1-2 hours. Note: greater sensitivity may be achieved using ABC techniques.
6. Wash x 4 for 5 minutes per cycle in TBS/Tween-20.
7. Develop color using the enhanced DAB reaction or develop film using ECL or other chemiluminescent techniques.