

General Procedure for Photorelease

This general protocol for the photorelease may be used as a starting point. For optimal result slight tuning of experimental conditions might be required.

1. Resuspend the washed resin in 1ml of PBS and transfer to a clear glass vial or quartz cuvette with a tight fitting cap.
2. Irradiate the resin suspension with light at 345-375nm with constant agitation for 1hr. This can be done using hand held long wave UV lamp such as a UVGL-25.¹
3. Agitate the sample at 37°C for 1hr after irradiation. Avoid using a stir bar as this can crush some resins.
4. Collect the eluant by centrifugation or using an empty spin column.
5. Resuspend the resin in 1ml of PBS and agitate for 2-16hrs. For more efficient recovery of enriched protein(s), use a buffer containing 0.1-1% detergent and/or 250mM - 1M NaCl.
6. Collect the second elution by centrifugation or using an empty spin column.

Troubleshooting

Problem	Possible Cause	Solution
Poor Photorelease	Light is not sufficiently intense	Use a lamp with a higher intensity.
	Incorrect wavelength of light	Ensure that the lamp is outputting light in the 345-368nm range.
	Insufficient agitation	Ensure that the beads are being properly mixed during photorelease
	Strong non-specific interactions	Consider using a detergent during photorelease or including more wash steps after photorelease

¹ For more efficient photorelease and shorter irradiation time following UV-Lamps can be used:

<http://uvp.com/3uvlamps.html>

<http://www.uvsystems.com/store/product.php?productid=16135&cat=250&page=1>

<http://www.uvsystems.com/store/product.php?productid=16193&cat=251&page=1>

Selected References:

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