

施万细胞糖尿病周围神经病变模型的构建及其对轴突再生机制的探究

汇报人：施奕欣

指导老师：余巍

2025.5.27



目录

01

研究背景及意义

02

研究方法和结果

03

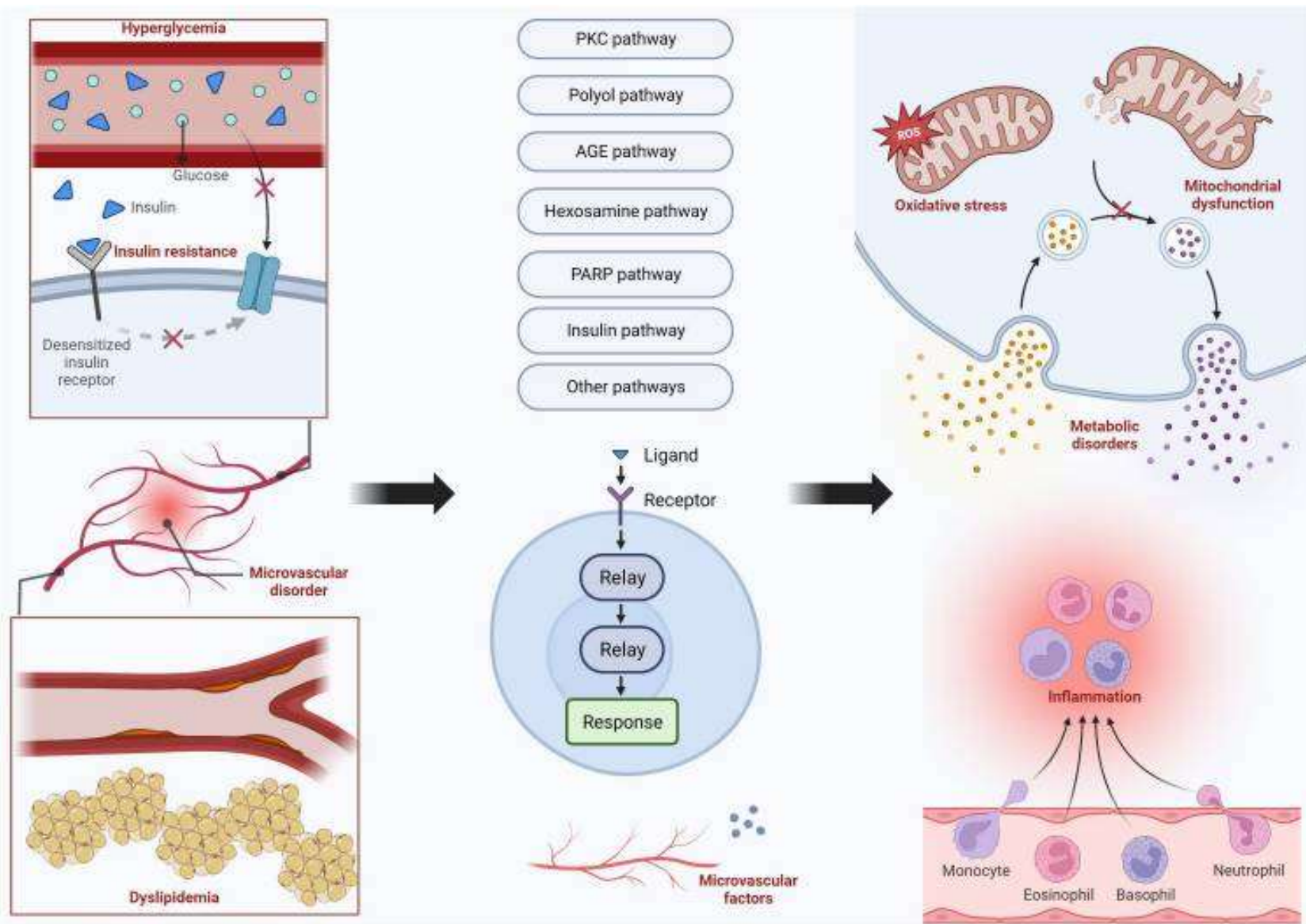
结论与展望

01

研究背景及意义



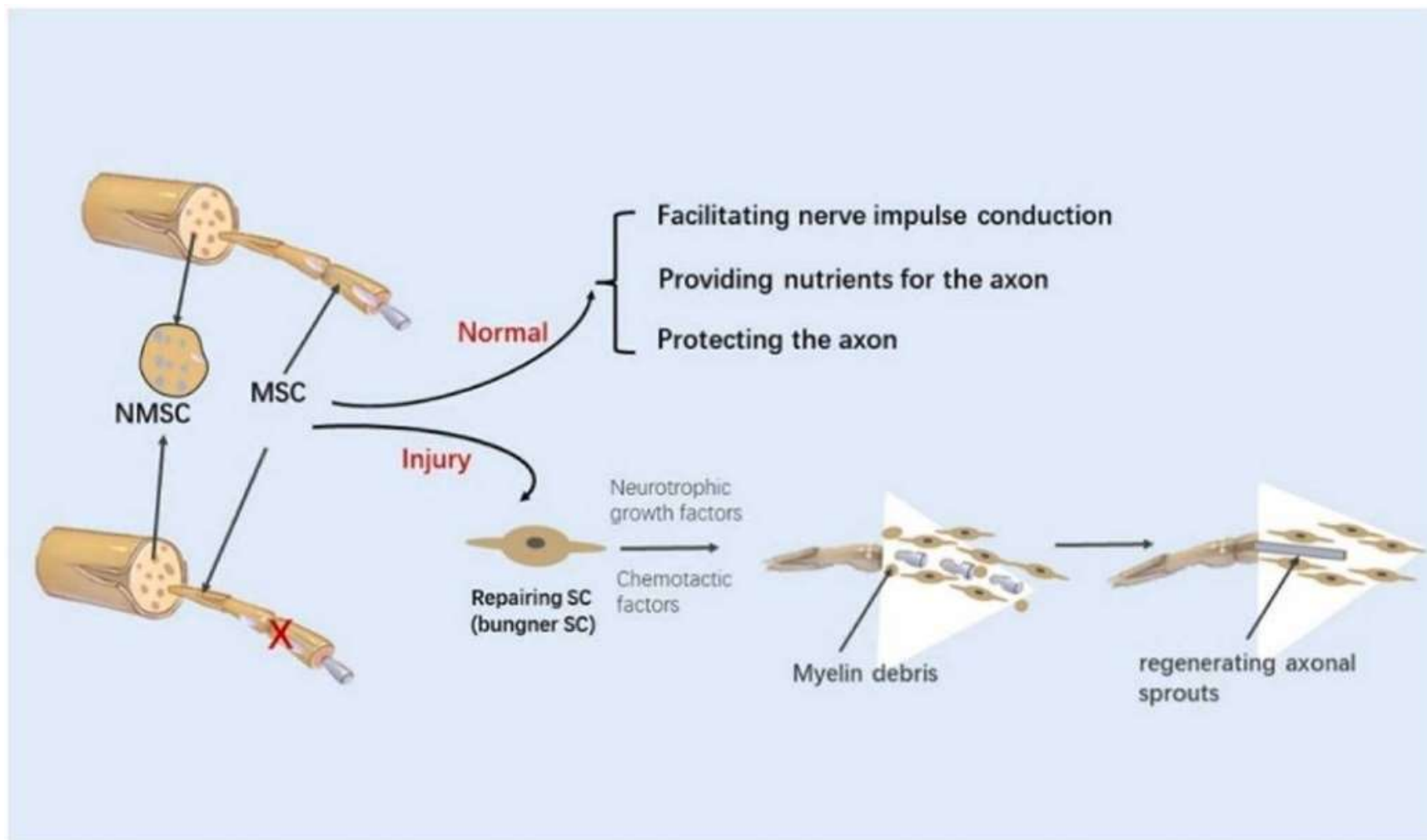
糖尿病周围神经病变 (DPN)



- 高血糖
- 血脂异常
- 胰岛素抵抗
- 微血管疾病



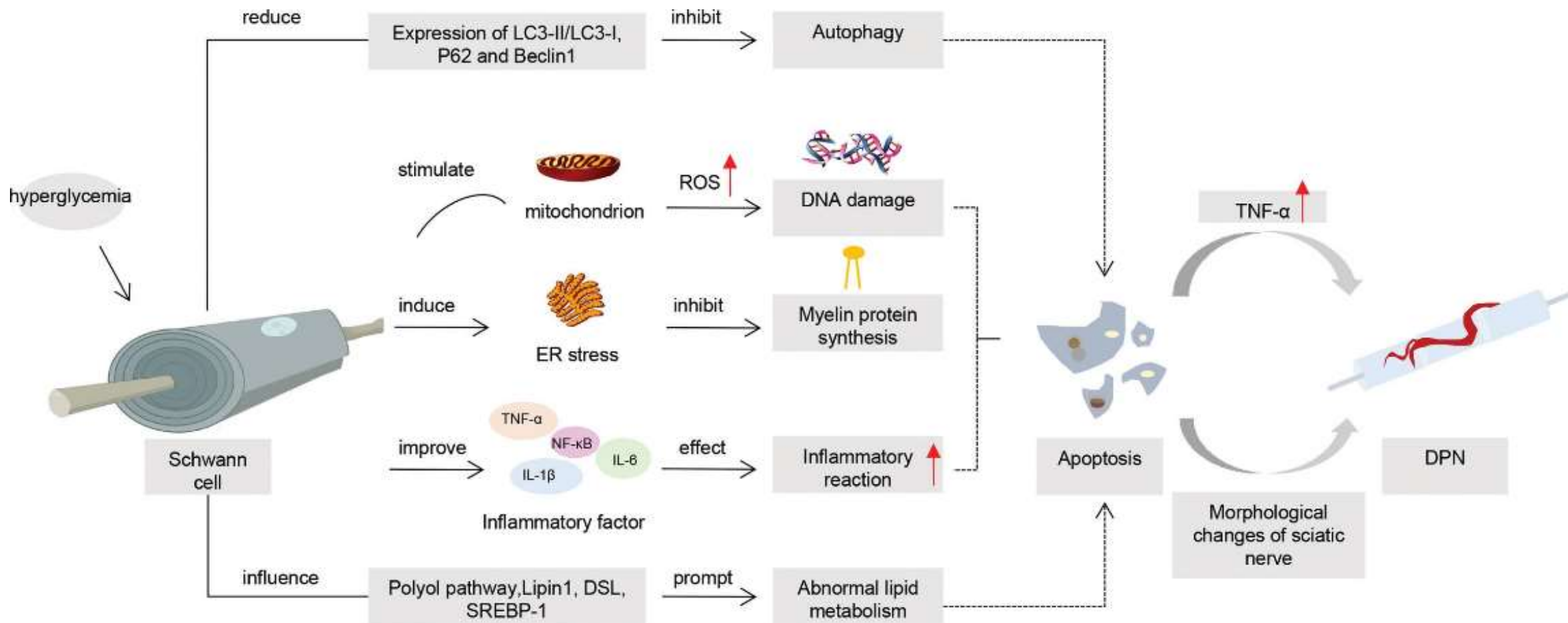
施万细胞



- 形成髓鞘
- 轴突能量供应
- 参与轴突再生
- 调节神经元功能和修复



施万细胞损伤导致DPN进展



Li J, et al., Medicine (Baltimore), 2023



施万细胞的高糖模型

cells/well and allowed to adhere overnight. Then cells were cultured in medium with 25-mM-high glucose and 0.1, 1, 10, 25 and 50- μ M loganin for 24, 48 and 72 h. After treatment, the medium was refreshed and 10 μ L of the CCK-8 solution was added to each well. After incubation for 2 h at 37 °C at 5% CO₂,

Cheng Y C, et al. Loganin Attenuates High Glucose Induced Schwann Cells Pyroptosis by Inhibiting ROS Generation and NLRP3 Inflammasome Activation, 2020

humidified 5% CO₂, 37 °C incubator. The cells were exposed to 30 mM high glucose (HG) for 6, 12, 24, 48 and 96 h, respectively. The

Li R, et al. NGF Attenuates High Glucose-Induced ER Stress, Preventing Schwann Cell Apoptosis by Activating the PI3K/Akt/GSK3 β and ERK1/2 Pathways, 2017

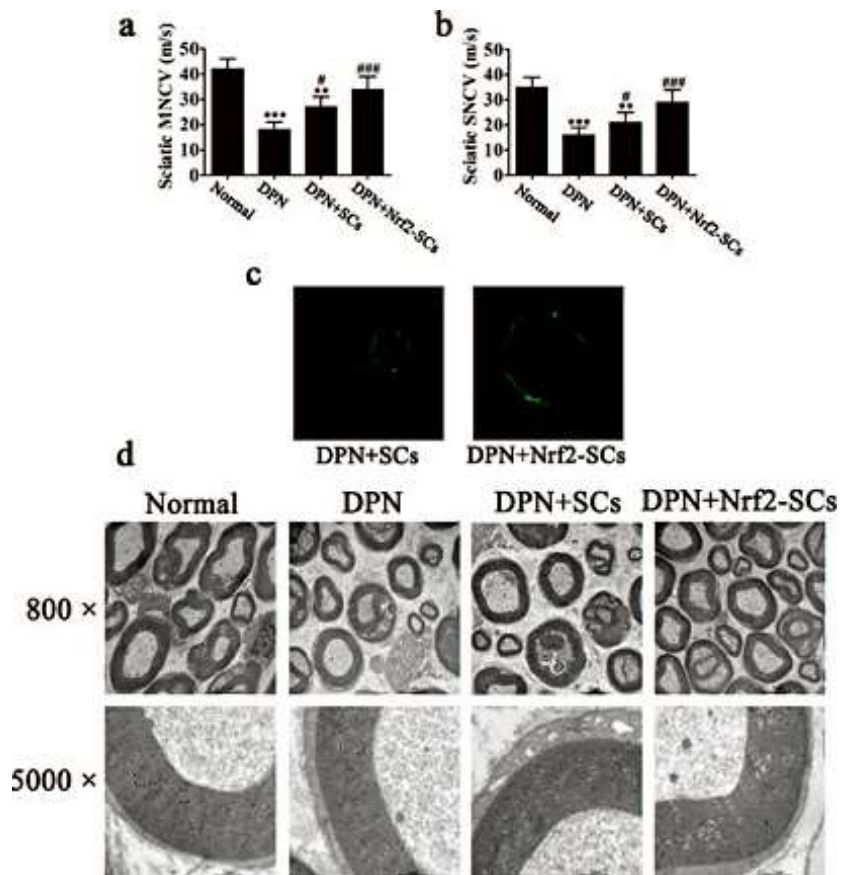
medium; HG group was treated with glucose (supplemented with 150 mM glucose dissolved in glucose-free DMEM) for 48 h; HNK groups were

Hu M, et al. Honokiol attenuates high glucose-induced peripheral neuropathy via inhibiting ferroptosis and activating AMPK/SIRT1/PGC-1 α pathway in Schwann cells, 2023

是否能建立一个长期高糖高脂环境下的施万细胞模型？

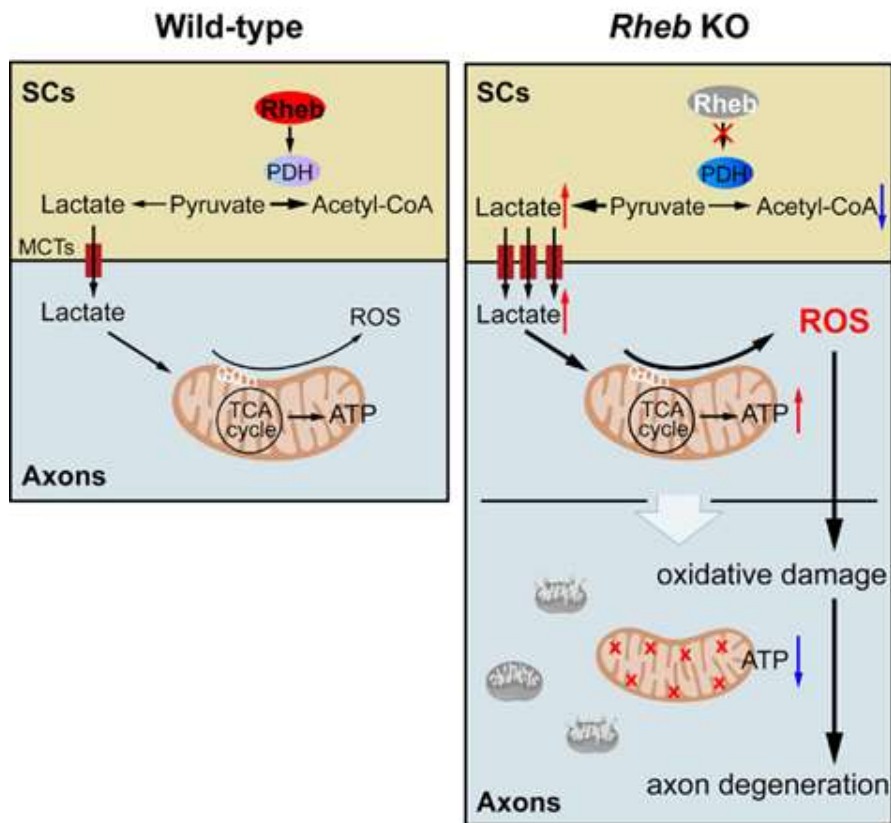


施万细胞修复神经损伤



改善施万细胞氧化应激状态能恢复神经传导速度

Tang W. et al., Cell Physiol Biochem, 2018



施万细胞中小GTP酶Rheb调控的丙酮酸代谢对轴突再生的影响

Jia L. et al., Dev Cell, 2021

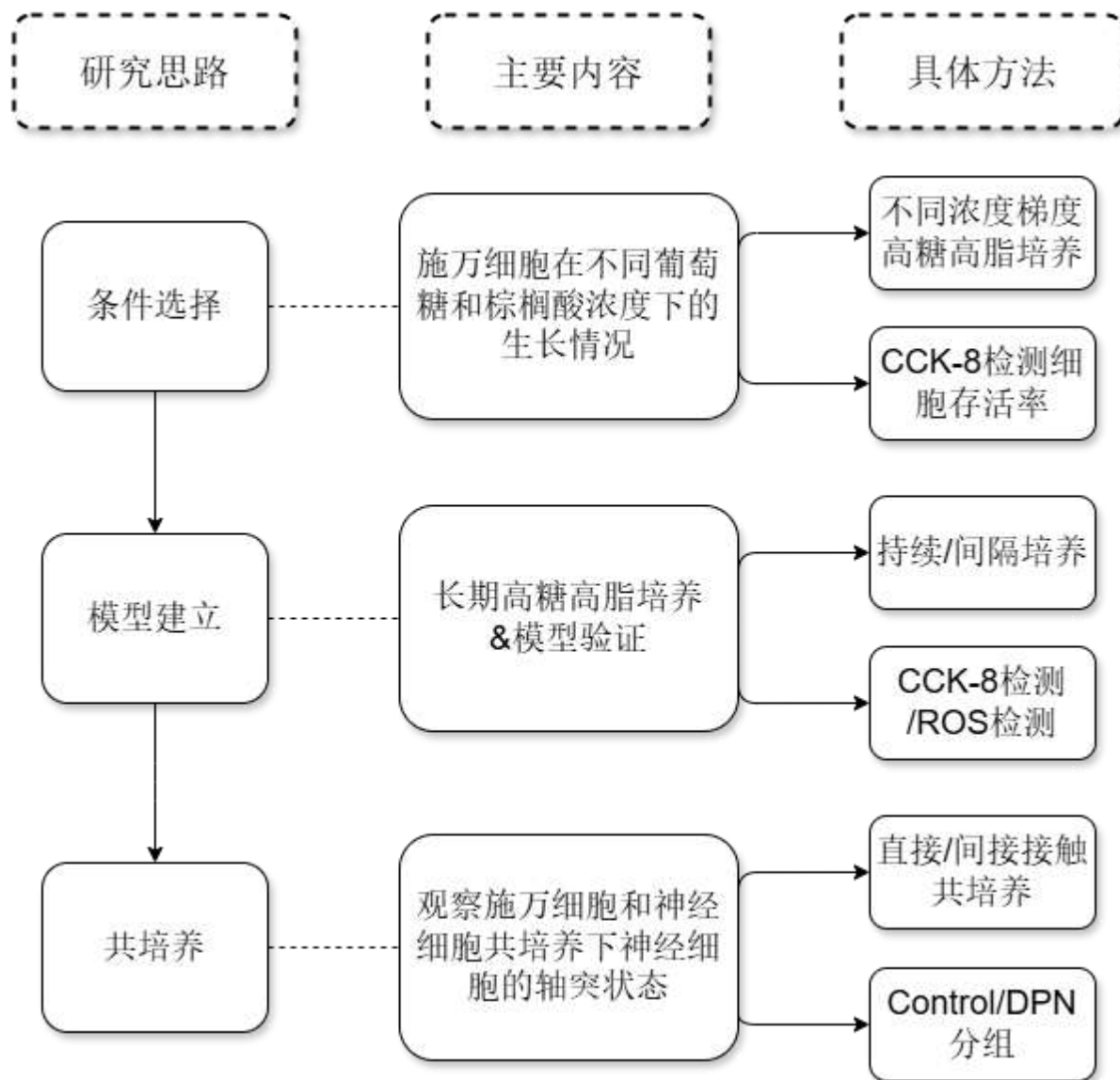
长期高糖高脂培养的施万细胞对神经细胞轴突再生有何影响？

02

研究方法和结果



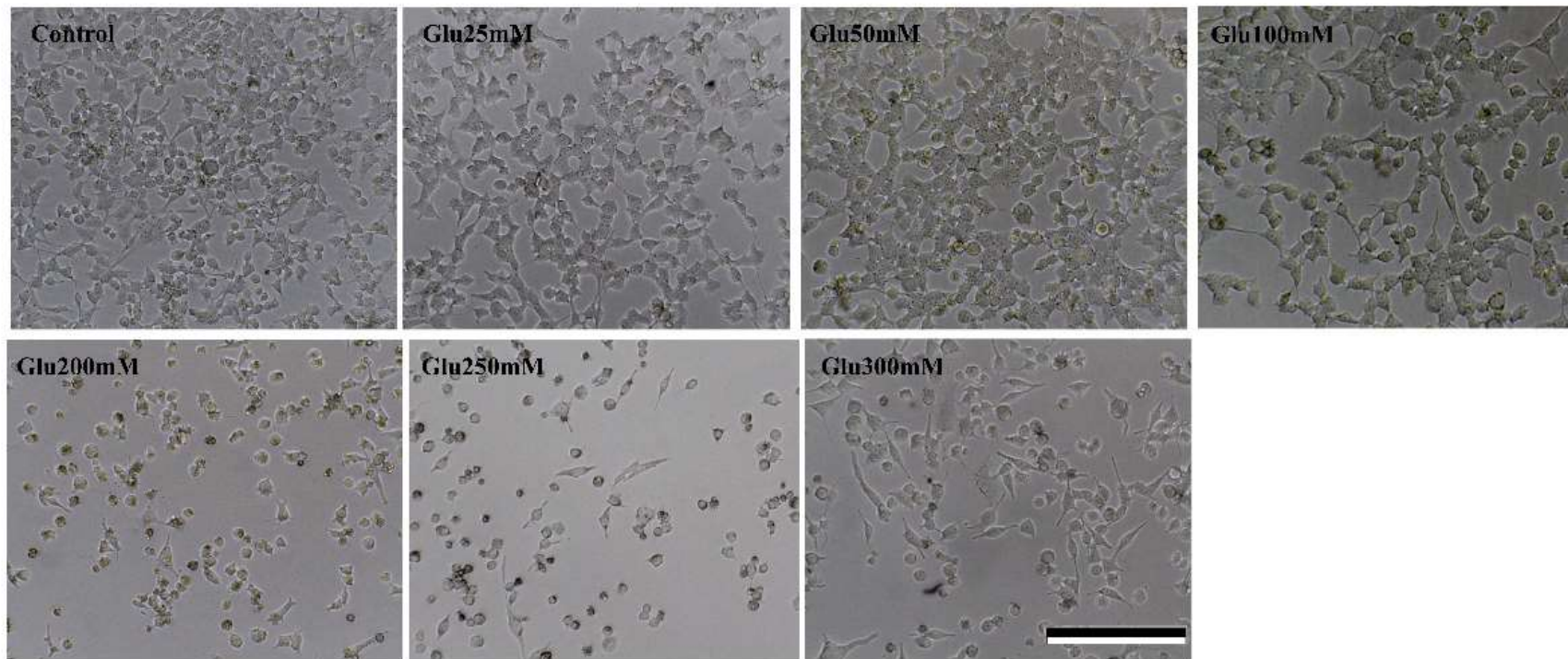
研究思路



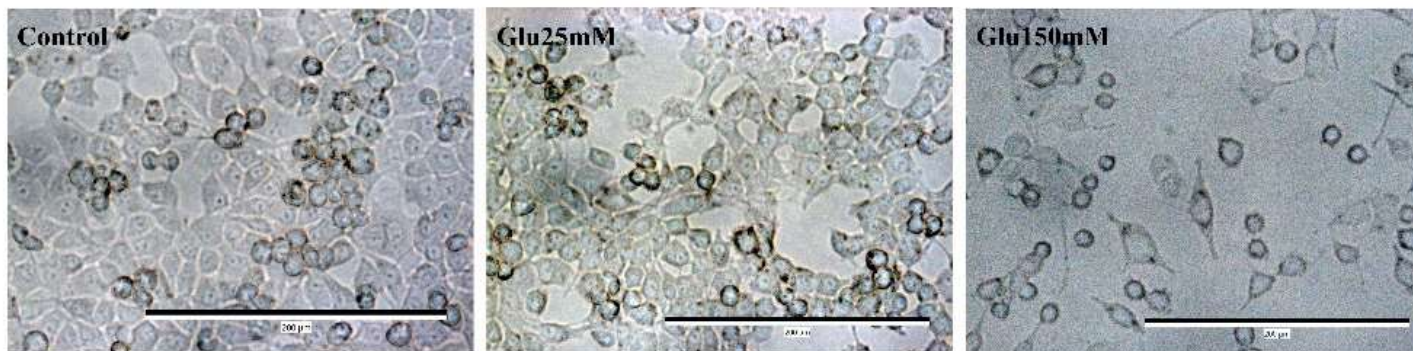


高浓度葡萄糖抑制施万细胞生长

A



B

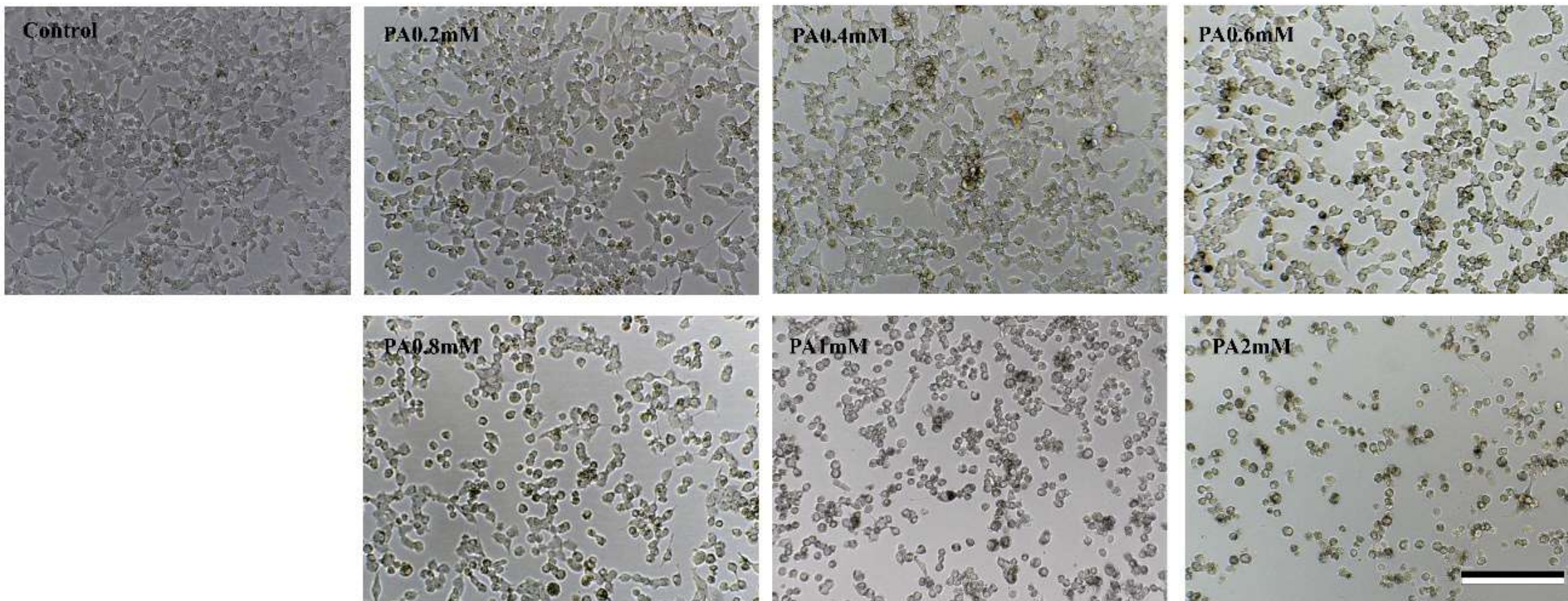


不同浓度葡萄糖下施万细胞生长情况（24h）

标尺200 μ m； Control: 完全培养基组； Glu25、50、100、200、250、300mM: 含25、50、100、200、250、300mM Glu 的完全培养基



高浓度棕榈酸抑制施万细胞生长



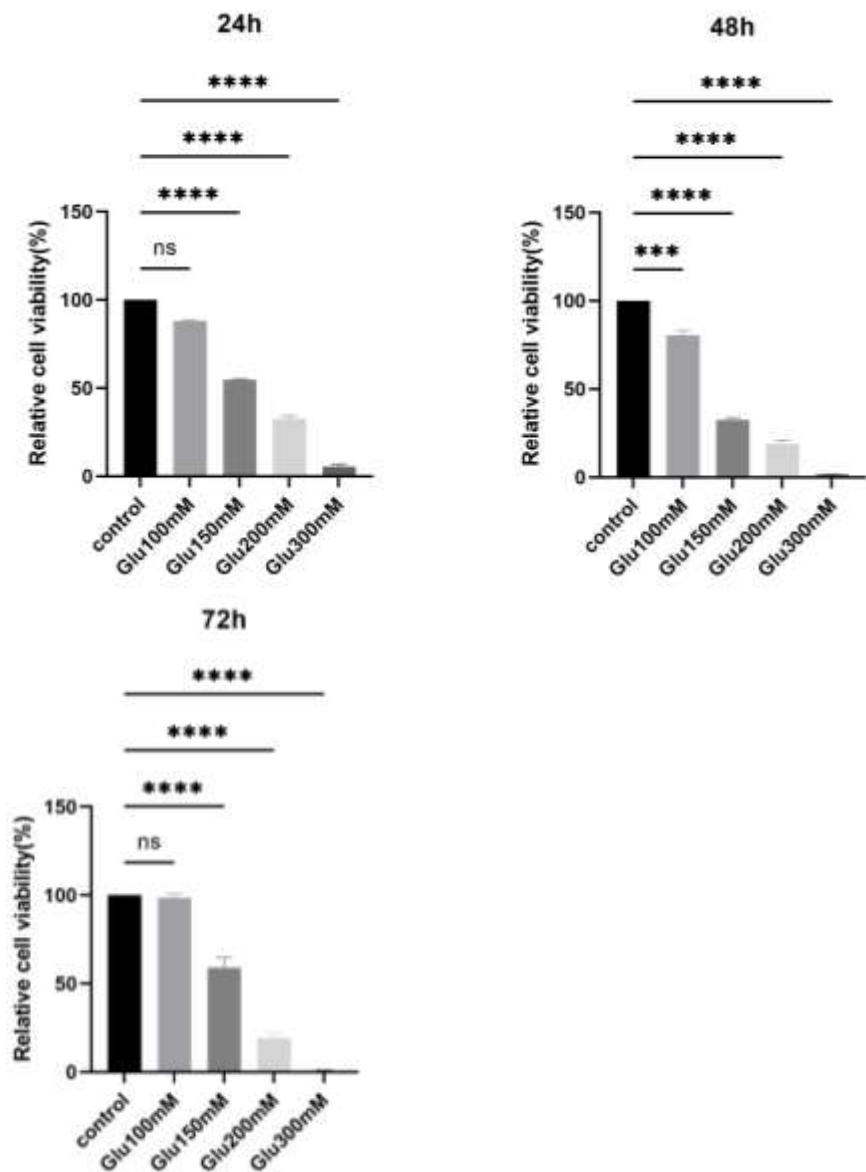
不同浓度棕榈酸下施万细胞生长情况（24h）

标尺200 μ m; Control: 完全培养基组; PA0.2、0.4、0.6、0.8、1、2mM: 含0.2、0.4、0.6、0.8、1、2mM PA 的完全培养基

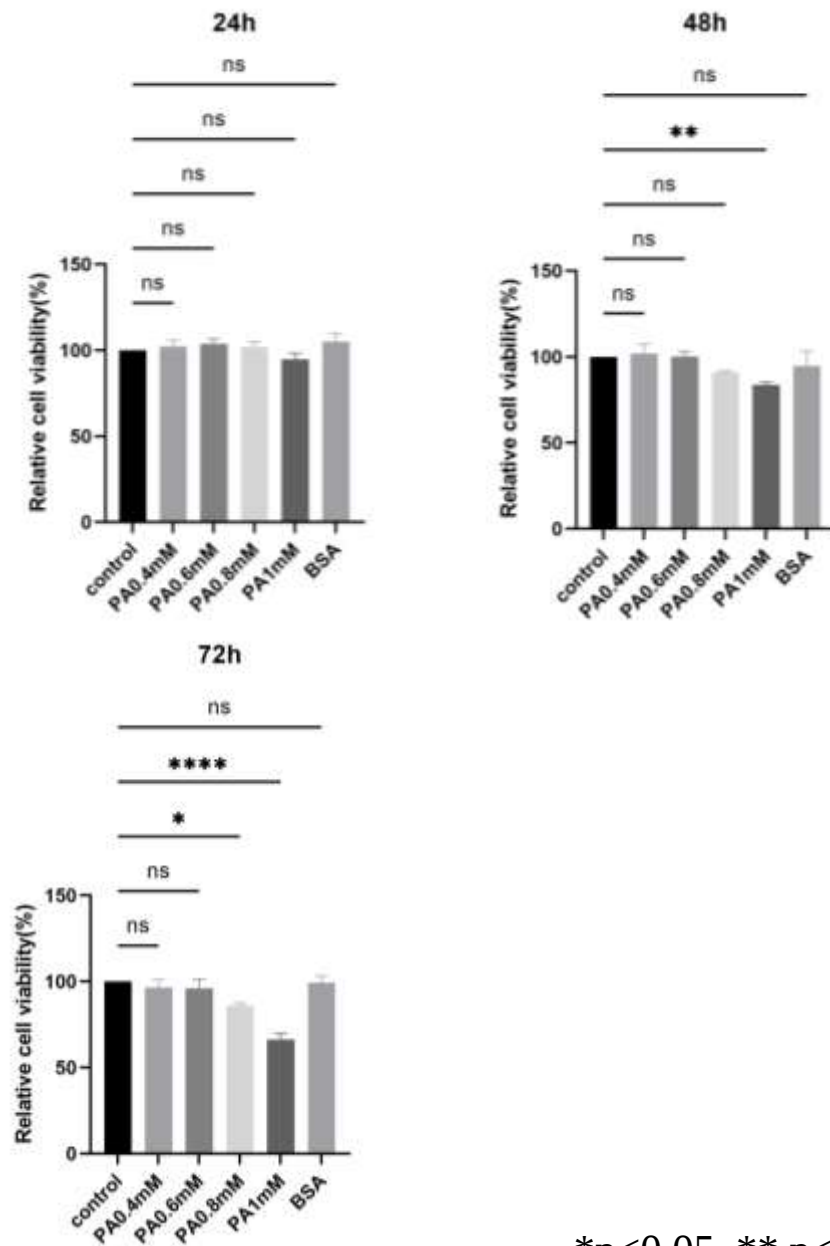


高浓度葡萄糖/棕榈酸抑制施万细胞生长

A



B

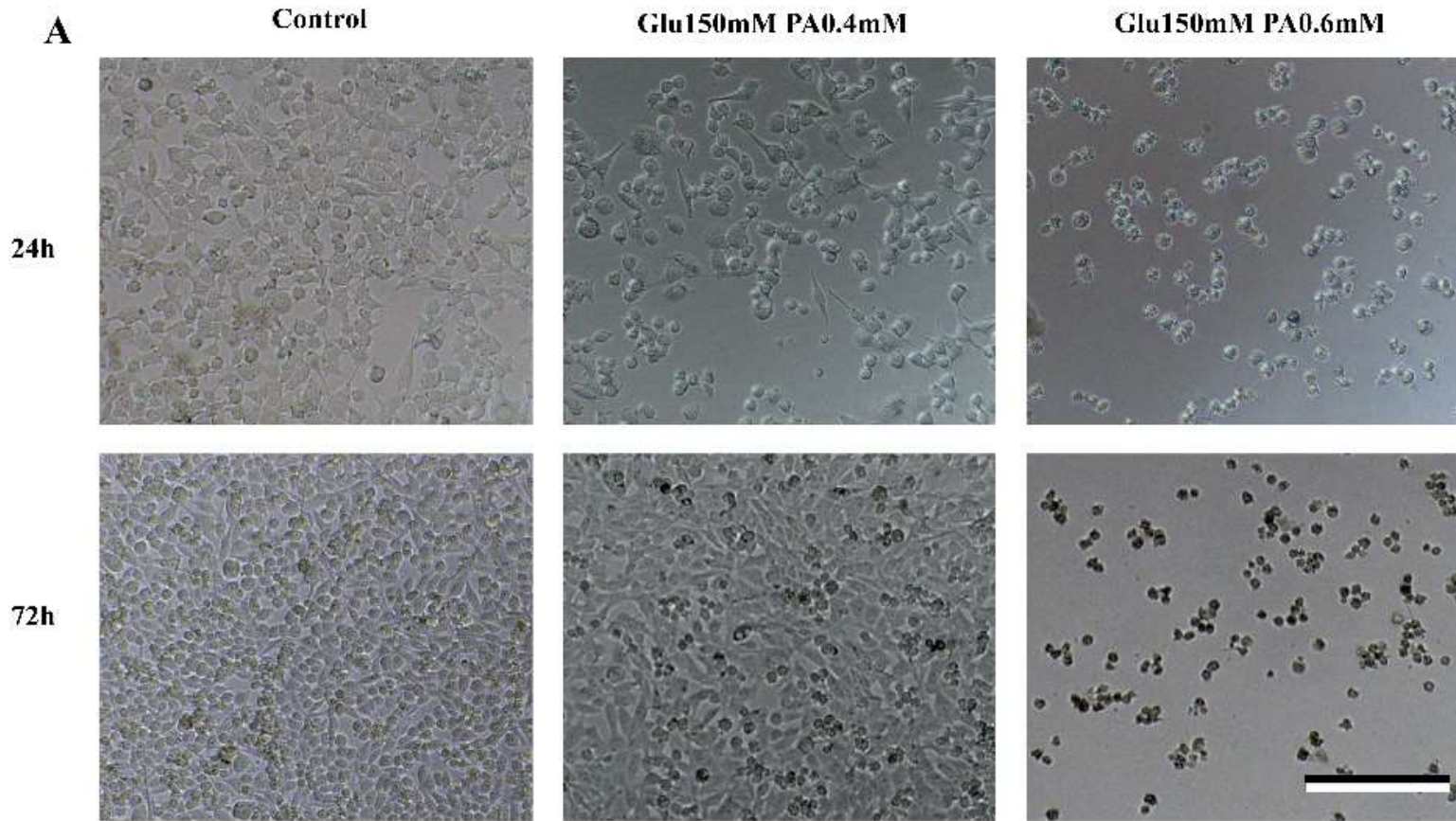


不同葡萄糖或棕榈酸浓度下施万细胞存活率（24、48、72h）

* $p < 0.05$, ** $p < 0.01$,
*** $p < 0.001$, **** $p < 0.0001$

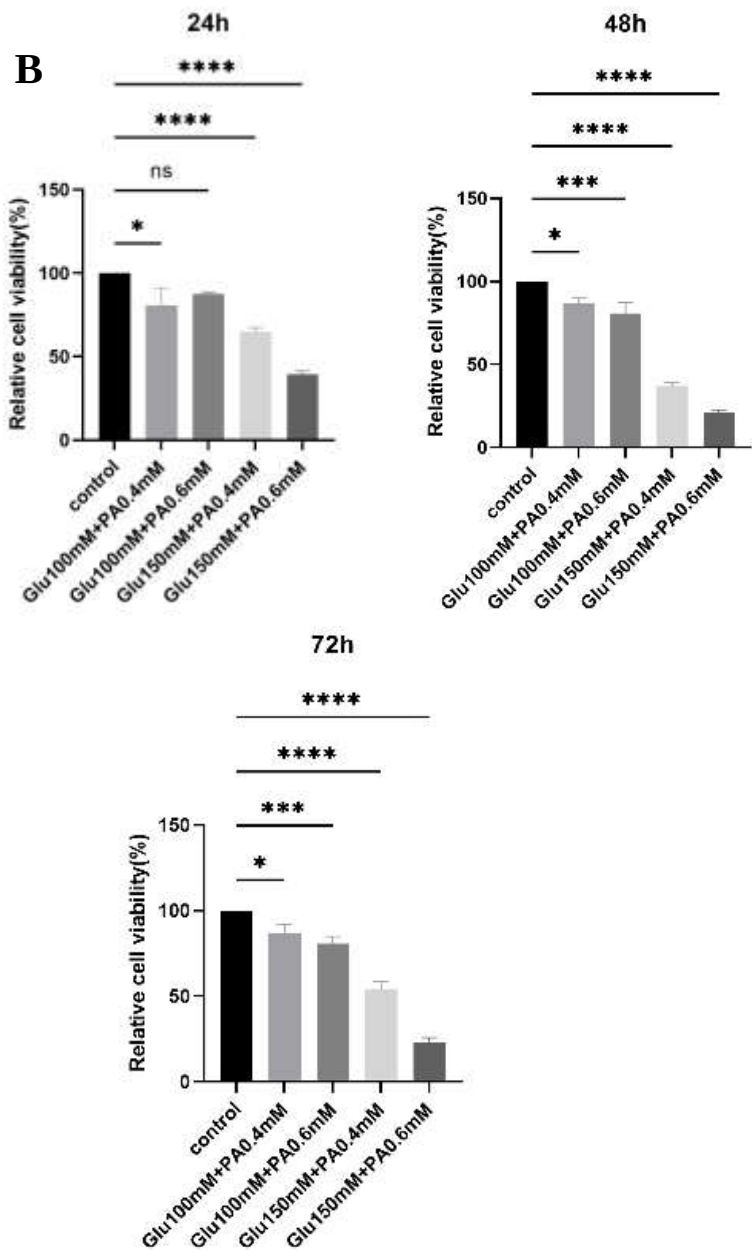


混合培养下施万细胞生长情况



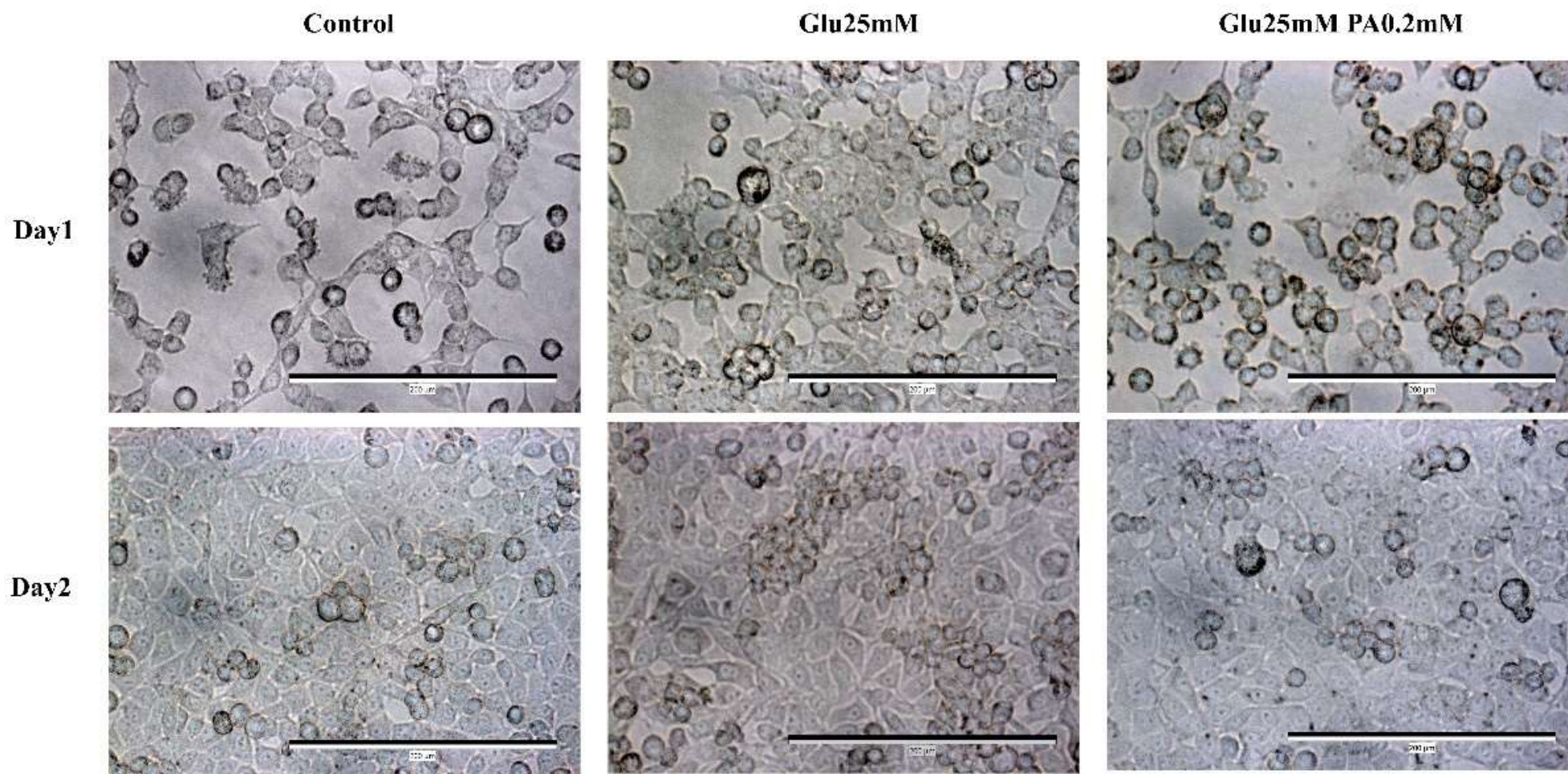
混合培养下施万细胞生长情况和存活率

150mM葡萄糖浓度的细胞存活率比100mM葡萄糖浓度组更低，且0.6mM棕榈酸浓度的细胞存活率也低于0.4mM棕榈酸浓度组。





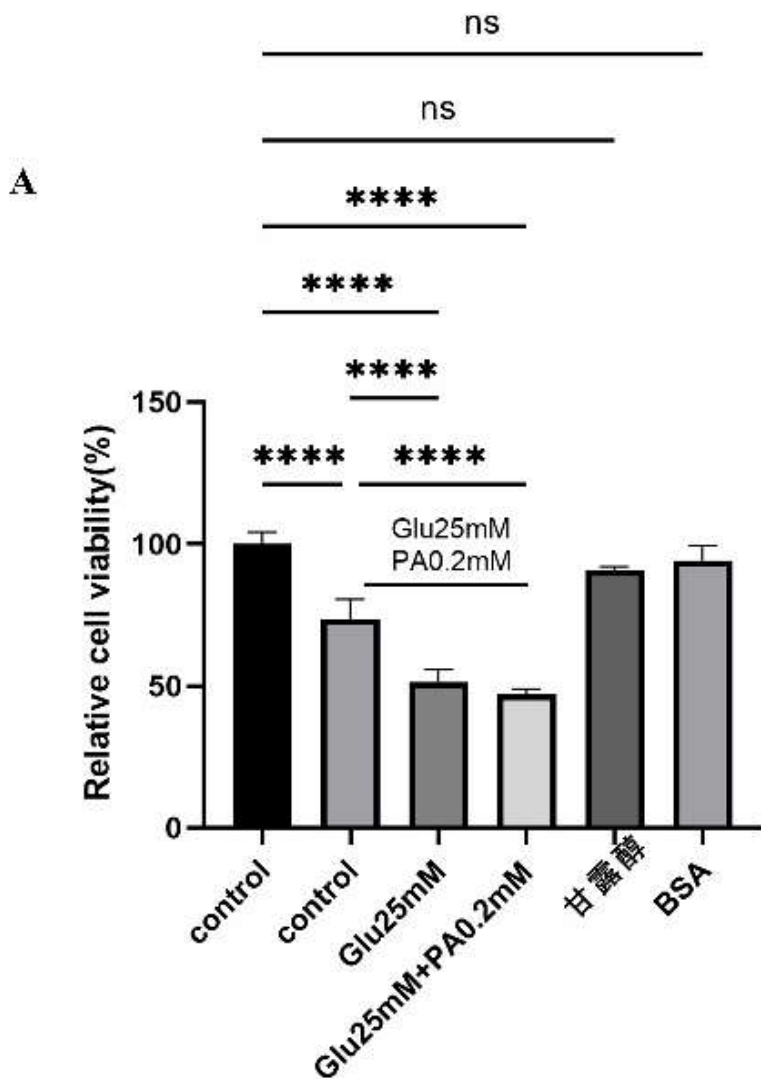
施万细胞长期高糖高脂培养过程





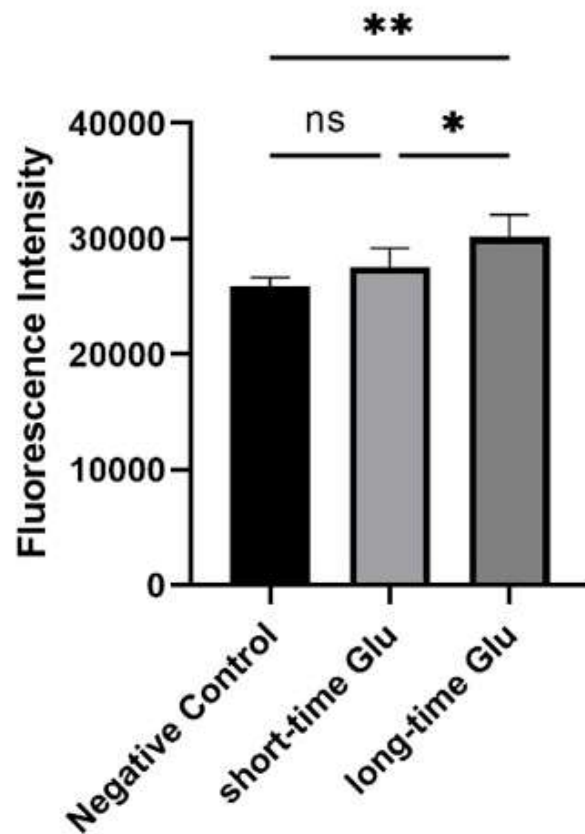
施万细胞模型建立检测

- 高糖高脂刺激组细胞存活率显著下降
- 长期高糖高脂组存活率相比短期刺激组进一步下降



A: CCK-8检测施万细胞在不同条件下的细胞存活率

B



B: 不同培养条件下施万细胞ROS水平

- 短期高糖高脂组ROS水平升高不明显
- 长期高糖高脂组ROS水平进一步提升



背根神经节高糖高脂培养

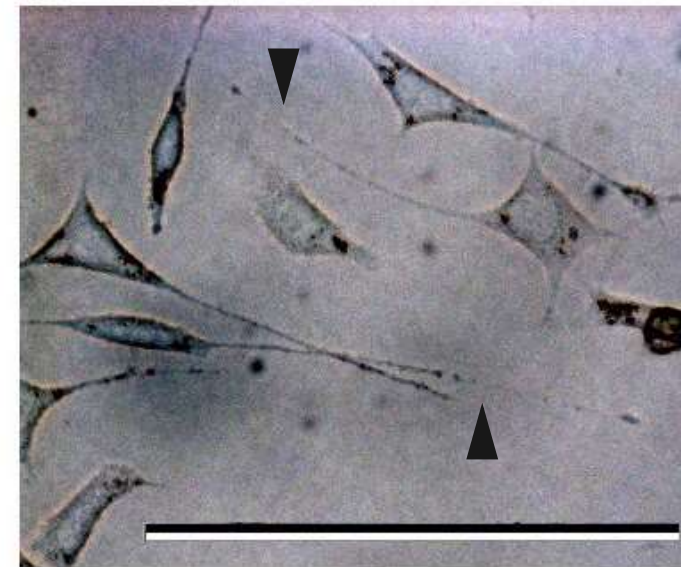
Control



Glu 25mM



Glu 25mM PA 0.2mM



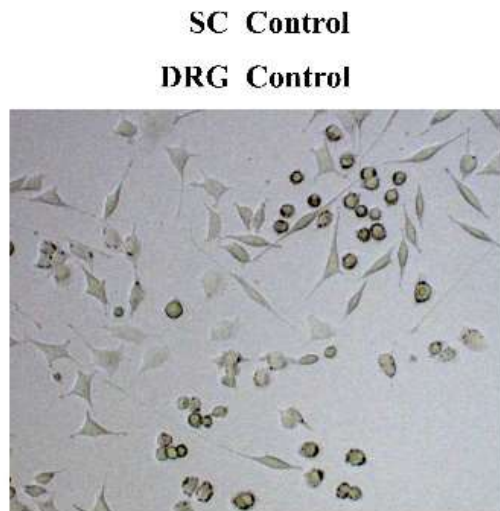
背根神经节（DRG）高糖高脂培养下的轴突损伤

标尺：200 μ m；Control: 完全培养基组；Glu25mM: 含 25mM Glu 完全培养基；Glu25mM PA0.2mM: 含25mM Glu+0.2mM PA 完全培养基；箭头所指为轴突损伤处

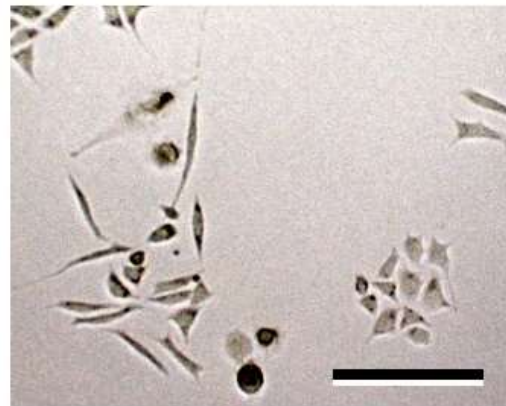


共培养下背根神经节生长情况

Normal



Glu25mM PA0.2mM

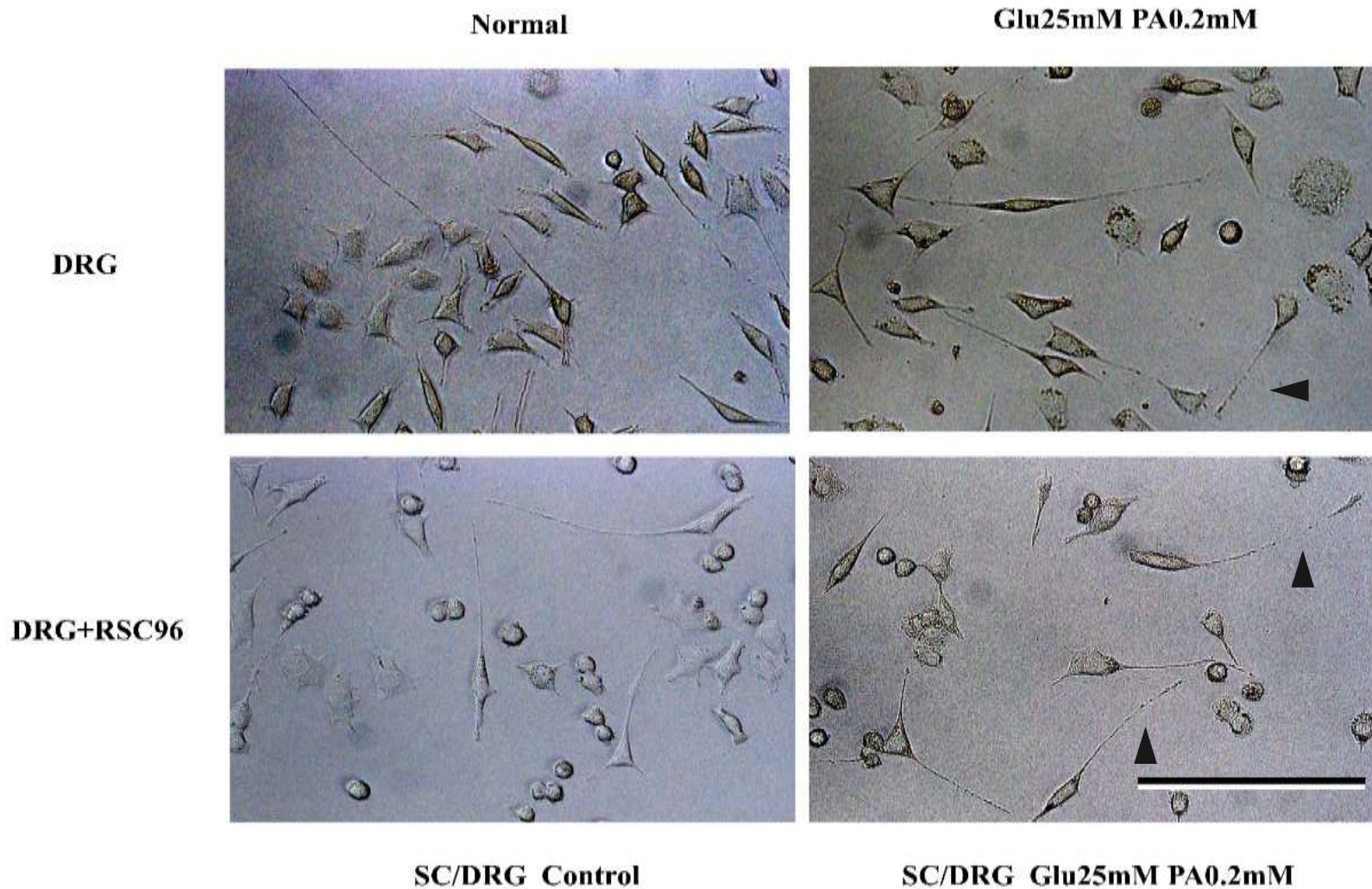


不同条件的施万细胞和DRG共培养下轴突损伤情况

标尺200 μ m; SC、DRG为细胞预先处理条件，左侧为共培养下培养基成分；Control/Normal: 完全培养基组；Glu25mM PA0.2mM: 含25mM Glu+0.2mM PA的完全培养基；施万细胞呈圆型，DRG呈多角型或梭型，有轴突



共培养下背根神经节生长情况



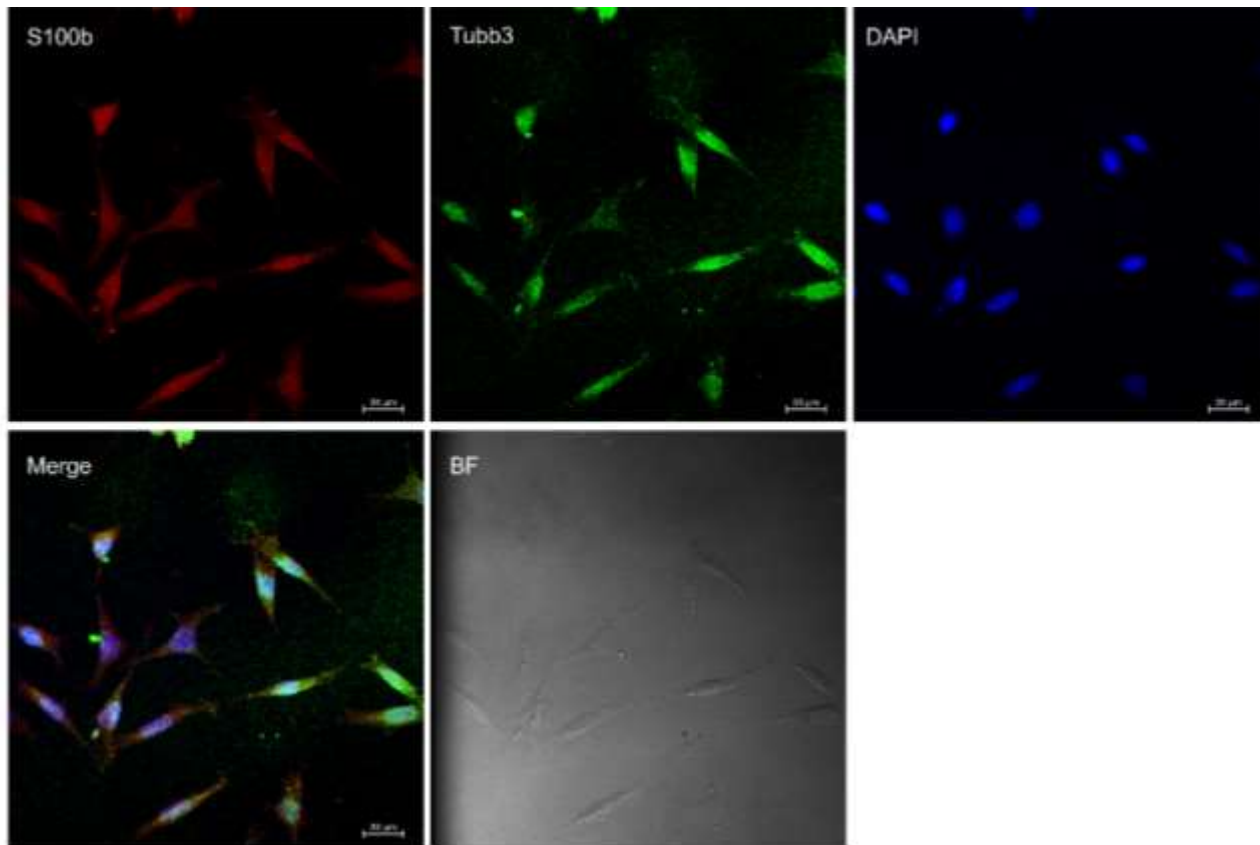
施万细胞和DRG高糖高脂共培养下轴突损伤情况

标尺：200 μ m；第一行为DRG单独培养，第二行为DRG/SC共培养；上方为共培养的培养基成分，下方为细胞预处理条件；Control: 完全培养基组；Glu25mM PA0.2mM: 含25mM Glu+0.2mM PA完全培养基；施万细胞呈圆型，DRG呈多角型或梭型，有轴突；箭头所指为轴突损伤处

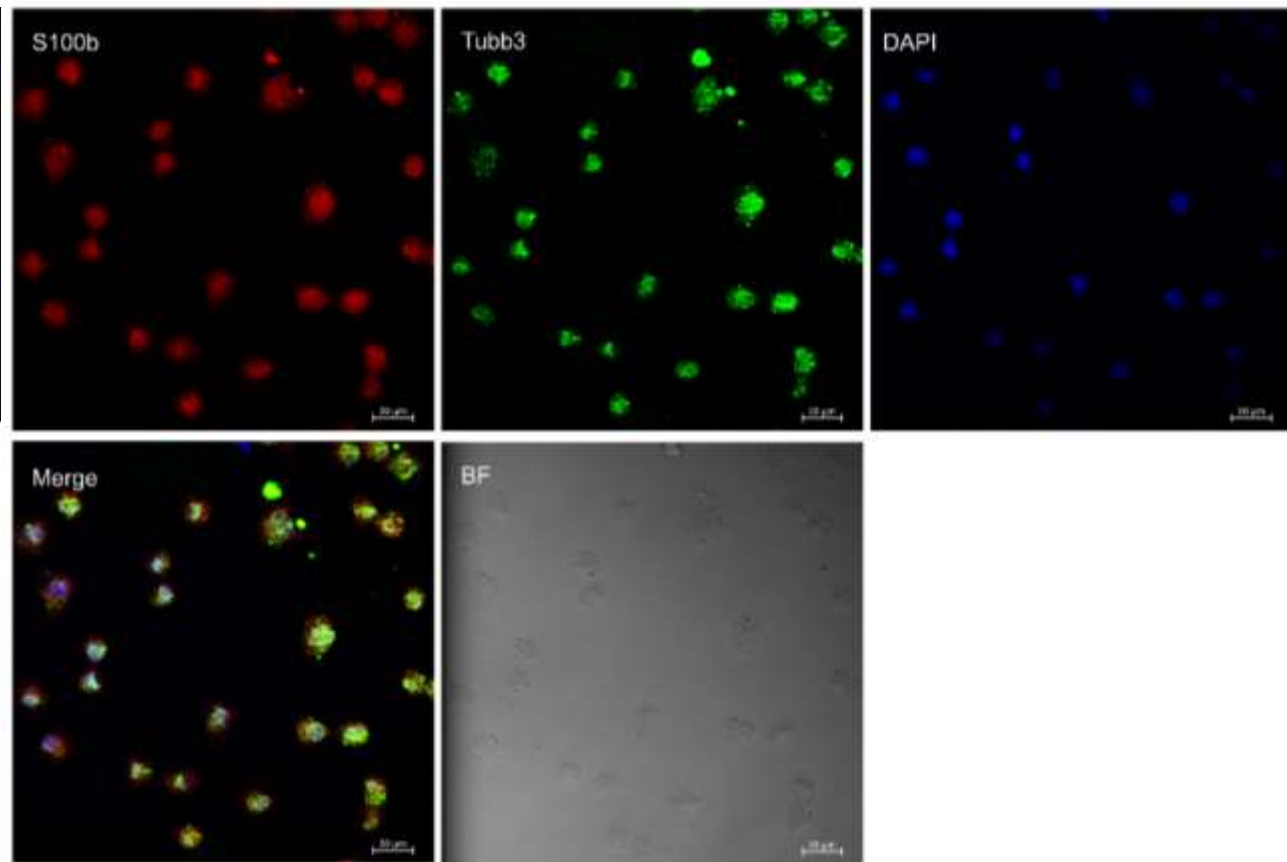


共培养下背根神经节生长情况

DRG: Normal



SC: Normal



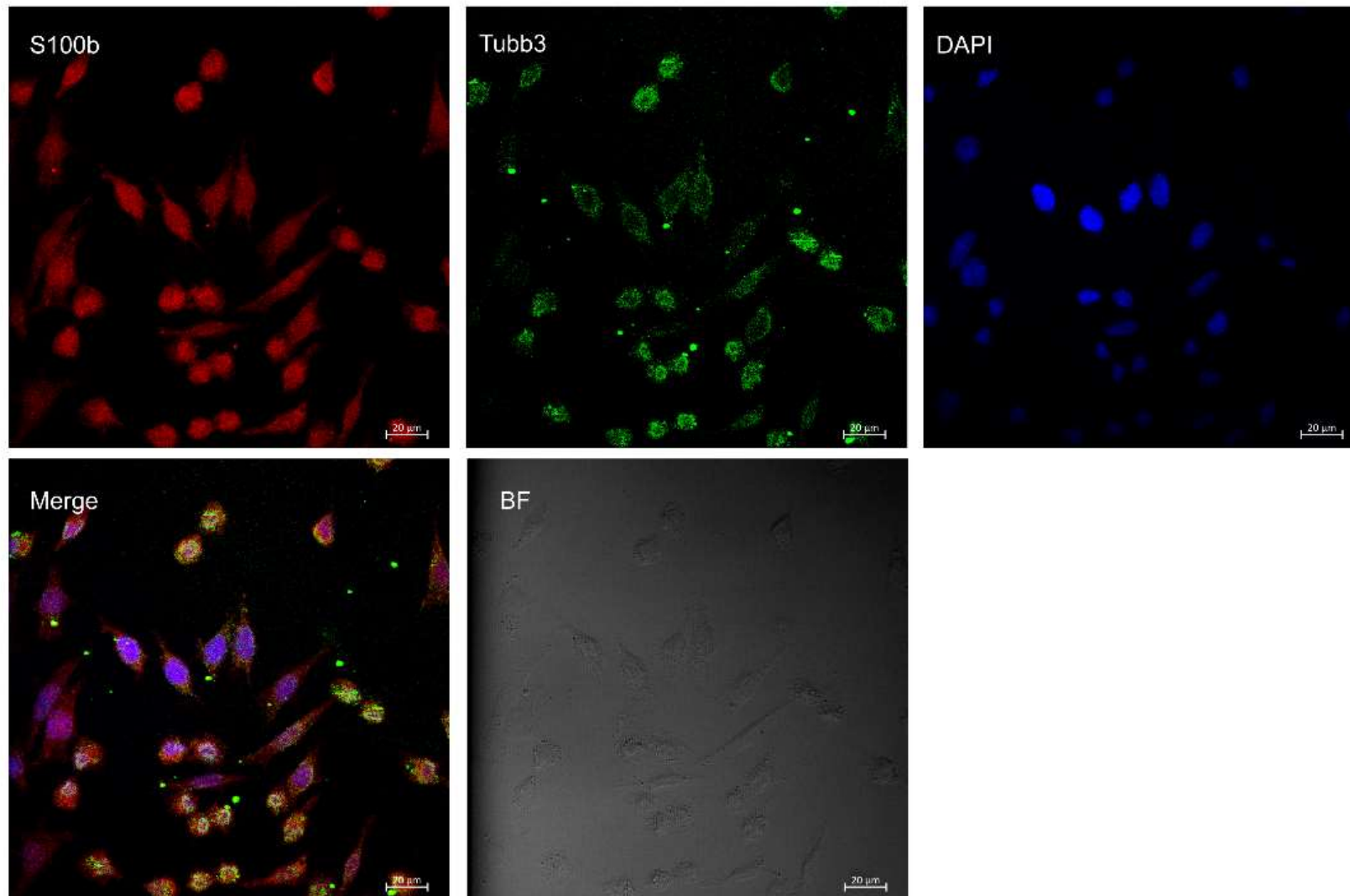
施万细胞和DRG共培养下生长情况

标尺: 20 μ m; S100b: S100钙结合蛋白B, 红色荧光; Tubb3: 微管蛋白 β 3, 绿色荧光; DAPI: 细胞核标志物, 蓝色荧光; Merge: 三色荧光通道叠加; BF: 明场通道



共培养下背根神经节生长情况

SC/DRG:Normal



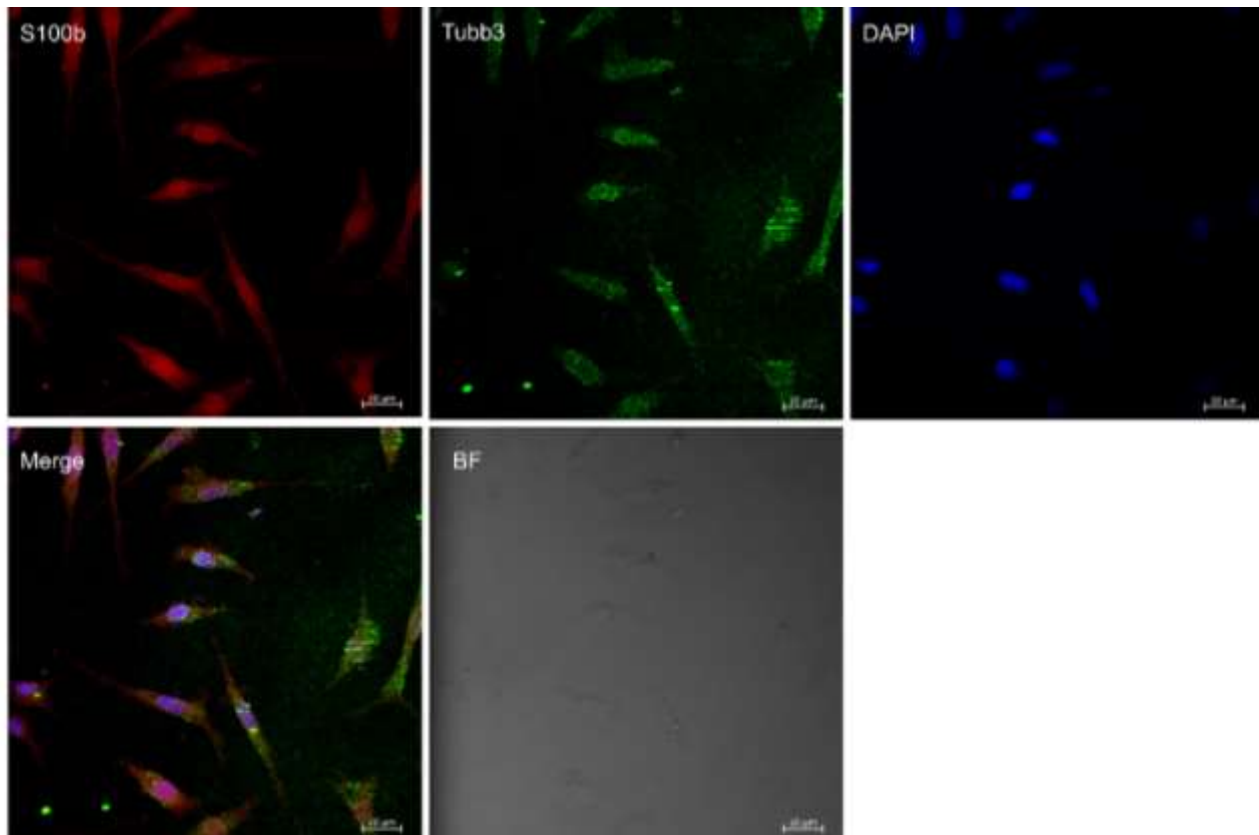
施万细胞和DRG共培养下生长情况



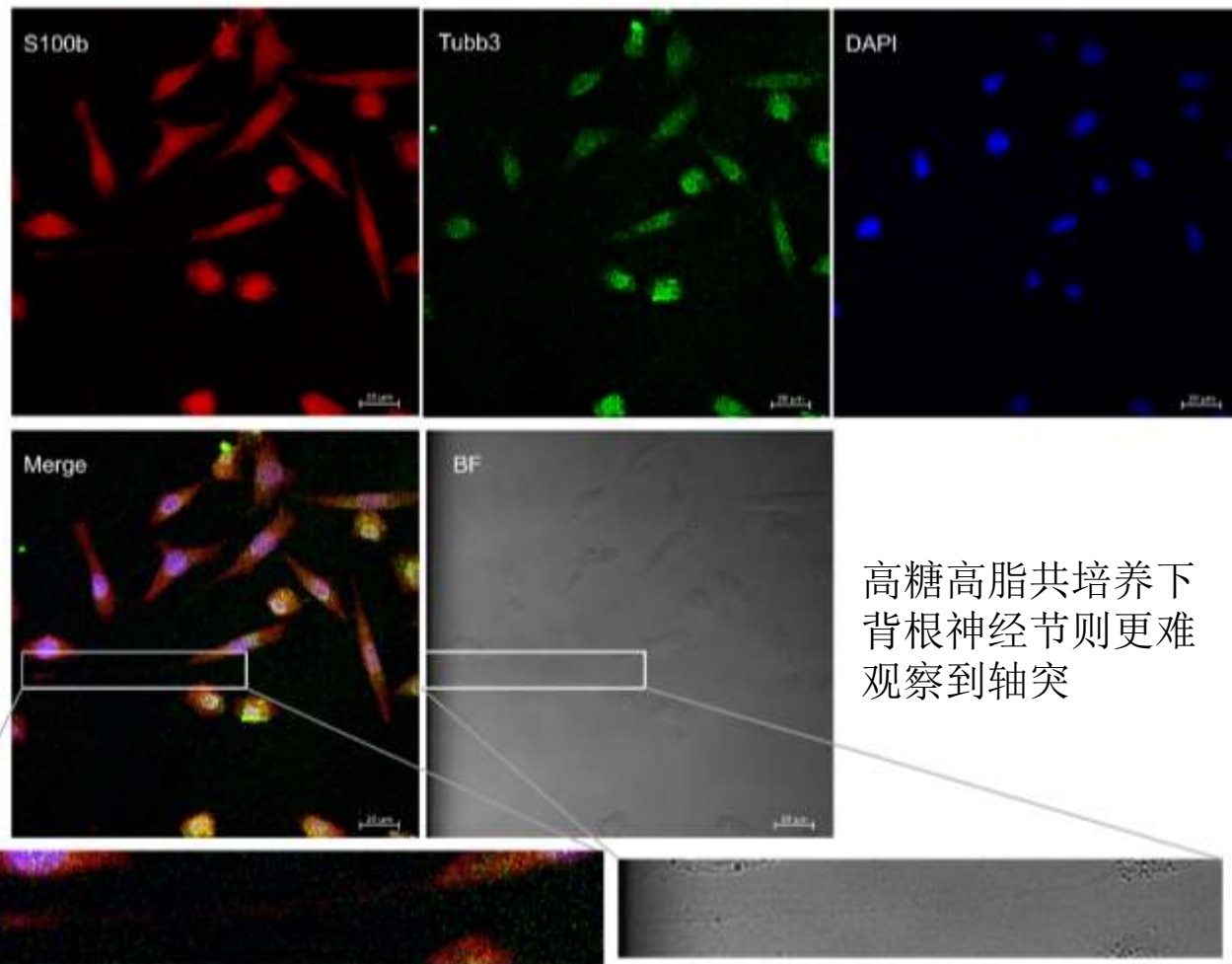
共培养下背根神经节生长情况

DRG: Glu25mM+PA0.2mM

SC/DRG: Glu25mM+PA0.2mM



高糖高脂条件下单独培养的背根神经节轴突未能被Tubb3 明显标记



高糖高脂共培养下背根神经节则更难观察到轴突

施万细胞和DRG共培养下生长情况

标尺: 20 μ m; S100b: S100钙结合蛋白B, 红色荧光; Tubb3: 微管蛋白 β 3, 绿色荧光; DAPI: 细胞核标志物, 蓝色荧光; Merge: 三色荧光通道叠加; BF: 明场通道

03

结论与展望

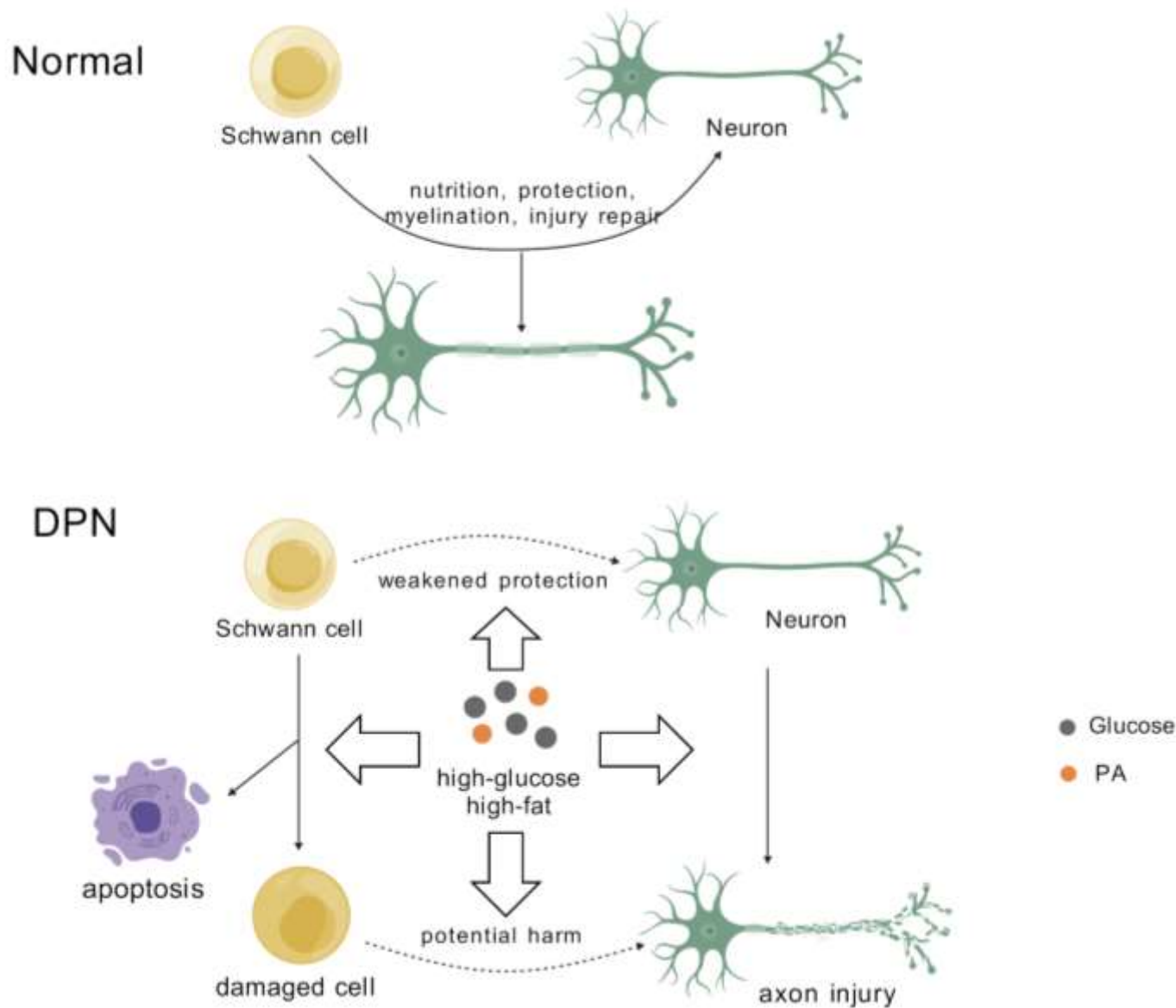


研究总结

- 长期高糖高脂培养模型
(Glu25mM+PA0.2mM)
- 高糖高脂环境下施万细胞帮助DRG轴突再生的能力可能减弱

展望

- 为DPN的发生发展机制提供了新视角
- 改善施万细胞功能可能是DPN的潜在治疗方向



施万细胞和DRG互作关系模式图



参考文献

1. Zhu J, Hu Z, Luo Y, et al. Diabetic peripheral neuropathy: pathogenetic mechanisms and treatment [J]. *Front Endocrinol (Lausanne)*, 2023, 14: 1265372.
2. Wang X, Xu G, Liu H, et al. Inhibiting apoptosis of Schwann cell under the high-glucose condition: A promising approach to treat diabetic peripheral neuropathy using Chinese herbal medicine [J]. *Biomed Pharmacother*, 2023, 157: 114059.
3. Li J, Guan R, Pan L. Mechanism of Schwann cells in diabetic peripheral neuropathy: A review [J]. *Medicine (Baltimore)*, 2023, 102(1): e32653.
4. Cheng Y C, Chu L W, Chen J Y, et al. Loganin Attenuates High Glucose Induced Schwann Cells Pyroptosis by Inhibiting ROS Generation and NLRP3 Inflammasome Activation [J]. *Cells*, 2020, 9(9).
5. Li R, Wu Y, Zou S, et al. NGF Attenuates High Glucose-Induced ER Stress, Preventing Schwann Cell Apoptosis by Activating the PI3K/Akt/GSK3 β and ERK1/2 Pathways [J]. *Neurochem Res*, 2017, 42(11): 3005–3018.
6. Hu M, Jiang W, Ye C, et al. Honokiol attenuates high glucose-induced peripheral neuropathy via inhibiting ferroptosis and activating AMPK/SIRT1/PGC-1 α pathway in Schwann cells [J]. *Phytother Res*, 2023, 37(12): 5787–5802.
7. Tang W, Chen X, Liu H, et al. Expression of Nrf2 Promotes Schwann Cell Mediated Sciatic Nerve Recovery in Diabetic Peripheral Neuropathy [J]. *Cell Physiol Biochem*, 2018, 46(5): 1879–1894
8. Jia L, Liao M, Mou A, et al. Rheb-regulated mitochondrial pyruvate metabolism of Schwann cells linked to axon stability [J]. *Dev Cell*, 2021, 56(21): 2980–2994.e2986.

请各位老师批评指正