www.biossusa.com
800.501.7654 [DOMESTIC]
+1.781.569.5821 [INT'L]
support@biossusa.com

MBP Ready-To-Use IHC Kit

Cat. No.: IHC0107

Sample Type: FFPE tissue **Size: 50T (including three control slides)**

Storage and Stability: Please store components at the temperatures indicated on the individual tube labels. The kit is stable for 6 months from the date of receipt.

General Information

Number	Component	Size	Concentration	Storage
1	PBS Buffer (powder)	2 L×2	20x	RT
2	Antigen Retrieval Buffer	20 ml	100x	2-8℃
3	Endogenous Peroxidase Blocking Buffer	3 ml	RTU	2-8℃
4	Blocking Buffer	3 ml	RTU	2-8℃
5	Primary Antibody (MBP Rabbit pAb)	6 ml	RTU	2-8℃
6	Secondary Antibody (HRP-Goat anti-Rabbit IgG pAb)	6 ml	RTU	2-8℃
7	Chromogen Component A	0.3 ml	RTU	-20℃
8	Chromogen Component B	0.3 ml	RTU	-20℃
9	Counter Staining Reagent	5 ml	RTU	RT
10	Mounting Media	5 ml	RTU	RT
11	Control slide (Human brain, mouse brain, rat brain)	3 slides	RTU	RT
12	Datasheet	1 copy		

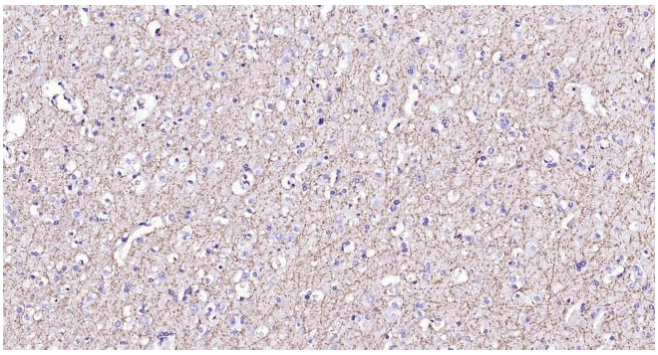
Background

The protein encoded by the classic MBP gene is a major constituent of the myelin sheath of oligodendrocytes and Schwann cells in the nervous system. However, MBP-related transcripts are also present in the bone marrow and the immune system. These mRNAs arise from the long MBP gene (otherwise called "Golli-MBP") that contains 3 additional exons located upstream of the classic MBP exons. Alternative splicing from the Golli and the MBP transcription start sites gives rise to 2 sets of MBP-related transcripts and gene products. The Golli mRNAs contain 3 exons unique to Golli-MBP, spliced in-frame to 1 or more MBP exons. They encode hybrid proteins that have N-terminal Golli aa sequence linked to MBP aa sequence. The second family of transcripts contain only MBP exons and produce the well characterized myelin basic proteins. This complex gene structure is conserved among species suggesting that the MBP transcription unit is an integral part of the Golli transcription unit and that this arrangement is important for the function and/or regulation of these genes.

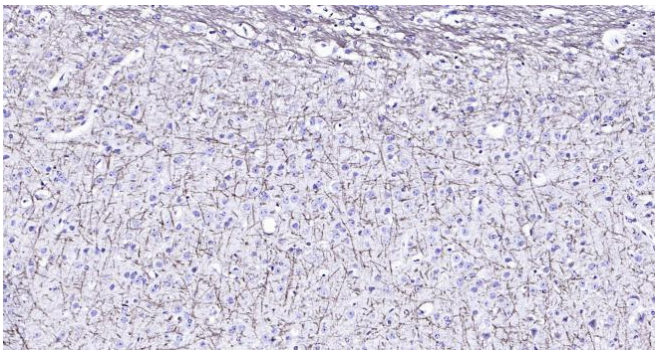
Synonyms

Myelin Basic Protein; Myelin basic protien; GDB; Golli MBP; Hemopoietic MBP; HMBPR; HUGO; MBP; MGC99675; MLD; Myelin A1 Protein; Myelin Deficient; Myelin Membrane Encephalitogenic Protein; SHI; Shiverer; SP; MBP_HUMAN.

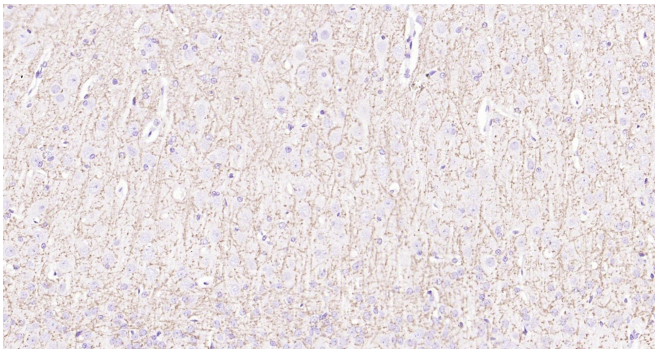
Validation Data



Immunohistochemical analysis of paraffin embedded human brain tissue slide using IHC0107 (MBP IHC Kit).



Immunohistochemical analysis of paraffin embedded mouse brain tissue slide using IHC0107 (MBP IHC Kit).



Immunohistochemical analysis of paraffin embedded rat brain tissue slide using IHC0107 (MBP IHC Kit).

Immunohistochemistry Protocol

1. Deparaffinization And Rehydration

Immerse slides in fresh xylene for 15 minutes and then repeat two more times using separate containers. Immerse slides sequentially in 100%, 95%, 90%, 80%, and 70% ethanol solutions for 5 minutes each. Rinse slides 3 times with distilled water for 5 minutes each.

2. Antigen Retrieval

Add 100×**Antigen Retrieval Buffer** into distilled water to prepare a 1×solution. Boil slides in 1×solution at 95°C-100°C for 15 minutes. Move the slides to 1×solution at room temperature (RT) and allow them to stand for 20 minutes. Rinse 3 times with **PBS Buffer** (dissolve the powder in 2L distilled water) for 5 minutes each.

3. Block Endogenous Peroxidase

Drain the liquid off the slides and then use a hydrophobic IHC pen to draw circles on the slides around tissue sections. Add 2-4 drops of **Endogenous Peroxidase Blocking Buffer** directly on slides, covering the whole tissue and block slides for 15 minutes at RT. Rinse 3 times with **PBS Buffer** for 5 minutes each.

4. Serum Blocking

Block with 2-4 drops of **Blocking Buffer** for 20 minutes at RT.

5. Primary Antibody Incubation

Drain blocking buffer from slides. Incubate slides with 2-4 drops of **MBP Rabbit pAb** overnight at 4°C or 1-2 hours at RT. Rinse 3 times with **PBS Buffer** for 5 minutes each.

6. Secondary Antibody Incubation

Incubate slides with 2-4 drops of **HRP-Goat anti-Rabbit IgG pAb** for 1-2 hours at RT. Rinse slides 3 times with **PBS Buffer** for 5 minutes each.

7. Signal Development

Remove residual liquid around the tissue section. Add 50ul fresh **DAB Buffer** (**Chromogen Component A : Chromogen Component B : PBS Buffer=1:1:18**) to cover the tissue. Monitor the reaction under the microscope until a brown color is visible (approximate 3-5 minutes at RT). Stop reaction immediately by rinsing with distilled water. Rinse slides 3 times with distilled water for 5 minutes each.

8. **Counterstain**

Counterstain with an appropriate amount of **Counter Staining Reagent** for 3-5 minutes at RT. Rinse slides with distilled water for 5 minutes. Use 2-4 drops of **Differentiation reagent** to cover the tissue for 30 seconds. Rinse slides twice with distilled water for 5 minutes each.

9. Dehydration Sheet

Immerse slides sequentially in 70%, 80%, 90%, 95%, and 100% ethanol for 5 minutes each at RT. Immerse slides in 2 changes of fresh xylene, 15 minutes each. Drop some **Mounting Media** on the tissue. Mount coverslips.

Notes

1. The positive control slide provided in the kit allows you to be sure that the experimental set-up is working properly.
2. Do not allow slides to dry at any time during this procedure.
3. Please don't replace the matching reagents in this product with other manufacturers' products.
4. As DAB is a carcinogen, please take necessary precautions.
5. PBS (reagent 1) can be stored for one week at 4℃ after preparation; The antigen retrieval buffer (1×reagent 2) and the chromogenic agent (the mixture of reagents 7 and 8) should be prepared right before each assay.

Please cite this product as "IHC0107, Bioss Antibodies". Citation example: "Tissue sections using MBP IHC Kit (IHC0107, Bioss Antibodies) were stained for MBP according to the manufacturer's instructions.

400-901-9800
sales@bioss.com.cn
techsupport@bioss.com.cn

MBP Ready-To-Use IHC Kit

髓鞘碱性蛋白/磷脂碱性蛋白即用型免疫组化试剂盒

产品货号：IHC0107
样本类型：FFPE 组织切片 **产品规格：50T （包含三个对照切片）**
保存条件：见下表。有效期 6 个月。

产品组分及规格

编号	组分	规格	浓度	储存
1	PBS 缓冲液（干粉）	2 L×2	20x	室温
2	抗原修复缓冲液	20 ml	100x	2-8℃
3	内源性过氧化物酶阻断剂	3 ml	RTU	2-8℃
4	封闭工作液	3 ml	RTU	2-8℃
5	一抗（MBP Rabbit pAb）	6 ml	RTU	2-8℃
6	二抗（HRP-Goat anti-Rabbit IgG pAb）	6 ml	RTU	2-8℃
7	DAB kit（20×）显色液	0.3 ml	RTU	-20℃
8	DAB kit（20×）稀释液	0.3 ml	RTU	-20℃
9	复染试剂	5 ml	RTU	室温
10	封片剂	5 ml	RTU	室温
11	对照切片（大鼠脑，小鼠脑，人脑）	3 张	RTU	室温
12	说明书	1 份		

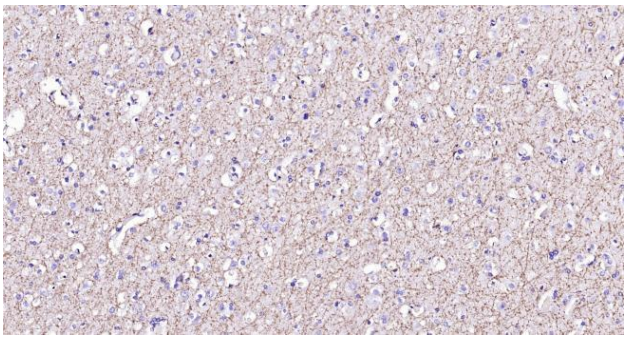
背景

The protein encoded by the classic MBP gene is a major constituent of the myelin sheath of oligodendrocytes and Schwann cells in the nervous system. However, MBP-related transcripts are also present in the bone marrow and the immune system. These mRNAs arise from the long MBP gene (otherwise called "Golli-MBP") that contains 3 additional exons located upstream of the classic MBP exons. Alternative splicing from the Golli and the MBP transcription start sites gives rise to 2 sets of MBP-related transcripts and gene products. The Golli mRNAs contain 3 exons unique to Golli-MBP, spliced in-frame to 1 or more MBP exons. They encode hybrid proteins that have N-terminal Golli aa sequence linked to MBP aa sequence. The second family of transcripts contain only MBP exons and produce the well characterized myelin basic proteins. This complex gene structure is conserved among species suggesting that the MBP transcription unit is an integral part of the Golli transcription unit and that this arrangement is important for the function and/or regulation of these genes.

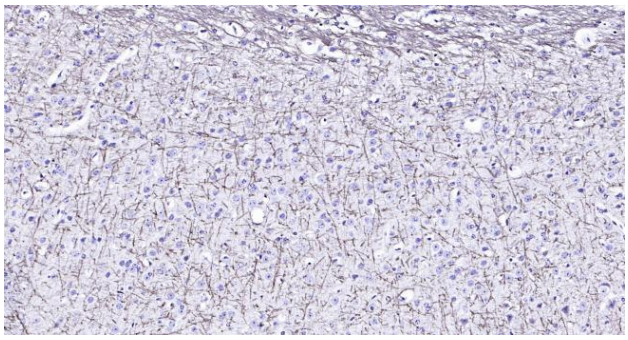
别名

Myelin Basic Protein; Myelin basic protien; GDB; Golli MBP; Hemopoietic MBP; HMBPR; HUGO; MBP; MGC99675; MLD; Myelin A1 Protein; Myelin Deficient; Myelin Membrane Encephalitogenic Protein; SHI; Shiverer; SP; MBP_HUMAN.

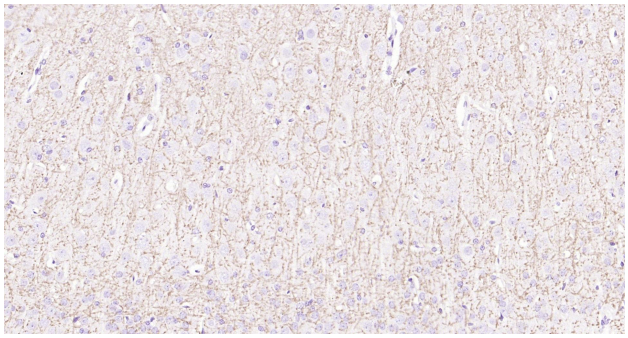
验证数据



使用 IHC0107（髓鞘碱性蛋白/磷脂碱性蛋白 IHC 试剂盒）对石蜡包埋的人脑组织切片进行免疫组化分析。



使用 IHC0107（髓鞘碱性蛋白/磷脂碱性蛋白 IHC 试剂盒）对石蜡包埋的小鼠脑组织切片进行免疫组化分析。



使用 IHC0107（髓鞘碱性蛋白/磷脂碱性蛋白 IHC 试剂盒）对石蜡包埋的大鼠脑组织切片进行免疫组化分析。

石蜡包埋组织的免疫组织化学方案

1. 脱蜡水化

石蜡切片置于新鲜二甲苯中浸泡脱蜡 3 次，每次 15 min；依次置于不同浓度（100%、95%、90%、80%、70%）乙醇浸泡各 5 min，再置于蒸馏水洗涤 5 min，重复 3 次。

2. 抗原修复

沸水浴修复：将 100×**抗原修复缓冲液（试剂 2）**用蒸馏水稀释成 1×抗原修复缓冲液，放入修复盒中并提前加热至 95-100℃（注意盖好以防液体蒸发），然后将切片放入修复盒中，在沸水浴环境中保持外沸状态 15 min，室温自然冷却；用 **PBS 缓冲液（试剂 1**，将干粉溶解在 2L 蒸馏水中）清洗 5 min，重复 3 次。

3. 阻断内源性过氧化物酶

用吸水纸吸去玻片上多余的液体，用免疫组化笔在组织周围画圈，加入 2-4 滴**内源性过氧化物酶阻断剂（试剂 3）**，室温下置于湿盒中孵育 15 min，用 PBS 洗涤 5 min，重复 3 次。

4. 血清封闭

用吸水纸吸去玻片上多余的液体，加入 2-4 滴**封闭工作液（试剂 4）**，置于湿盒内 37℃封闭 20 min，以减少非特异性染色。

5. 一抗孵育

用吸水纸吸去玻片上多余的液体，加入 2-4 滴 **MBP 兔多抗工作液（试剂 5）**，置于湿盒中，4℃孵育过夜或 37℃孵育 1-2 h。

6. 复温

4℃孵育过夜后，室温下复温 15 min（若在室温下孵育一抗，则直接进入下一步清洗）；用 PBS 洗涤 5 min，重复 3 次。

7. 二抗孵育

用吸水纸吸玻片上多余的液体，加入 2-4 滴 **HRP 标记羊抗兔 IgG 工作液（试剂 6）**，置于湿盒中，37℃孵育 1-2 h；用 PBS 洗涤 5 min，重复 3 次。

8. 显色

用吸水纸吸去玻片上多余的液体，在每张切片上滴加约 50 μL 新配制的 **DAB 工作液（试剂 7:试剂 8:PBS=1:1:18）**，作用 3-5 min。显微镜下观察结果，达到合适的显色强度后，用蒸馏水冲洗切片以终止反应，用蒸馏水冲洗 5 min，重复 3 次。

9. 复染

滴加适量**复染试剂（试剂 9）**复染 3-5 min，蒸馏水冲洗 5 min，滴加盐酸酒精分化约 30 s，蒸馏水洗涤 5 min，重复 2 次。

10. 脱水封片

将玻片依次置于不同浓度（70%、80%、90%、95%、100%）乙醇，各 5 min；然后置于新鲜二甲苯中浸泡脱蜡 3 次，每次 15 min。用吸水纸吸去多余的二甲苯，滴加适量**封片剂（试剂 10）**在组织上，将盖玻片盖在组织上，避免产生气泡。

注意事项

1. 建议检测时进行阴性及阳性对照，以提高实验的可靠性。
2. 本品中的配套试剂，请不要用其他生产商产品替换使用。
3. DAB 为致癌物质，请采取必要的防范措施。
4. PBS 洗涤液（试剂 1）配制后在 4℃可保存一周；抗原修复液（试剂 2）及显色剂（试剂 7 和 8）的工作液需每次实验时现用现配。

*5. 发表论文时引用本产品的写作建议 "IHC0107, Bioss Antibodies"。引用示例: “Tissue sections using MBP IHC Kit (IHC0107, Bioss Antibodies) were stained for MBP according to the manufacturer's instructions.”