BIOMOL GmbH
Waidmannstr. 35
22769 Hamburg
info@biomol.de
www.biomol.de
Phone: +49-40-8532600 or 0800-2466651 (D)
Fax: +49-40-85326022 or 0800-2466652 (D)

# Phosphoserine Detection Kit

**Order No.:** **0701/PSER-KIT**

02/080507

## Background and Specificity

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the nucleus. Phosphorylated epitopes may serve as docking sites for the assembley of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein interactions.
Modification of proteins on tyrosine residues is mediated by protein tyrosin kinases. Tyrosine phosphorylation may alter the biological activity or mediate the assembly of protein complexes via the interaction of phosphotyrosine residues with SH2 or PTB domains.
Antibodies direct against phosphorylated epitopes recognize the phosphorylated amino acid in the context of the surrounding amino acid sequence. Recognition is therefore dependent on 2 criteria: 1) phosphorylation and 2) the surrounding amino acid motif. If one of the two criteria is not fulfilled, the antibody will not detect the phosphorylation site. Since the amino acid sequence varies between different phosphorylation sites, certain proteins - though phosphorylated - may not be detected by the antibody. Phosphorylation patterns in a given cell extract may differ when probed with different antibodies due to sequence specificity.

The Phosphoserine Detection Kit contains 6 different phosphotyrosine specific monoclonal antibodies.

**Do not use Milk or Casein based blocking and incubation buffers.**

| clone | isotype | order number |
|---|---|---|
| 1C8 | IgM | 0018-025 |
| 4A3 | IgM | 0019-025 |
| 4A9 | IgM | 0020-025 |
| 4H4 | IgM | 0021-025 |
| 7F12 | IgG1 | 0022-025 |
| 16B4 | IgM | 0023-025 |

**Postive control**

This product contains the following positive control for immunoblot applications:
#0901-PSRECO phosphoproteins from rabbit muscle

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Fax: +49-40-85326022 or 0800-2466652 (D)

## Mouse Monoclonal Antibody to

# Phosphoserine

# clone 1C8

**Order No.:** **0018-025/PSER-1C8**

**Size (µg)** 25

**Lot No.:** 0018S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgM | human, mouse, rat, dog | WB, ELISA, IP | pattern | | | phosphoserine conjugated to KLH |

**Background and Specificity:**

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-1C8** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

**Related Products**

**mab against Phosphoserine**

#0019-100/pSer-4A3

#0020-100/pSer-4A9

#0021-100/pSer-4H4

#0022-100/pSer-7F12

#0023-100/pSer-16B4

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.**

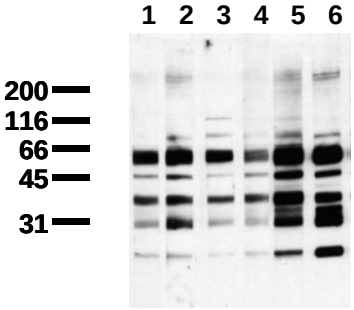
**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

**All products are supplied for research and investigational use only. Not for use in humans or laboratory animals.**

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## Mouse Monoclonal Antibody to

# Phosphoserine

# clone 4A3

**Order No.:** **0019-025/PSER-4A3**

**Size (µg)** 25

**Lot No.:** 0019S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgM | human, mouse, rat, dog | WB, ELISA, IP | pattern | | | phosphoserine conjugated to KLH |

**Background and Specificity:**

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-4A3** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.**

**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml

**All products are supplied for research and investigational use only. Not for use in humans or laboratory animals.**

**Related Products**

**mab against Phosphoserine**

#0018-100/pSer-1C8

#0020-100/pSer-4A9

#0021-100/pSer-4H4

#0022-100/pSer-7F12

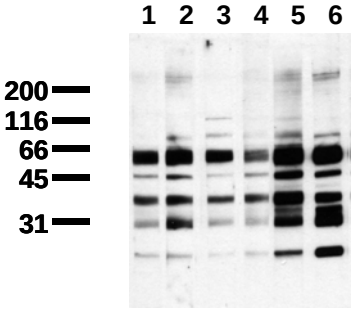
#0023-100/pSer-16B4

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

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# Mouse Monoclonal Antibody to

# Phosphoserine

# clone 4A9

**Order No.:** **0020-025/PSER-4A9**

**Size (µg)** 25

**Lot No.:** 0020S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgM | human, mouse, rat, dog | WB, ELISA, IP | pattern | | | phosphoserine conjugated to KLH |

## Background and Specificity:

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-4A9** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

## Related Products

**mab against Phosphoserine**

#0018-100/pSer-1C8

#0019-100/pSer-4A3

#0021-100/pSer-4H4

#0022-100/pSer-7F12

#0023-100/pSer-16B4

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.**

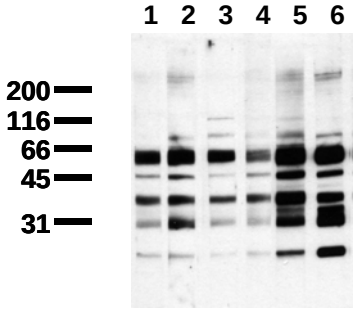
**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

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## Mouse Monoclonal Antibody to

# Phosphoserine

# clone 4H4

**Order No.:** **0021-025/PSER-4H4**

**Size (µg)** 25

**Lot No.:** 0021S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgM | human, mouse, rat, dog | WB, ELISA, IP | pattern | | | phosphoserine conjugated to KLH |

## Background and Specificity:

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-4H4** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

## Related Products

**mab against Phosphoserine**

#0018-100/pSer-1C8

#0019-100/pSer-4A3

#0020-100/pSer-4A9

#0022-100/pSer-7F12

#0023-100/pSer-16B4

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.**

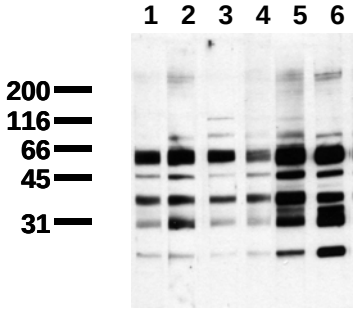
**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

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## Mouse Monoclonal Antibody to

# Phosphoserine

# clone 7F12

**Order No.:** **0022-025/PSER-7F12**

**Size (µg)** 25

**Lot No.:** 0022S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgG1 | human, mouse, rat, dog | WB, ELISA, IP | pattern | | | phosphoserine conjugated to KLH |

## Background and Specificity:

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-7F12** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

## Related Products

**mab against Phosphoserine**

#0018-100/pSer-1C8

#0019-100/pSer-4A3

#0020-100/pSer-4A9

#0021-100/pSer-4H4

#0023-100/pSer-16B4

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.v**

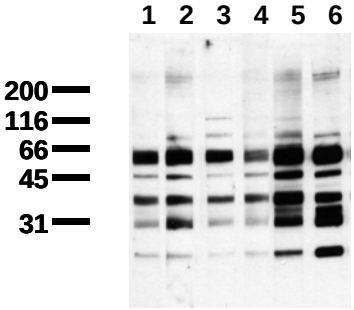
**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

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## Mouse Monoclonal Antibody to

# Phosphoserine

# clone 16B4

**Order No.:** **0023-025/PSER-16B4**

**Size (µg)** 25

**Lot No.:** 0023S

03/160307F

| Isotype | Species Reactivity | Applications | Mol. Weight | Ref.Cell Line | Epitope | Immunogen |
|---|---|---|---|---|---|---|
| IgM | human, mouse, rat, dog | WB, ELISA, IP | pattern | | ...pSer - Pro...;...pSer - Lys | phosphopeptide conjugated to KLH |

## Background and Specificity:

Phosphorylation and dephosphorylation of cellular proteins are central steps in transducing extracellular signals to the cell nucleus. Phosphorylated epitopes may serve as docking sites for the assembly of protein complexes or may alter the 3-dimensional protein structure thus modulating enzymatic activity or the ability to undergo protein-protein-interactions. Modification of proteins on serine residues is mediated by serine/threonine kinases.

**Please note that phosphoserine detection by monoclonal antibodies is always dependent on the surrounding amino acid sequence!**

**Mab PSER-16B4** recognizes a broad range of serine-phosphorylated proteins in crude cell extracts, preferring positively charged amino acids directly neighboured to phosphoserine.

## Related Products

**mab against Phosphoserine**

#0018-100/pSer-1C8

#0019-100/pSer-4A3

#0020-100/pSer-4A9

#0021-100/pSer-4H4

#0022-100/pSer-7F12

**mab against Phosphothreonine**

#0024-100/pThr-1E11

#0025-100/pThr-4D11

#0026-100/pThr-14B3

**Purification:** The antibody was purified from serum-free cell culture supernatant by subsequent thiophilic adsorption and size exclusion chromatography.

**Formulation:** lyophilized from 1 ml 2 x PBS / 0.09 % Na-azide / PEG and Sucrose.

**Reconstitution:** Reconstitute with 1 ml $H_2O$ (15 min, RT).

**Stability:** For long-term storage, freeze lyophilizate upon arrival (-20°C). Upon reconstitution, aliquote and freeze in liquid nitrogen; reconstituted antibody can be stored frozen at -80°C up to 1 year. Thaw aliquots at 37°C. Thawed aliquots may be stored at 4°C up to 1 week.

**Avoid repeated freeze / thaw cycles.**

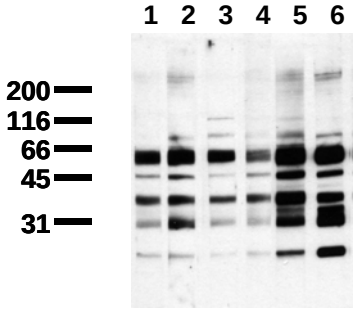
**Positive Control:** #0901: phosphoserine/phosphothreonine positive control

**Immunoblotting:** 1 µg/ml for HRPO/ECL detection
**Recommended blocking buffer:** BSA/Tween 20 based blocking buffer.
**DO NOT USE MILK OR CASEIN FOR BLOCKING!**

**Immunoprecipitation:** use at 1 - 10 µg per $10^6$ pervanadate-treated A431 cells

**Immunocytochemistry:** ND

**ELISA:** use at 0.05 µg/ml




**Phosphoserine Detection**
Phosphoprotein Positive Control was probed with
lane 1: mab 1C8 (IgM), 1 µg/ml
lane 2: mab 4A3 (IgM), 1 µg/ml
lane 3: mab 4A9 (IgM), 1 µg/ml
lane 4: mab 4H4 (IgM), 1 µg/ml
lane 5: mab 7F12 (IgG), 1 µg/ml
lane 6: mab 16B4 (IgM), 1 µg/ml

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# pSer / pThr Molecular Weight Marker

03/100407F

**Order No.:** **0901/PSERCO**

**Lot:** **0901**

**Size** **20 Blots**

## Formulation

The pSer/pThr molecular weight marker contains rabbit muscle phosphoproteins isolated by Fe3+/IDA - affinity chromatography. Proteins are lyophilized from PBS/NaF/PEG/Sucrose/ Bromo-phenolblue and Na - azide. After reconstitution the solution contains 0.09% Na-azide.

## Stability

Reconstitute by addition of 200 µl $H_2O$. After complete solubilization add 200 µl 2x SDS-PAGE sample buffer, mix and incubate at 90°C for 5 min.

**Aliquote and store frozen. Avoid repeated freeze/thaw cycles.**

## Application

The pSer/pThr molecular weight marker is recommended for immunoblot applications. Use 20µl molecular weight marker per lane.

Note: Use BSA based blot incubation buffers. Milk, Casein and Blotto might interfere with antibody - antigen interaction.