

Rb α Arc | Ch α GFP | Protocol
06/01/22

DAY 1

1. **Wash x3:** 10min, RT, 1X PBS
2. **Block x1:** 2hr, RT, 10% NDS in 1X PBST (0.5% Triton X-100)
3. **Primary Abs x1:** 24hr, 4°C, 10% NDS in 1X PBST
 - a. Rb α Arc
 - i. 1:1,000
 - ii. SySy 156 003; Rb polyclonal
 - b. Ch α GFP
 - i. 1:2,000
 - ii. Abcam ab13970; Ch polyclonal

DAY 2 (1 day later)

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| <ol style="list-style-type: none">6. Wash x3: 10min, RT, 1X PBS7. Secondary Abs x1: 2hr, RT, 1X PBS<ol style="list-style-type: none">a. Dk α Rb<ol style="list-style-type: none">i. 1:500ii. Alexa Fluor 647b. Dk α Ch<ol style="list-style-type: none">i. 1:250ii. Cy28. Wash x3: 5min, RT, 1X PBS | <p style="text-align: center;">***OPTIONAL***</p> <ol style="list-style-type: none">9. Nuclear Stain x1: 15min, RT, 1X PBS<ol style="list-style-type: none">a. Hoechst stain<ol style="list-style-type: none">i. 1:10,00010. Wash x3: 5min, RT, 1X PBS |
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