Macro collection for the estimation of molecular weight in gels

This is a collection of 4 macros for the estimation of molecular weight in gel electrophoresis.
At least 2 reference bands with known molecular weight are required for setting up the calibration curve. If more than 2 references are entered, the algorithm calculates the regression line for the coordinates distance from start and natural logarithm of molecular weight.

Installation:
Copy the file ***MolWt.txt*** into the macro folder of ***ImageJ*** and do Install (***Plugins -> Macros-> Install***, select **MolWt.txt** from the macro folder). The macros were successfully tested with ***ImageJ Vers. 1.34*** (Windows) ***.***

Procedure:
The estimation is performed in the density plots derived from ***Analyze -> Gels ...***
The whole procedure is divided into 4 Steps. All 4 required macros are part of file *MolWt.txt*.

- **Before starting this procedure, please be shure that the result and log windows are empty or closed!**
- **Allow 4 decimal places for the result window (please see *Analyze -> Set Measurements, Box Decimal Places*).**

1. Step

Run macro ***Open MW File*** (or simply press b). This opens the file selection dialog box. Select the .txt file which is containing the molecular weights of your reference bands. This file must have the .txt extension. The file must be written in the format like

210000
106000
50000
10000
No other information, just the molecular weights of the reference bands, one per row. For generating such a file, please use an editor (i.e. *Notepad*).
These values are copied to the **log** window by the macro.

2. Step

- Now select the ***point selection tool*** from the icon bar.
- In the density plots derived from the gel analysis module, click to one of the peaks originated from a reference band.
- Run now macro ***Select Reference Bands*** (or simply press I). A dialogbox is popping up showing the molecular weights of all reference bands in the pull down box.
- Select the appropriate value and press **OK.**
- Repeat this for every reference peak.

The peaks become annotated with the corresponding molecular weight.
In the result window, you will find the peak coordinates and corresponding molecular weight of the reference bands. Peaks selected by mistake may be cleared in the result window.

3. Step

Run macro ***Calculate Regression Line*** (or simply press q).
In the result window there are now 3 new columns: ***slope, intercept*** and ***Corr.***, containing the constants of the calculated regression line in the first row. All other cells of these columns are filled with 0. Corr. is the correlation coefficient indicating the quality of the fit. An absolutely perfect fit would result in -1.

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4. Step

- Select the ***point selection tool*** from the icon bar.
- In the density plots, click to a peak whose molecular weight is to be estimated.
- Run macro ***Estimate unknown MW*** (or simply press y).
- The peak becomes annotated
- Repeat this for all interesting peaks

THAT'S IT !

In principle the above procedure can also be applied to the gel image directly. However you have to turn the image first, so that the sample slots are seen on the left side. Cut off those parts form the image which are left from the sample slots or sample start and carry out steps 1 to 4 but click to the desired bands in the gel image.

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