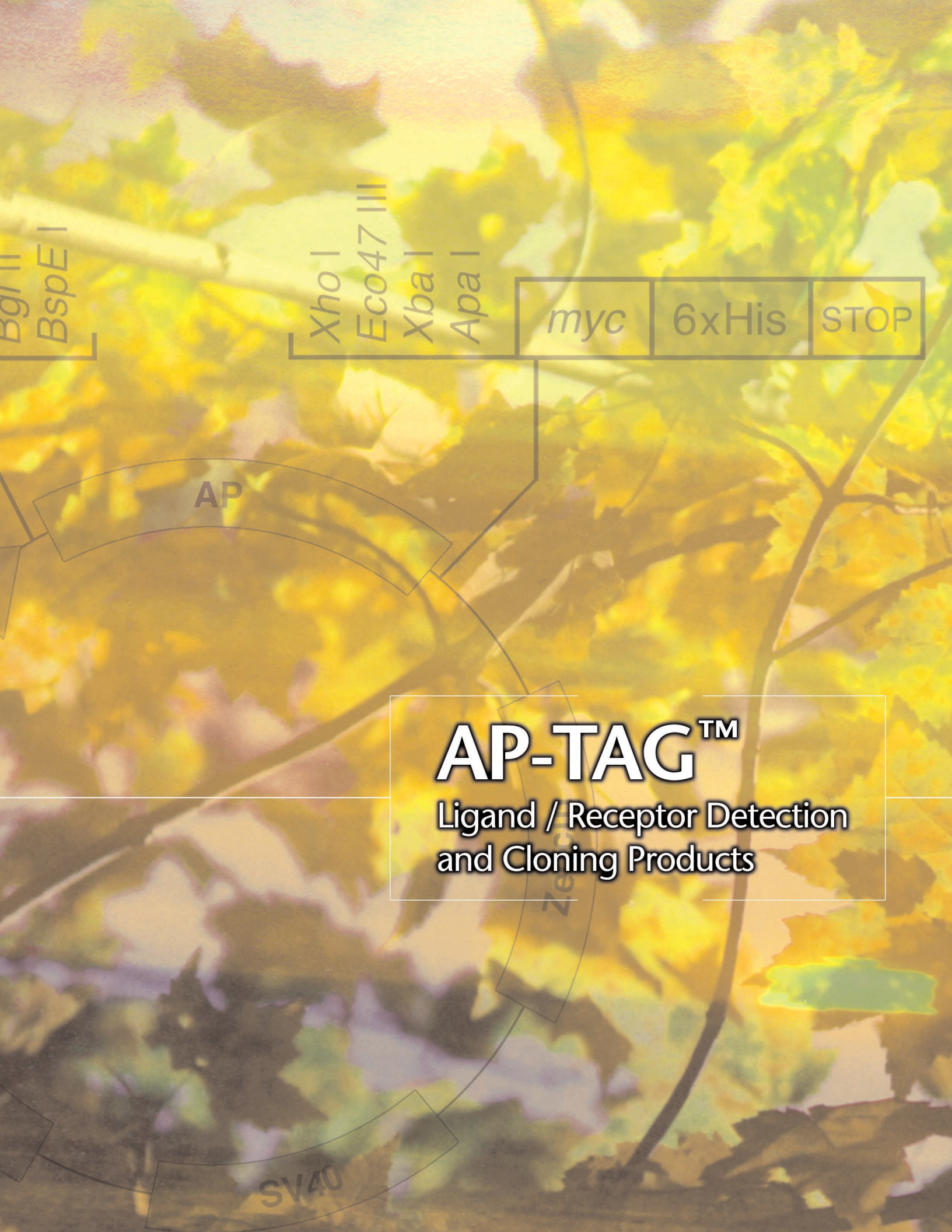


AP-TAG™

Ligand / Receptor Detection and Cloning Products

AP-TAG® Introduction

AP-TAG® technology (US patents 5,554,499 and 5,801,000; ref. 1), invented by Drs. J. Flanagan and P. Leder at Harvard Medical School, has revolutionized the way cell surface receptors and ligands are detected and cloned. GenHunter is proud to be the exclusive licensee of this powerful method. Purchase of an AP-TAG® Kit or any pAPtag vector comes with a limited, single-user and non-transferable sublicense for use in research applications only. No part of the kit or pAPtag plasmid vectors shall be disseminated, propagated or distributed outside the user's own laboratory without written permission from GenHunter. A separate license is required for drug screening or other commercial applications. Contact GenHunter for details.

The essence of this invention is to allow a cDNA sequence encoding any secreted polypeptide ligand or extracellular domain of a receptor to be in-frame fused to human placental secreted alkaline phosphatase (AP) in pAPtag cloning vectors. The resulting AP fusion protein, designated as an AP-body™, when expressed in 293T cells, can be secreted at high levels into the culture medium and thus easily detected by either the AP activity assay or Western blot analysis using antibody against AP. The ligand-AP or soluble receptor-AP fusion proteins thus can serve as affinity agents much like antibodies, which allow the most convenient, safe, and sensitive detection and cloning of their corresponding cell surface receptors or ligands. Unlike the conventional radioactive ^{125}I labeling method, AP-TAG® is safe and does not require ligand/soluble receptor purification.

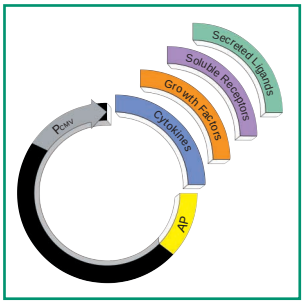
Since its invention, many important cell surface receptors and ligands have been cloned by AP-TAG™ technology including receptors for **Leptin**, **Semaphorin III**, **Nogo-66**, **IL-24**, **Jelly Belly**, and ligands for **Kit**, **Mek4** and **Sek receptor tyrosine kinases** (see references on page 46). A more extensive list of publications using AP-TAG® technology can be found on page 59 or on our website.

GenHunter was extremely pleased to be able to add this innovative method into our product line as a powerful tool for applications downstream of differential display (DD). If you are working with a secreted protein or cell surface molecule cloned by DD or other methods, AP-TAG® technology may allow you to functionally characterize these genes further.

Comparison of AP-TAG® technology and the conventional radioactive ^{125}I ligand-labeling method:

	AP-TAG®	^{125}I labeling
Ligand purification	Not required	Required
Labeling Reaction	Not required	Required
Hazardous	No	Yes
Detection	Colorimetric	Scintillation counting
Sensitivity	High	High
Cell Staining	Yes	No
Expression Cloning	Yes	Yes
Ligand-Receptor Binding Kinetics/Affinity	Yes	Yes

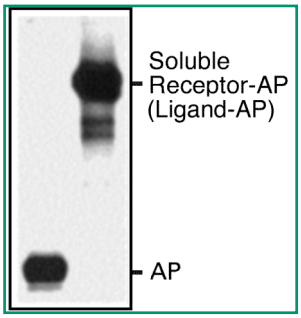
Schematic Illustration of AP-TAG® technology and its major applications



AP fusion construct

Create an in-frame fusion of your cDNA encoding a secreted ligand or soluble receptor with either the N- or C-terminus of secreted alkaline phosphatase (AP) in pAPtag expression vectors.

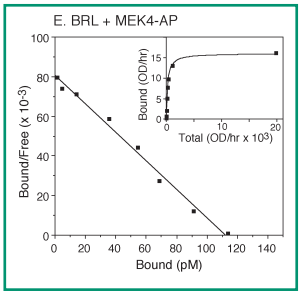
GenHunter Products: AP-TAG® Kit A
AP-TAG® Kit B



AP fusion protein expression

After transfecting the above AP fusion plasmid construct into 293T or NIH 3T3 cells, the expression of the secreted AP fusion protein (AP-body™) can be measured by either colorimetric AP activity assay or immunoblotting (or IP) with antibody to AP.

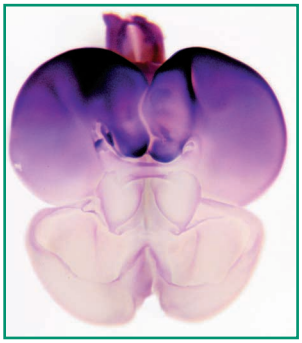
GenHunter Products: 293T Cells
AP Antibody (Polyclonal and Monoclonal)
AP Assay Reagent A
Monoclonal AP Antibody Sepharose Beads



Receptor/ligand binding assay

The culture medium containing the secreted AP fusion protein can be used directly to measure the presence or absence of a cell surface receptor (or ligand) of interest by assaying the AP activity bound to the cells. The secreted AP alone is used as a negative control.

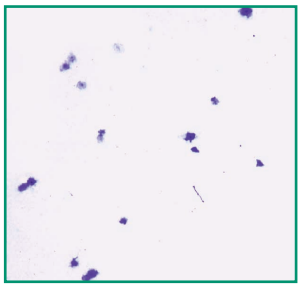
GenHunter Products: AP Assay Reagent A
293T/pAPtag-4 stable cell line
AP control AP-body™



in situ staining of receptor/ligand

The secreted AP fusion protein can be used much like an antibody to detect the tissue distribution of a cell surface receptor/ligand of interest. An expression cDNA library thus can be made with mRNA isolated from tissues that express the highest level of the receptor/ligand for subsequent expression cloning.

GenHunter Products: AP Assay Reagent S



Expression cloning of receptor/ligand

The secreted AP fusion protein can be used as a probe to clone a cell surface receptor or ligand of interest by traditional expression cloning strategy (panning).

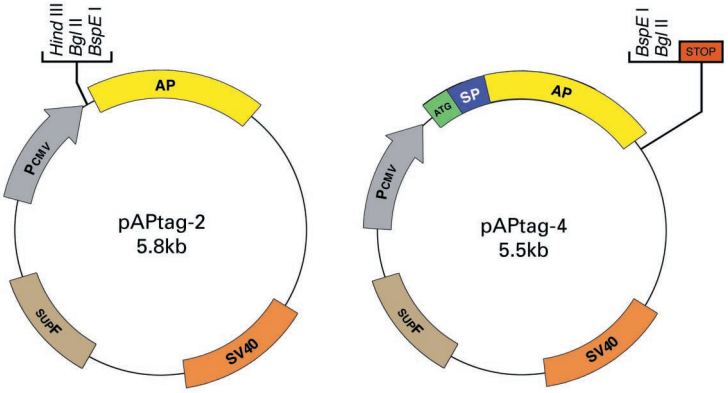
GenHunter Products: Expression Cloning Kit
AP Assay Reagent S
Kit-AP AP-body™
Kit Ligand Positive Control Vector
pMT21 Expression Vector

AP-TAG® Kit A

For Academic/Non-Profit: Cat. No.: Q201 Price: $935
For Industry: Cat. No.: Q201P Price: $3240

For non-radioactive detection of receptor/ligand interaction

This is the second generation of AP-TAG® technology. A secreted ligand or soluble receptor can be fused with secreted alkaline phosphatase (AP) at either its N- or C-terminus to produce an "AP-body™". The resulting AP fusion protein can be expressed as a secreted protein and used directly as highly sensitive affinity agents much like an antibody.



Features	pAPtag-2	pAPtag-4
Size (kb)	5.8	5.5
AP fusion Type	Ligand-AP (Receptor-AP)	AP-Ligand (AP-Receptor)
Cloning Sites	*Hind* III, *Bgl* II, *BspE* I	*Bgl* II, *BspE* I
Promoter	CMV	CMV
SV40 Ori.	Yes	Yes
E. coli Host	GH2/P3	GH2/P3
Vector selection	Tet/Amp	Tet/Amp
Secretion Signal	From insert	From AP
AP negative Control (secretes AP alone)	No	Yes

AP-TAG® Kit A

1. pAPtag-2 (10 µg)	40 µL
2. pAPtag-4 (10 µg)	40 µL
3. L-AP Primer (2 µM)	100 µL
4. R-AP Primer (2 µM)	100 µL
5. Colony lysis buffer	2 X 1 mL

NOTE: pAPtag-2 and pAPtag-4 plasmid vectors can only be transformed or propagated in E. coli host cells with P3 episome such as **GH2/P3 Supercompetent cells (Cat. No. T601)**.

The L(left)- and R(right)-AP primers flanking the cloning sites of pAPtag-2 are used in PCR to check for the presence and size of DNA insert cloned into the vector and for sequence verification of the Ligand-AP or soluble receptor-AP fusion constructs. The L-AP4 and R-AP4 primers (not included in kit) flank the cloning sites of pAPtag-4 and can be purchased separately (see below).

This kit is shipped on dry ice via overnight delivery.
A detailed step-by-step protocol is included.

References:

1. Flanagan, J. G. and Leder, P. (1990). The kit ligand: A cell surface molecule altered in steel mutant fibroblasts. ***Cell*** 63, 185-194.
2. Cheng, H.J., and Flanagan, J.G. (1994). Identification and cloning of ELF-1, a developmentally expressed ligand for Mek4 and Sek receptor tyrosine kinases. ***Cell*** 79, 157-168.
3. Tartaglia, L.A. et al. (1995). Identification and expression cloning of a leptin receptor, OB-R. ***Cell*** 83, 1263-1271.
4. He, Z. and Tessier-Lavigne, M. (1997). Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. ***Cell*** 90, 739-751.
5. Flanagan, J.G., et al. (2000). Alkaline phosphatase fusions of ligands or receptors as *in situ* probes for staining of cells, tissues and embryos. ***Methods in Enzymology*** 327, 17-35.
6. Flanagan, J.G., and Cheng, H.-J. (2000). Alkaline phosphatase fusion proteins for molecular characterization and cloning of ligands and receptors. ***Methods in Enzymology*** 327, 198-210.
7. US patents 5,554,499 and 5,801,000.

See page 59 for an extensive list of AP-TAG® References.

Individual components for the AP-TAG® Kit A sold separately:

Description	Volume	Cat. No. Academic	Cat. No. Industry	Price Academic	Price Industry
pAPtag-2 (10 µg)	40 µL	QV2	QV2P	$920	$3225
pAPtag-4 (10 µg)	40 µL	QV4	QV4P	$920	$3225
L-AP Primer (2 µM)	100 µL	Q210	Q210	$53	$53
R-AP Primer (2 µM)	100 µL	Q211	Q211	$53	$53
L-AP4 Primer (2 µM)	100 µL	Q213	Q213	$53	$53
R-AP4 Primer (2 µM)	100 µL	Q214	Q214	$53	$53
Colony Lysis Buffer	5 mL	L102	L102	$57	$57

AP-TAG® Kit B

For Academic/Non-Profit: Cat. No.: Q202 Price: $935
For Industry: Cat. No.: Q202P Price: $3240

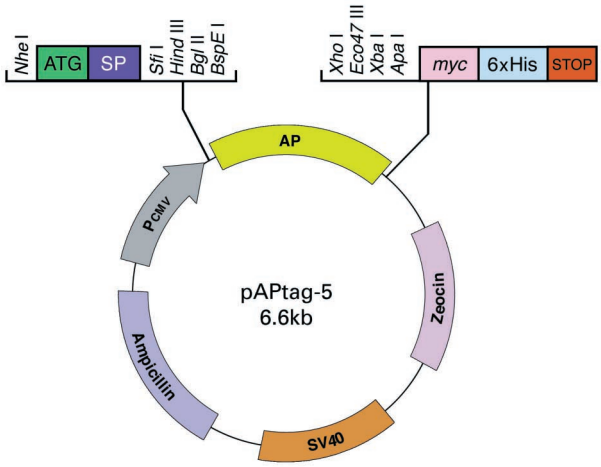
For non-radioactive detection of receptor/ligand interaction

This single vector system is the third generation AP-TAG® technology. A secreted ligand or soluble receptor can be fused with secreted alkaline phosphatase (AP) at either its N- or C-terminus to produce an "AP-body™". The resulting AP fusion protein can be expressed as a secreted protein and used directly as highly sensitive affinity agents much like an antibody. The epitope tags (6xHis and *myc*) allow easy purification, detection and interaction assays (IP) of the AP-fusion proteins. Other improved features are listed below.

Features	pAPtag-5
Size (kb)	6.6
AP fusion Type	Ligand-AP or AP-Ligand (Receptor-AP or AP-Receptor)
Cloning Sites	*Sfi* I, *Hind* III, *Bgl* II, *BspE* I or *Xho* I, *Eco47* III, *Xba* I *Apa* I
Promoter	CMV
SV40 Ori.	Yes
E. coli Host	GH (no need for P3)
Vector Selection	Ampicillin
Secretion Signal	From vector or insert
AP negative Control (secretes AP alone)	Yes
Affinity Tags	6xHis and *myc*
Transfection marker	Zeocin (Invitrogen)

AP-TAG® Kit B

1. pAPtag-5 (20 µg)	40 µL
2. L-AP5 Primer (2 µM)	100 µL
3. R-AP Primer (2 µM)	100 µL
4. Colony lysis buffer	2 X 1 mL



NOTE: pAPtag-5 plasmid vector can be transformed or propagated in **GH Competent cells (Cat. No. L301)**.

The L(left)-AP5 and R(right)-AP primers flank the N-terminal cloning site of pAPtag-5 and are used to check for the presence and size of DNA insert cloned into the vector and for sequence verification of the Ligand-AP or soluble receptor-AP fusion constructs (N-terminal of AP only). For C-terminal cloning, the L-AP5C and R-AP5C primers (not included in the kit) can be used (see below).

This kit is shipped on dry ice via overnight delivery.
A detailed step-by-step protocol is included.

References:

1. Flanagan, J.G., et al. (2000). Alkaline phosphatase fusions of ligands or receptors as *in situ* probes for staining of cells, tissues and embryos. ***Methods in Enzymology*** 327, 17-35.
2. Flanagan, J.G., and Cheng, H.-J. (2000). Alkaline phosphatase fusion proteins for molecular characterization and cloning of ligands and receptors. ***Methods in Enzymology*** 327, 198-210.
3. US patents 5,554,499 and 5,801,000.

See page 59 for an extensive list of AP-TAG® References.

Individual components for the AP-TAG® Kit B sold separately:

Description	Volume	Cat. No. Academic	Cat. No. Industry	Price Academic	Price Industry
pAPtag-5 (20 µg)	40 µL	QV5	QV5P	$920	$3225
L-AP5 Primer (2 µM)	100 µL	Q212	Q212	$53	$53
R-AP Primer (2 µM)	100 µL	Q211	Q211	$53	$53
L-AP5C Primer (2 µM)	100 µL	Q215	Q215	$53	$53
R-AP5C Primer (2 µM)	100 µL	Q216	Q216	$53	$53
Colony Lysis Buffer	5 mL	L102	L102	$57	$57

GH2/P3 Supercompetent Cells

CAT. NO.: T601 **PRICE: $200**

For transformation of pAPtag-2 and pAPtag-4 vectors

The pAPtag-2 and pAPtag-4 AP-fusion cloning vectors contain the supF gene which confers both ampicillin and tetracycline resistance when transformed into the GH2/P3 Supercompetent Cells. pAPtag-2 and pAPtag-4 vectors will not confer antibiotic resistance in an *E. coli* host which does not contain the P3 episome. A tube of 1000X AT antibiotics mix is included for your convenience to prepare ampicillin (25 µg/mL) and tetracycline (10 µg/mL) plates for 1 L of LB-agar.

Detailed protocol included.

GH2/P3 Supercompetent Cells

1. GH2/P3 Supercompetent Cells	5 x 0.4 mL
2. 1000X AT Antibiotics Mix (Amp and Tet)	1 mL

AT Antibiotics Mix (1000X)

CAT. NO.: Q601 **SIZE: 1 mL** **PRICE: $41**

For selection of pAPtag-2 and pAPtag-4 plasmids

Each tube of AT antibiotics mix is conveniently packaged to prepare ampicillin (25 µg/mL) and tetracycline (10 µg/mL) plates for 1 L of LB-agar.

GH Competent Cells

CAT. NO.: L301 **SIZE: 6 X 0.5 mL** **PRICE: $200**

For transformation of pAPtag-5 vector

The pAPtag-5 AP fusion cloning vector contains the ampicillin resistance gene. It can be easily and efficiently transformed and propagated in GH Competent cells.

293T Cells

Cat. No.: Q401 **Size: 5 x 10^6 Cells / Vial** **Price: $215**

For transfection with pAPtag vectors

293T is a human embryonic kidney (HEK) cell line commonly used for transfection assays. Due to the expression of the large T antigen in the cells, plasmids with SV40 origin of replication (such as pAPtag-2, pAPtag-4, and pAPtag-5) can be transiently transfected and give extremely high levels of expression of AP fusion proteins (e.g. ligand-AP fusion proteins). Thus, we strongly recommend using this cell line for your production of AP fusion proteins with pAPtag vectors. The fusion proteins can be easily monitored 2-3 days after transfection by alkaline phosphatase assay (See our AP Assay Reagent A, Cat. No. Q501) or by Western blot using AP Antibody. But for long term production of AP fusion proteins, we recommend that a stable cell line be cloned by co-transfecting with a puromycin or hygromycin-resistant plasmid (293T is G418 resistant). See below for information on the co-transfection vectors we offer.

Detailed protocol included.

293T-S Cells (for Serum Free)

Cat. No.: Q401-S **Size: 5 x 10^6 Cells / Vial** **Price: $289**

For transfection with pAPtag vectors for serum free production

The 293T-S cell line is a clone of the same HEK 293T cell line (above), but has it has already been adapted for use in serum-free (SFM) production. They can be used with the pAPtag-2, pAPtag-4, and pAPtag-5 vectors as well. Just like the standard 293T cells, the fusion proteins can be easily monitored after transfection by alkaline phosphatase assay or by Western blot using AP Antibody. But for long term production of AP fusion proteins, we recommend that a stable cell line be cloned using a co-transfection vectors (see below).

Detailed protocol included.

Co-transfection Vectors

For use as a selectable marker for transfection of cultured mammalian cells.

The pSV2-Hygro or pBabe-Hygro vectors confer hygromycin resistance and the pBabe-Puro vector confers puromycin resistance when co-transfected into cells.

Vector	Selectable Marker	Cat. No.	Volume	Price
pSV2-Hygro co-transfection vector	Hygromycin	Q455	stab	$156
pBabe-Hygro co-transfection vector	Hygromycin	Q455-B	stab	$156
pBabe-Puro co-transfection vector	Puromycin	Q456	10 µg	$156

293T/pAPtag-4 Stable Cell Line

Cat. No.: Q402 **Size: 5 x 10^6 Cells / Vial** **Price: $281**

For production of high levels of AP alone

The 293T/pAPtag-4 stable cell line is used to produce high levels of secreted human placental alkaline phosphatase (AP) which can be used as a negative control for a ligand-AP or soluble receptor-AP fusion protein in cell surface binding assays or cell staining. High level production of secreted AP can be achieved with sub-confluent to confluent culture a few days after medium change. The secretion of AP can be monitored easily with the culture medium by AP activity assay using GenHunter AP Assay Reagent A (Cat. No. Q501).

Detailed protocol included.

AP Western Blot Kit

Cat. No.: Q310 **Price: $395**

For immunoblotting of AP fusion proteins

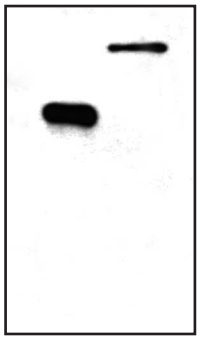
This kit contains the AP Antibody (rabbit polyclonal) which is specific to human placental secreted AP as well as two controls for AP Western blots. The antibody works optimally when used for Western blot analysis of secreted AP fusion proteins from culture media. It should be noted that although this antibody works extremely well for Western blot detection of AP fusion proteins, AP itself (with a MW of 67 KD) may not be detected directly from the culture media due to the amount of albumin which runs at a similar MW. Therefore, the purified AP from human placenta and a known soluble receptor AP fusion protein are provided as positive controls for the antibody. In addition, this antibody only recognizes the denatured form of AP. The Monoclonal AP Antibody (Cat. # Q320) can be used for applications where recognition of the native form is required.

Detection Limit: 20 mU of AP

AP Western Blot Kit

1. AP Antibody (human placenta) - Polyclonal From rabbit	100 µL
2. Purified AP (Western blot control) human placenta, 1 unit/mL	100 µL
3. Kit-AP fusion protein control media 1unit/mL	200 µL

This kit is shipped on dry ice via overnight delivery.
A detailed step-by-step protocol is included.



— **Kit-AP (150 KD)**

— **AP (67 KD)**

Western blot analysis of AP fusion proteins (antibody dilution 1:2000)

Individual components for the AP Western Blot Kit sold separately:

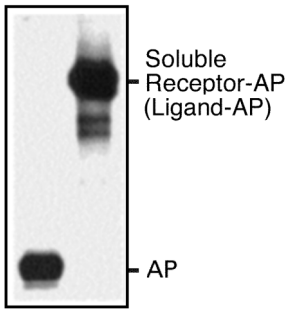
Description	Cat. No.	Volume	Price
AP Antibody (human placenta) - Polyclonal	Q301	100 µL	$281
Purified AP (Western blot control)	Q302	100 µL	$70
Kit-AP fusion protein media (positive control)	Q303	200 µL	$70

AP Antibody (human placenta) - Monoclonal

For ELISA Assay of Alkaline Phosphatase or IP (Immunoprecipitation)

This Monoclonal AP Antibody (human placental) is purified IgG 2a with a concentration of approximately 2.3 mg/mL. It is purified by DEAE chromatography and is in 15mM potassium phosphate buffer, 150mM sodium chloride, 0.1% sodium azide, pH 7.2. This antibody does not recognize denatured AP and therefore cannot be used for Western Blot directly.

	Cat. No.	Volume	Price
AP Antibody (human placenta) - Monoclonal	Q320	100 µg	$308
AP Antibody (human placenta) - Monoclonal	Q320-5	500 µg	$936
AP Antibody (human placenta) - Monoclonal	Q321	1 mg	$1520



IP with Monoclonal AP Antibody

Monoclonal AP Antibody-Sepharose Beads

For one-step purification of AP-fusion proteins and proteins interacting with AP-fusion proteins

Applications:

1) Concentrating AP fusion protein from the conditioned media
2) Purification of AP fusion proteins
3) Affinity column purification of protein(s) or molecules interacting with the ligand/receptor-AP fusion protein
4) IP Western analysis of the AP fusion proteins (see Polyclonal AP Antibody)

Technical parameters:

Coupling condition: 1 mg of pure IgG/mL of beads

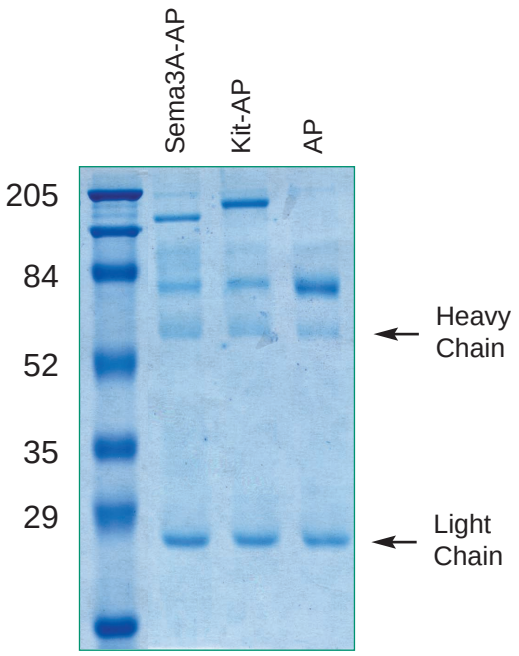
AP binding capacity: up to 200 Units/mL of beads

Specificity of the antibody: Native human placental AP

Interference of AP activity: The antibody binding does not interfere with the AP activity

Product Form:

The Monoclonal AP antibody-sepharose beads are supplied in a 50% suspension in 0.1 M $NaHCO_3$, 0.5 M NaCl, pH 8.4.



Coomassie blue stain of different secreted AP-fusion proteins purified with our Monoclonal AP Antibody-Sepharose Beads.

	Cat. No.	Total Volume	Price
AP Antibody-Sepharose Beads	Q330	100 µL	$308
AP Antibody-Sepharose Beads	Q331	500 µL	$1190
AP Antibody-Sepharose Beads	Q332	1 mL	$1741

Antigen Elution Solution

For eluting AP fusion protein from the Monoclonal AP Antibody-Sepharose Beads

The Antigen Elution Solution is specially formulated for eluting AP or AP fusion proteins from the monoclonal AP Antibody-sepharose beads. This solution breaks the extremely tight interaction between the antibody-antigen complex, allowing up to 80% recovery of the bound antigen.

It is available in both Acidic and Basic versions, depending on the pH sensitivity of your protein.

	Cat. No.	Volume	Price
Antigen Elution Solution (Acidic)	Q340A	10 mL	$43
Antigen Elution Solution (Acidic)	Q341A	50 mL	$106
Antigen Elution Solution (Basic)	Q340B	10 mL	$43
Antigen Elution Solution (Basic)	Q341B	50 mL	$106

Expression Cloning Kit

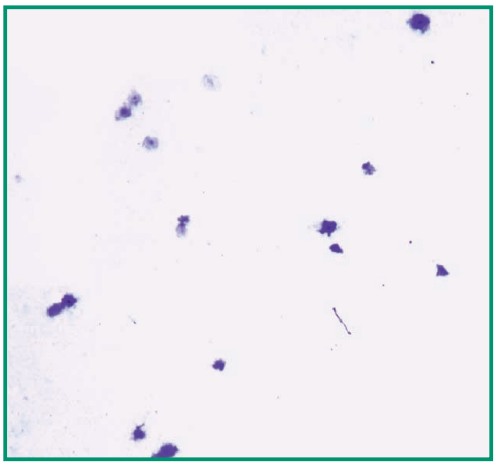
CAT. NO.: Q450 **PRICE: $413**

For expression cloning of cell surface receptor/ligand using AP fusion proteins (AP-bodies™)

This kit consists of a clonally purified cos-1 host cell line ideal for expression cloning by panning and a positive control receptor/ligand-AP pair.

An expression cDNA library potentially containing the receptor/ligand gene of interest can be transiently transfected into these cells. Positive cDNA pools can be identified by staining the transfected cells with your AP fusion protein. Cells over-expressing the corresponding cell surface receptor/ligand will be stained blue with GenHunter AP Activity Assay Reagent S (see figure below).

The Kit ligand positive control vector [containing a 1 kb cDNA encoding the transmembrane form of Kit ligand (stem cell factor), Ref. # 1 below] can be transfected into the cos-1 host cell line. Cells overexpressing cell surface Kit ligand will be stained blue by soluble receptor Kit-AP fusion protein using GenHunter AP Assay Reagent S.



Expression cloning of cell surface receptor/ligand by panning

Expression Cloning Kit

1. cos-1 Host Cell Line	1 x 10^6 cells / vial
2. Kit-AP fusion protein (media) 1 unit/mL	10 mL
3. Kit ligand (stem cell factor) positive control vector	10 µg

This kit is shipped on dry ice via overnight delivery.
A detailed step-by-step protocol is included.

References:

1. Flanagan, J.G. *et al*. (1991). Transmembrane Form of the *kit* Ligand Growth Factor is Determined by Alternative Splicing and is missing in the SI^d Mutant. ***Cell*** 64, 1025-1035.
2. Cheng, H.J., and Flanagan, J.G. (1994). Identification and cloning of ELF-1, a developmentally expressed ligand for Mek4 and Sek receptor tyrosine kinases. ***Cell*** 79, 157-168.
3. Tartaglia, L.A. *et al*. (1995). Identification and expression cloning of a leptin receptor, OB-R. ***Cell*** 83, 1263-1271.
4. He, Z. and Tessier-Lavigne, M. (1997). Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. ***Cell*** 90, 739-751.

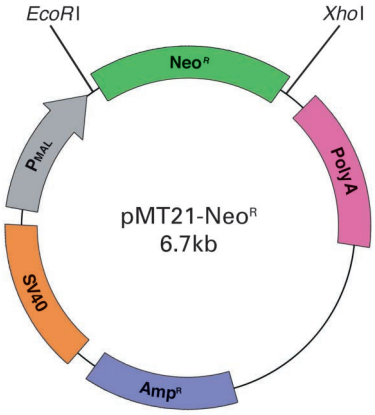
Individual components for the Expression Cloning Kit sold separately:

DESCRIPTION	CAT. NO.	VOLUME	PRICE
cos-1 Host Cell Line	Q451	1 x 10^6 cells/vial	$185
Kit-AP fusion protein (media), 1 unit/mL	Q452	10 mL	$102
Kit ligand (stem cell factor) positive control vector	Q453	10 µg	$192

pMT21-Neo Mammalian Expression Cloning Vector

CAT. NO.: Q454 **PRICE: $432**

For construction of expression cDNA libraries.



This cloning vector has been used extensively to construct mammalian expression cDNA libraries (see below references). The vector contains an SV40 origin of replication and the major adeno late promoter (P_{MAL}) in front of Neo resistance cDNA insert flanked by an *EcoR* I and *Xho* I site. Using the Stratagene cDNA Synthesis Kits generally results in cDNA ends with *EcoR* I and *Xho* I sites, which can be directionally cloned into the pMT21-Neo^R vector.

Amount:	20 µg
Cloning Sites:	*EcoR* I, *Xho* I (the excised Neo^R insert is 2kb)
Promoter:	Major Adeno Late Promoter (P_{MAL})
Antibiotic Resistance:	Ampicillin

References:

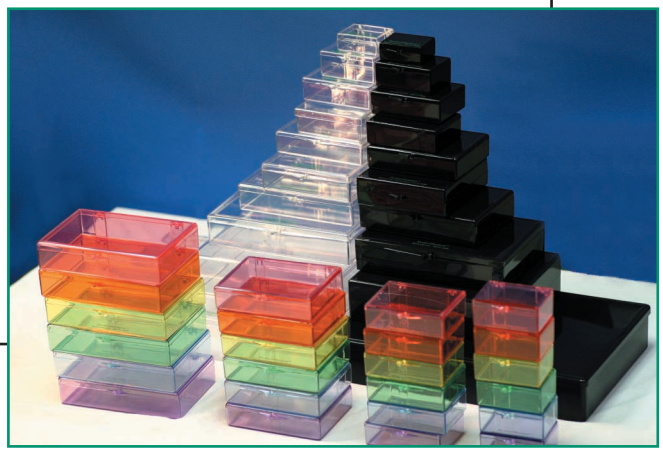
1. He, Z. and Tessier-Lavigne, M. (1997). Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. ***Cell*** 90, 739-751.

2. Kolodkin, A.L. *et al.* (1997). Neuropilin is a Semaphorin III receptor. ***Cell*** 90, 753-762.

Want to save money on Antibodies and get a PerfectWestern®?

See page 56 for information on our PerfectWestern® Containers!!

These containers come in 23 different sizes to fit different size membranes. This allows you to save money on expensive antibodies because significantly less volume can be used.



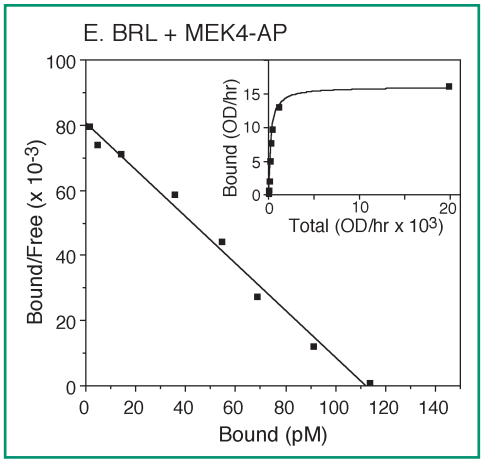
AP Assay Reagent A (For 200 Reactions)

Cat. No.: Q501 **Size: 10 mL** **Price: $122**

For AP Activity Assay

The AP assay reagent A is formulated specifically for measuring the enzymatic activity of the alkaline phosphatase (AP). The secretion of human placenta AP or a secreted ligand(receptor)-AP fusion protein can be easily monitored by this assay using the culture media in which the transfected cells are grown. The dephosphorylation of the p-Nitrophenyl phosphate by AP leads to the generation of yellow color which serves as both qualitative (by eye) and quantitative (Absorbance 405 nm) measurement of AP activity.

Detailed protocol included.



Receptor/Ligand Binding Assay using AP Assay Reagent A

Reference:

1. Flanagan, J. G. and Leder, P. (1990). The kit Ligand: A cell surface molecule altered in steel mutant fibroblasts. ***Cell*** 63, 185-194.

AP Assay Reagent S

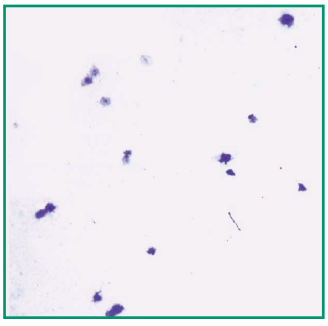
Cat. No.: Q502 **Size: 10 mL** **Price: $49**
Cat. No.: Q502L **Size: 100 mL** **Price: $342**

For Cell Staining

The AP Assay Reagent S is formulated specifically for cell staining or affinity blotting analysis of enzymatic activity of the alkaline phosphatase (AP). The AP substrate BCIP upon dephosphorylation forms an insoluble blue precipitate, thus it can be used for tissue or cell staining for the presence of receptors to which the ligand-AP fusion proteins bind. This assay kit can also be used during expression cloning of a receptor (Cheng and Flanagan, 1994, Cell 79:157-168).

Detailed protocol included.

***in situ* staining of receptor/ligand** (embryonic chick brain)



Expression cloning of receptor/ligand by panning

Reference:

1. Flanagan, J. G. and Leder, P. (1990). The kit Ligand: A cell surface molecule altered in steel mutant fibroblasts. ***Cell*** 63, 185-194.

HBHA Wash Buffer

For Receptor Binding Assay

HBHA wash buffer consists of Hank's balanced salt solution with 0.5 mg/mL BSA and 20 mM HEPES, pH 7.0. This buffer has been used extensively for ligand-receptor binding assays.

Detailed protocol included.

	Cat. No.	Volume	Price
HBHA Wash Buffer	Q503S	100 mL	$54
HBHA Wash Buffer	Q503L	500 mL	$194

Reference:

1. Flanagan, J. G. and Leder, P. (1990). The kit Ligand: A cell surface molecule altered in steel mutant fibroblasts. ***Cell*** 63, 185-194.

Cell Lysis Buffer

Cat. No.: Q504 **Size: 100 mL** **Price: $54**

For Cell Lysis in Receptor Binding Assay

The cell lysis buffer is used in ligand-receptor binding assay. This buffer allows rapid lysis of the cells and removal of cell nuclei before bound AP activity is measured.

Detailed protocol included.

Reference:

1. Flanagan, J. G. and Leder, P. (1990). The kit Ligand: A cell surface molecule altered in steel mutant fibroblasts. ***Cell*** 63, 185-194.

PerfectWestern® Containers - *23 Sizes Now Available!*

For optimal Western blot analysis

Save Money on Antibodies - now less volume is required!

Due to the popularity of these containers and based on continuing feedback from our customers, GenHunter has expanded this product line again and now offers 23 different sized containers to fit your needs.

For the most complete information and a real-size layout, please download or request the 21-page 2011 PerfectWestern Brochure at *www.PerfectWestern.com*

PerfectWestern®-Dura: Brand New Material

New, more flexible, proprietary polystyrene blend. Still crystal clear, but is virtually unbreakable. Currently, only available in the Medium size. See details and pricing next page.

These specially designed containers perfectly accommodate Western blots of different size, including whole blots, half blots, and strips. They come in 23 different sizes in both clear and black plastic. Our PerfectWestern® containers significantly reduce the volume of expensive antibodies required while still producing beautiful Western blots!

These containers were conceived by our own frustration in searching for suitable containers for Western blots. The choices were often much too large and never flat enough, causing waste of antibody and concern about scratching the membrane. We decided to design our own containers offering many different sizes and options to truly achieve a PerfectWestern®. Our containers are made of durable polystyrene plastic, with ultra-smooth flat inner surfaces and lids to reduce evaporation. Their 90° edges create excellent membrane movement for more efficient incubation.

All containers come in both clear and black versions. The clear container allows easy visualization of the blot, whereas the black version keeps light-sensitive reagents protected. In addition, 4 of our most popular sizes (Small, Medium, Large, and Extra Large) are available in 6 translucent colors: pink, orange, yellow, green, blue, and violet.

See next page for more information including dimensions, catalog numbers, and prices. **We highly recommend downloading or requesting the complete 2011 PerfectWestern brochure including a real-size layout of all 23 sizes at *www.PerfectWestern.com*.**

Which PerfectWestern® Container fits your Gel System?

Gel Manufacturer	Our Container	Cat. No.
Biometra (formerly from Invitrogen)		
Mini-V 8•10	Medium	B101
V15•17 Vert. Gel. Elect.	Jumbo/Enormo	B138/B111
V16 or V16-2	Jumbo/Enormo	B138/B111
Maxigel	Enormo/Colosso/2D Tray	B111/B140/B142
Bio-Rad Systems		
Mini-PROTEAN 3	Medium	B101
Criterion Precast	Extra Large	B109
PROTEAN II xi, 16cm	Jumbo/Enormo	B138/B111
PROTEAN II xi, 20cm	Enormo	B111
PROTEAN II XL, 20cm	Enormo/Colosso/2D Tray	B111/B140/B142
PROTEAN Plus	Colosso/2D Tray	B140/B142
GE Healthcare / Hoefer (formerly Amersham)		
Mighty Small SE250	Medium/Mini Square	B101/B144
Mighty Small SE260	Mini Square/Large	B144/B107
miniVE Vert. Gel Elect.(SE300)	Large/Square	B107/B119
SE600/SE400 (18 x 8)	Doublewide/Extra Large	B136/B109
SE600/SE400 (18 x 16)	Jumbo/Enormo	B138/B111
SE640 (18 x 8)	Mini Doublewide	B136
SE660 (24 x 18)	Enormo/Colosso/2D Tray	B111/B140/B142
Ettan Dalt II/six/twelve (2D gel)	Colosso/2D Tray	B140/B142
Multiphor II, Excel 2D 12.5	Colosso/2D Tray	B140/B142
Multiphor II, Excel XL 12-14	Colosso/2D Tray	B140/B142
Invitrogen / Novex		
Novex XCell SureLock Mini	Mini Square	B144
Novex XCell SureLock Midi	Extra Large	B109
E-PAGE Gels	Extra Large	B109
Lonza (formerly Cambrex/FMC/BMA)		
PAGEr Gold Precast (8.3 x 7.1)	Medium	B101
PAGEr Gold Precast (8.3 x 8.1)	Mini Square	B144
Owl Systems		
Puffin P81	Large/Square	B107/B119
Penguin P8DS	Large/Square	B107/B119
Penguin P9DS	Jumbo/Enormo	B138/B111
Penguin P10DS	Enormo/Colosso/2D Tray	B111/B140/B142

Important Notes: 1) All above catalog numbers are given for the clear version. If black or colored versions are desired, check on catalog number. **2)** The above recommendations are not guaranteed to be correct. Please check the exact size of gels from your apparatus and compare it to the PerfectWestern® container (inside dimensions are given) to be sure.

New Product Available: The PerfectMembrane™

- **Available in PVDF or Nitrocellulose (NC)**
- **Available in 2 sizes: 8.3 x 6 cm or 8.3 x 7.3 cm**
- **Pre-cut to fit in the "Medium" or "Mini Square" size**
- **Moves well during incubation**
- **Saves time and materials**

Size	Membranes	Price	PVDF Cat. #	NC Cat. #
8.3 x 6 cm	10	$44	B301-10	B311-10
8.3 x 6 cm	25	$97	B301-25	B311-25
8.3 x 6 cm	50	$166	B301-50	B311-50
8.3 x 7.3 cm	10	$53	B321-10	B331-10
8.3 x 7.3 cm	25	$116	B321-25	B331-25
8.3 x 7.3 cm	50	$199	B321-50	B331-50

23 PerfectWestern® Container Sizes Now Available

Download/Request the complete 2011 PerfectWestern brochure at www.PerfectWestern.com

Standard Rectangular Type Containers

Extra Small - For Portions of Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Extra Small, Clear	B121	$13	$10	$8	$7	$6.50
Extra Small, Black	B122	$13	$10	$8	$7	$6.50

Dimensions (LxWxH): 5.3 x 4.0 x 2.2 cm • 2 1/16 x 1 9/16 x 7/8 inches

Small - For Mini-Protein gels cut in half

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Small, Clear	B104	$14	$11	$9	$8	$7.50
Small, Black	B105	$14	$11	$9	$8	$7.50
Small, one specific color (below*)	B104?*	$15	$12	$10	$9	$8.50
Small Rainbow Set (all 6 colors)	B104S	$82	$62	$48	$43	$40

*6 Colors: Pink (B104P), Orange (B104O), Yellow (B104Y), Green (B104G), Blue (B104B), Violet (B104V)

Dimensions (LxWxH): 7.3 x 5.1 x 3.3 cm • 2 7/8 x 2 x 1 5/16 inches

NEW HEIGHT

New "Thin" Version For Small Size! Save Space; ~ 40% Shorter

Small-Thin, Clear	B104-T	$14	$12	$9	$8	$7.50
Small-Thin, Black	B104-T	$14	$12	$9	$8	$7.50

Dimensions (LxWxH): 7.3 x 5.1 x 1.9 cm • 2 7/8 x 2 x 3/4 inches

Medium - For Whole Mini-Protein gels — MOST POPULAR SIZE

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Medium, Clear	B101	$15	$12	$10	$9	$8.50
Medium, Clear Lid/Black Base	B102	$15	$12	$10	$9	$8.50
Medium, Black	B103	$15	$12	$10	$9	$8.50
Medium, one specific color (below*)	B101?*	$16	$13	$11	$10	$9.50
Medium Rainbow Set (all 6 colors)	B101S	$86	$66	$51	$46	$43

*6 Colors: Pink (B101P), Orange (B101O), Yellow (B101Y), Green (B101G), Blue (B101B), Violet (B101V)

Dimensions (LxWxH): 9.1 x 6.6 x 2.9 cm • 3 5/8 x 2 5/8 x 1 1/8 inches

"Thin" Version For Medium Size! Save Space; ~ 2/3 The Height.

Medium-Thin, Clear	B101-T	$15	$12	$10	$9	$8.50
Medium-Thin, Black	B103-T	$15	$12	$10	$9	$8.50

Dimensions (LxWxH): 9.1 x 6.6 x 1.9 cm • 3 5/8 x 2 5/8 x 3/4 inches

NEW MATERIAL

NEW "Dura" Material for "Medium" size: Virtually Unbreakable!

More flexible, proprietary polystyrene blend. Still crystal clear, but will last much longer.

PerfectWestern-Dura, Medium, Clear	B101-D	$19	$16	$14	$13	$12.50
PerfectWestern-Dura, Medium, Black	B103-D	$19	$16	$14	$13	$12.50

Dimensions (LxWxH): 9.1 x 6.6 x 3.2 cm • 3 5/8 x 2 5/8 x 1 1/4 inches

Mini Square - For Larger Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Mini Square, Clear	B144	$20	$15	$13	$12	$11
Mini Square, Black	B145	$20	$15	$13	$12	$11

Dimensions (LxWxH): 8.6 x 8.6 x 3.3 cm • 3 13/32 x 3 13/32 x 1 1/4 inches

Large - For Larger Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Large, Clear	B107	$21	$16	$14	$13	$12
Large, Black	B108	$21	$16	$14	$13	$12
Large, one specific color (below*)	B107?*	$22	$17	$15	$14	$13
Large Rainbow Set (all 6 colors)	B107S	$120	$86	$72	$66	$63

*6 Colors: Pink (B107P), Orange (B107O), Yellow (B107Y), Green (B107G), Blue (B107B), Violet (B107V)

Dimensions (LxWxH): 11.7 x 9.0 x 2.9 cm • 4 5/8 x 3 9/16 x 1 1/8 inches

Square - For Larger Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Square, Clear	B119	$25	$21	$18	$16	$15
Square, Black	B120	$25	$21	$18	$16	$15

Dimensions (LxWxH): 11.7 x 11.7 x 3.5 cm • 4 9/16 x 4 9/16 x 1 3/8 inches

Extra Large - For Larger Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Extra Large, Clear	B109	$25	$21	$18	$16	$15
Extra Large, Black	B110	$25	$21	$18	$16	$15
Extra Large, one color (below*)	B109?*	$26	$22	$19	$17	$16
Extra Large Rainbow Set (all 6 colors)	B109S	$140	$106	$88	$81	$76

*6 Colors: Pink (B109P), Orange (B109O), Yellow (B109Y), Green (B109G), Blue (B109B), Violet (B109V)

Dimensions (LxWxH): 15.4 x 10.4 x 3.6 cm • 6 1/16 x 4 1/16 x 1 1/2 inches

Jumbo - For Very Large Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Jumbo, Clear	B138	$27	$23	$19	$18	$17
Jumbo, Black	B139	$27	$23	$19	$18	$17

Dimensions (LxWxH): 18.8 x 12.5 x 4.1 cm • 7 7/16 x 4 15/16 x 1 5/8 inches

Enormo - For Very Large Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Enormo, Clear	B111	$29	$24	$21	$19	$18
Enormo, Black	B112	$29	$24	$21	$19	$18

Dimensions (LxWxH): 20.7 x 15.9 x 5.2 cm • 8 1/8 x 6 1/4 x 2 1/16 inches

Largest Sizes For Staining 2D Gels

Colosso - For Largest Protein gels (2D gels, etc.)

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Colosso, Clear	B140	$62	$51	$44	$41	$38
Colosso, Black	B141	$82	$71	$64	$61	$58

Dimensions (LxWxH) Clear version: 32.1 x 21.3 x 5.7 cm • 12 5/8 x 8 3/8 x 2 1/4 inches
Black version: 31.7 x 21.0 x 5.6 cm • 12 7/16 x 8 9/32 x 2 3/16 inches

NEW SIZE

2D Gel Staining Tray - For Largest Protein gels (2D gels, etc.)

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
2D Gel Staining Tray, Clear	B142	$62	$51	$44	$41	$38
2D Gel Staining Tray, Black	B143	$62	$51	$44	$41	$38

Dimensions (LxWxH): 29.3 x 23 x 7.7 cm • 11 1/2 x 9 x 3 1/16 inches
Notes: The bottom of the container is not as flat as all other PerfectWesterns, so more volume may be required.

Compartmented Containers For Strips

3-Sectional - For Thin Strips from Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
3-Sectional, Clear	B106	$20	$15	$12	$11	$10
3-Sectional, Black	B125	$20	$15	$12	$11	$10

Well Dimensions (LxWxH): 3 wells each 7.3 x 1.6 x 1.3 cm • 2 7/8 x 5/8 x 1/2 inches
Overall Dimensions (LxW): 7.3 x 5.1 cm • 2 7/8 x 2 inches
Height: Clear is 1.6 cm, 5/8 inches. Black is 2.5 cm, 1 inch.

6-Sectional-Short - For Strips from Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
6-Sectional-Short, Clear	B130	$46	$39	$32	$29	$27
6-Sectional-Short, Black	B131	$56	$49	$42	$39	$37

Note: Each of the 6 compartments is of a size between the Small Strip and Small Strip EXT, but is priced to be significantly cheaper than purchasing 6 of those containers.

Well Dimensions (LxWxH): 6 wells each 8.4 x 2.7 x 3.2 cm • 3 5/16 x 1 1/16 x 1 1/4 inches
Overall Dimensions (LxWxH): 17.3 x 8.4 x 3.2 cm • 6 13/16 x 3 5/16 x 1 1/4 inches

6-Sectional-Mid - For Strips from Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
6-Sectional-Mid, Clear	B132	$51	$43	$36	$34	$30
6-Sectional-Mid, Black	B133	$61	$53	$46	$44	$40

Note: Each of the 6 compartments is almost identical to the Small Strip EXT, but is priced to be significantly cheaper than purchasing 6 of those containers.

Well Dimensions (LxWxH): 6 wells each 10 x 3.2 x 3.3 cm • 3 15/16 x 1 9/32 x 1 3/8 inches
Overall Dimensions (LxWxH): 20.2 x 10 x 3.3 cm • 8 x 3 15/16 x 1 3/8 inches

6-Sectional-Long - For Longer Strips from Mini-Protein gels

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
6-Sectional-Long, Clear	B134	$58	$47	$41	$38	$36
6-Sectional-Long, Black	B135	$68	$57	$51	$48	$46

Note: Each of the 6 compartments is slightly shorter and wider than the Long Strip, but is priced to be significantly cheaper than purchasing 6 of those containers.

Well Dimensions (LxWxH): 6 wells each 15.8 x 4 x 4.4 cm • 6 3/16 x 1 9/16 x 1 3/4 inches
Overall Dimensions (LxWxH): 26.7 x 15.8 x 4.4 cm • 10 1/2 x 6 3/16 x 1 3/4 inches

Non-Compartmented Containers For Strips

Item	Cat. No.	1-4	5-9	10-19	20-49	50+
Small Strip, Clear	B113	$13	$10	$8	$7	$6.50
Small Strip, Black	B114	$13	$10	$8	$7	$6.50
Dimensions (LxWxH): 7.4 x 3.1 x 1.9 cm • 2 7/8 x 1 1/4 x 3/4 inches						
Note: The 6-Sectional-Short (B130, above) container is similar to 6 Small Strips with one lid.						
Small Strip EXT, Clear	B123	$14	$11	$9	$8	$7.50
Small Strip EXT, Black	B124	$14	$11	$9	$8	$7.50
Dimensions (LxWxH): 9.5 x 3.0 x 1.9 cm • 3 7/8 x 1 3/16 x 3/4 inches						
Note: The 6-Sectional-Mid (B132, above) container is similar to 6 Small Strip EXT's with one lid.						
Slimmer, Clear	B128	$16	$13	$11	$10	$9.50
Slimmer, Black	B129	$16	$13	$11	$10	$9.50
Dimensions (LxWxH): 14.3 x 1.4 x 1.7 cm • 5 9/16 x 9/16 x 11/16 inches						
NEW Slim, Clear	B146	$17	$14	$12	$11	$10.50
Slim, Black	B147	$17	$14	$12	$11	$10.50
Dimensions (LxWxH): 15.0 x 1.85 x 1.9 cm • 5 15/16 x 11/16 x 3/4 inches						
Medium Strip, Clear	B115	$15	$12	$10	$9	$8.50
Medium Strip, Black	B116	$15	$12	$10	$9	$8.50
Dimensions (LxWxH): 11.1 x 5.2 x 2.6 cm • 4 3/8 x 2 1/16 x 1 inches						
Large Strip, Clear	B117	$22	$17	$15	$14	$13
Large Strip, Black	B118	$22	$17	$15	$14	$13
Dimensions (LxWxH): 14.7 x 6.7 x 3.6 cm • 5 3/4 x 2 5/8 x 1 7/16 inches						
Mini Doublewide, Clear	B136	$25	$21	$18	$16	$15
Mini Doublewide, Black	B137	$25	$21	$18	$16	$15
Dimensions (LxWxH): 17.8 x 8.9 x 3 cm • 7 x 3 9/16 x 1 1/8 inches						
Long Strip, Clear	B126	$18	$14	$11	$10	$9.50
Long Strip, Black	B127	$18	$14	$11	$10	$9.50
Dimensions (LxWxH): 19.0 x 3.8 x 2.2 cm • 7 1/2 x 1 1/2 x 13/16 inches						
Note: The 6-Sectional-Long (B134, above) container is similar to 6 Long Strips with one lid.						

All dimensions given for the PerfectWestern® containers are container inside dimensions.

Prices listed are only for the U.S., Canada, & other countries without distributors and are subject to change without notice! Find your distributor on page 92 or at www.GenHunter.com/distributors

Premade AP-body™ Collection

As the world-wide exclusive licensee for the AP-TAG® technology reagent business, GenHunter Corporation is pleased to continue its effort to build up a collection of AP-bodies™ (ligand or soluble receptor-AP fusion proteins) made with AP-TAG® technology and make them available to the biomedical research community. If you have already made and/or published an AP fusion construct to your ligand or soluble receptor gene of interest, please consider depositing your vector constructs and the stable production cell lines with GenHunter. In return, GenHunter will provide you or your lab with 6% royalty on sales of any products associated with your deposition. Please contact GenHunter for details regarding such depositions (800-311-8260, Fax: 615-832-9461, or email: info@genhunter.com).

Please be reminded that, according to our AP-TAG® licensing agreement with Harvard, even if you obtained the APtag vectors from Harvard prior to March 1, 1999, when our exclusive sublicensing agreement went into effect, you do not have the right to distribute the APtag vectors or their derivatives such as a ligand-AP fusion construct or cell line harboring such a construct to others without written permission from GenHunter Corporation.

Please consider depositing your AP-body™ with GenHunter, which saves you not only from legal issues regarding AP-TAG® technology, but also hassles in dealing with end-user requests.

AP-bodies™ Now Available:

All AP-bodies™ are supplied as either culture medium or AP-body™ producing plasmid vectors (as bacterial stock). All AP-bodies™ are filter-sterilized, buffered with 20 mM HEPES, pH 7.5, with a minimum amount of AP activity between 0.6 and 1 units/mL, unless otherwise specified.

AP-body™	Receptor/Ligand Recognized	Cat. No.	Volume	Price
AP control	Negative control	Q701	50 mL	$432
AP control, in serum free media	Negative control	Q701-SFM	50 units	$432
Kit-AP	Stem Cell Factor	Q702	50 mL	$432
ELF2-AP	Elk, Cek5, Cek10	Q703	vial	$432
Sema3A-AP	Neuropilin	Q704	vial	$432
mP84-AP	IAP/CD47	Q705	vial	$432
IAP/CD47-AP	mP84	Q706	vial	$432
hIzumo-AP	Unknown	Q707	vial	$432
sTNFR2-AP	TNF-alpha, CD120	Q708	vial	$432
sCD4-AP	gp120, MHC class II molecules	Q709	vial	$432
IL-24-AP (Human)	IL-20R1/IL-20R2 and	Q710	vial	$432
IL-24-AP (Rat)	IL-22R1/IL-20R2 Receptors	Q711	vial	$432
AP-Collagen (Human, C-terminal of alpha1, Type I)	Endo180	Q712	vial	$432
AP-Collagen (Human, C-terminal of alpha1, Type III)	Unknown	Q713	vial	$432
AP-Collagen (Rat, C-terminal of alpha1, Type I)	Endo180	Q714	vial	$432
AP-LRR2-hSlit1	Robo1 and Robo2 Receptors	Q715	vial	$432
AP-LRR2-hSlit2	Robo1 and Robo2 Receptors	Q716	vial	$432

Same receptors for both constructs

AP-TAG® Reference List

1. Flanagan, J. G. and Leder, P. The kit ligand: A cell surface molecule altered in steel mutant fibroblasts. **Cell** 1990, 63:185-194.
2. Flanagan, J.G. et al.: Transmembrane form of the kit ligand growth factor is determined by alternative splicing and is missing in the SId mutant. **Cell** 1991, 64:1025-1035.
3. Morrison, B.W. and Leder, P.: A receptor binding domain of mouse interleukin-4 defined by a solid- phase binding assay and in vitro mutagenesis. **J. Biol. Chem.**, 1992, 267:11957-11963.
4. Zimmer, Y. et al: Multiple structural elements determine ligand binding of fibroblast growth factor receptors. Evidence that both Ig domain 2 and 3 define receptor specificity. **J. Biol. Chem.**, 1993, 268:7899-7903.
5. Cheng, H.J. and Flanagan, J.G.: Identification and cloning of ELF-1, a developmentally expressed ligand for the Mek4 and Sek receptor tyrosine kinases. **Cell** 1994, 79:157-168.
6. Kasahara, N. et al.: Tissue-specific targeting of retroviral vectors through ligand-receptor interactions. **Science** 1994, 266:1373-1376.
7. Aviezer, D. et al: Differential structural requirements of heparin and heparan sulfate proteoglycans that promote binding of basic fibroblast growth factor to its receptor. **J. Biol. Chem.**, 1994, 269:114-121.
8. Tessler, S. et al: Heparin modulates the interaction of VEGF165 with soluble and cell associated flk-1 receptors. **J. Biol. Chem.** 1994, 269:12456-12461.
9. Tzahar, E. et al: ErbB-3 and ErbB-4 function as the respective low and high affinity receptors of all Neu differentiation factor/heregulin isoforms. **J. Biol. Chem.** 1994, 269:25226-25233.
10. Krause, D.S. et al: Characterization of murine CD34, a marker for hematopoietic progenitor and stem cells. **Blood.** 1994, 84:691-701.
11. Ju, S.-T. et al: Fas(CD95)/FasL interactions required for programmed cell death after T-cell activation. **Nature**. 1995, 373:444-448.
12. Cheng H.J. et al.: Complementary gradients in expression and binding of ELF-1 and Mek4 in development of the topographic retinotectal projection map. **Cell** 1995, 82:371-381.
13. Tartaglia, L.A. et al.: Identification and expression cloning of a leptin receptor, OB-R. **Cell.** 1995, 83:1263-1271.
14. Han, X.D. et al: Identification of a unique membrane-bound molecule on a hematopoietic stem-cell line and on multipotent progenitor cells. **Proc. Natl. Acad. Sci. USA.** 1995, 92:11014-11018.
15. Chen, X. et al: Induction of heparin-binding EGF-like growth factor expression during myogenesis. **J. Biol. Chem.** 1995, 270:18285-18294.
16. Bellosta, P. et al.: The receptor tyrosine kinase ark mediates cell-aggregation by hemophilic binding. **Mol. Cell Biol.** 1995, 15:614-625.
17. Bergemann, A.D. et al: ELF-2, a new member of the Eph ligand family, is segmentally expressed in mouse embryos in the region of the hindbrain and newly forming somites. **Mol. Cell. Biol.** 1995, 15:4921-4929.
18. Qin, Z. et al: Human lymphotoxin has at least equal antitumor activity in comparison to human tumor necrosis factor but is less toxic in mice. **Blood**. 1995, 85:2779-2785.
19. Nakamoto, M. et al: Topographically specific effects of ELF-1 on retinal axon guidance *in vitro* and retinal axon mapping in vivo. **Cell**. 1996, 86:755-766.
20. Cao, Y. et al: Heterodimers of placenta growth factor/vascular endothelial growth factor. **J. Biol. Chem.** 1996, 271:3154-3162.
21. Yang, X. and Cepko, C.L.: Flk-1, a receptor for vascular endothelial growth factor (VEGF), is expressed by retinal progenitor cells. **Journal of Neuroscience**. 1996, 16:6089-6099.
22. He, Z.G. and Tessier-Lavigne, M.: Neuropilin is a receptor for the axonal chemorepellent Semaphorin III. **Cell.** 1997, 90:739-751.
23. Kolodkin, A.L. et al.: Neuropilin is a Semaphorin III receptor. **Cell.** 1997, 90:753-762.
24. Koppel, A.M. et al: A 70 amino acid region within the semaphorin domain activates specific cellular response of semaphorin family members. **Neuron**. 1997, 19:531-537.
25. Feiner, L. et al: Secreted chick semaphorins bind recombinant neuropilin with similar affinities but bind different subsets of neurons *in situ*. **Neuron**. 1997, 19:539-545.
26. Chen, H. et al: Neuropilin-2, a novel member of the neuropilin family, is a high affinity receptor for the Semaphorins Sema E and Sema IV but not Sema III. **Neuron**. 1997, 19:547-559.
27. Takahashi, T. et al: Neuronal and non-neuronal collapsin-1 binding sites in developing chick are distinct from other semaphorin binding sites. **Journal of Neuroscience**. 1997, 17:9183-9193.
28. Bonneh-Barkay, D. et al: Identification of glypican as a dual modulator of the biological activity of fibroblast growth factors. **J. Biol. Chem.** 1997, 272:12415-12421.
29. Monschau, B. et al: Shared and distinct functions of RAGS and ELF-1 in guiding retinal axons. **EMBO J.** 1997, 16:1258-1267.
30. Labrador, J.P. et al: The N-terminal globular domain of Eph receptors is sufficient for ligand binding and receptor signaling. **EMBO J.** 1997, 16:3889-3897.
31. Adams, R.H. et al: The chemorepulsive activity of secreted semaphorins is regulated by furin-dependent proteolytic processing. **EMBO J.** 1997, 16:6077-6086.
32. Brennan, C. et al: Two Eph receptor tyrosine kinase ligands control axon growth and may be involved in the creation of the retinotectal map in the zebrafish. **Development.** 1997, 124:655–664.
33. Johnston, S. H. et al. A family of mammalian Fringe genes implicated in boundary determination and the Notch pathway. **Development.** 1997, 124:2245–2254.
34. Polleux, F. et al.: Patterning of cortical efferent projections by semaphorin-neuropilin interactions. **Science.** 1998, 282:1904-1906.
35. Takahashi, T. et al: Semaphorins A and E act a antagonists of neuropilin-1 and agonists of neuropilin-2 receptors. **Nature Neuroscience**. 1998, 1:487-493.
36. Nakamura, F. et al: Neuropilin-1 extracellular domains mediate semaphorin D/III-induced growth cone collapse. **Neuron**. 1998, 21:1093-1100.
37. Giger, R.J. et al.: Neuropilin-2 is a receptor for semaphorin IV: Insight into the structural basis of receptor function and specificity. **Neuron.** 1998, 21:1079-1092.
38. Chen, H. et al: Semaphorin–neuropilin interactions underlying sympathetic axon responses to class III semaphorins. **Neuron**. 1998, 21:1283-1290.
39. Feldheim, D.A. et al: Topographic guidance labels in a sensory projection to the forebrain. **Neuron**. 1998, 21:1303-1313.
40. Denda, S. et al: Identification of osteopontin as a novel ligand for the integrin alpha 8 beta 1 and potential roles for this integrin-ligand interaction in kidney morphogenesis. **Mol. Biol. Cell.** 1998, 9:1425-1435.
41. Ichijo, H. and Bonhoeffer F.: Differential withdrawal of retinal axons induced by a secreted factor. **J. Neurosci.** 1998, 18:5008-5018.
42. Durbin, L. et al.: Eph signaling is required for segmentation and differentiation of the somites. **Gene Dev.** 1998, 12:3096-3109.
43. Kang, J.S. et al.: CDO, a Robo-related cell surface protein that mediates myogenic differentiation. **J. Cell Biology.** 1998, 143:403-413.
44. Escribano, L. et al: Expression of the c-kit (CD117) molecule in normal and malignant hematopoiesis. **Leukemia Lymphoma**. 1998, 30:459-466.
45. Brose, K. et al: Slit proteins bind Robo receptors and have an evolutionarily conserved role in repulsive axon guidance. **Cell**. 1999, 96:795-806.
46. Li, H.S. et al.: Vertebrate slit, a secreted ligand for the transmembrane protein roundabout, is a repellent for olfactory bulb axons. **Cell.** 1999, 96:807-818.
47. Tamagnone, L. et al.: Plexins are a large family of receptors for transmembrane, secreted, and GPI-anchored semaphorins in vertebrates. **Cell.** 1999, 99:71-80.
48. Senzaki, K. et al: Proteins of the CNR family are multiple receptors for reelin. **Cell.** 1999, 99:635-647.
49. Chin-Sang, I.D. et al.: The ephrin VAB-2/EFN-1 functions in neuronal signaling to regulate epidermal morphogenesis in C. *elegans*. **Cell.** 1999, 99:781-790.
50. Nguyen-Ba-Charvet, K.T. et al: Slit2-mediated chemorepulsion and collapse of developing forebrain axons. **Neuron**. 1999, 22:463-473.
51. Burstyn-Cohen, T. et al.: F-spondin is required for accurate pathfinding of commissural axons at the floor plate. **Neuron.** 1999, 23:233-246.
52. Jiang, P.H. et al: Integrin-associated protein is a ligand for the P84 neural adhesion molecule. **J. Biol. Chem.** 1999, 274:559-562.
53. Chellaiah, A. et al.: Mapping ligand binding domains in chimeric fibroblast growth factor receptor molecules - Multiple regions determine ligand binding specificity. **J. Biol. Chem.** 1999, 274:34785-34794.
54. Sher, I. et al.: Mutations uncouple human fibroblast growth factor (FGF)-7 biological activity and receptor binding and support broad specificity in the secondary receptor binding site of FGFs. **J. Biol. Chem.** 1999, 274:35016-35022.
55. Berman, B. et al.: Similarities and differences between the effects of heparin and glypican-1 on the bioactivity of acidic fibroblast growth factor and the keratinocyte growth factor. **J. Biol. Chem.** 1999, 274:36132-36138.
56. Behar, O. et al.: Semaphorin 3A growth cone collapse requires a sequence homologous to tarantula hanatoxin. **Proc. Natl. Acad. Sci. USA.** 1999, 96:13501-13505.
57. Ishiguro, Y. et al: The role of amino acids surrounding tyrosine 1062 in Ret in specific binding of the Shc phosphotyrosine-binding domain. **Endocrinology.** 1999, 140:3992-3998.
58. Walter, J.J. and Sane, D.C.: Angiostatin binds to smooth muscle cells in the coronary artery and inhibits smooth muscle cell proliferation and migration *in vitro*. **Arterioscler, Thromb. & Vasc. Biol.** 1999, 19:2041-2048.
59. Feinstein, Y. et al.: F-spondin and mindin: two structurally and functionally related genes expressed in the hippocampus that promote outgrowth of embryonic hippocampal neurons. **Development.** 1999, 126:3637-3648.
60. Oohashi, T. et al.: Mouse ten-m/odz is a new family of dimeric type II transmembrane proteins expressed in many tissues. **J. Cell Biology.** 1999, 145:563-577.
61. Miyazaki, N. et al.: Mouse semaphorin H inhibits neurite outgrowth from sensory neurons. **Neurosci. Res.** 1999, 33:269-274.

This list has just the first 61 of at least 212 References using our AP-TAG technology. For the complete list, go to: http://www.genhunter.com/support/aptag_references/