



Supporting raw data for the western blotting analysis. (a) BMP-2, (b) Runx2, (c) OPN, (d) β -actin.

We followed the traditional protocol in the implementation of western blotting experiments and there are several experimental links that we think it is necessary to explain.

1) In the experiment, we used the pre-stained protein marker which ultimately did not appear on the X-ray film (in fact, the pre-stained protein marker has almost completely replaced the traditional protein marker in the past ten years).

2. The use of pre-stained protein marker is mainly conducive to saving antibodies, ECL and other related reagents. We generally do not incubate the entire PVDF membrane. Instead, after the transfer is completed, we evaluate the degree of completion of the transfer using ponceau S stain, and then the location of the target band was evaluated by the pre-stained protein marker. Then we cut out the PVDF membrane with a height of about 1cm above and below the location of the target bands for subsequent experiments.