Supplementary Data

Characterization of Recombinant Human Lactoferrin Expressed in *Komagataella Phaffii*

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Supplementary Data

Supplementary figures

Fig. S1.

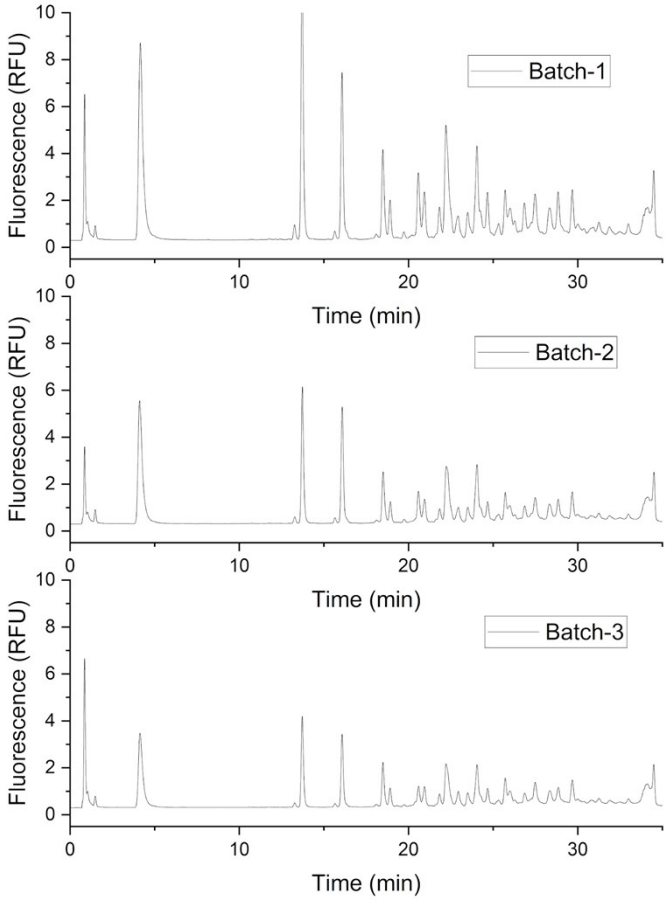


Fig. S1. The HPLC-fluorescence chromatograms of the released N-glycan profiles of three batches of Helaian rhLF. The glycans were released, labeled by InstantPC and measured by UHPLC-Fluoresence detection as described in the Material and Methods section.

Supplementary Data

Fig. S2.

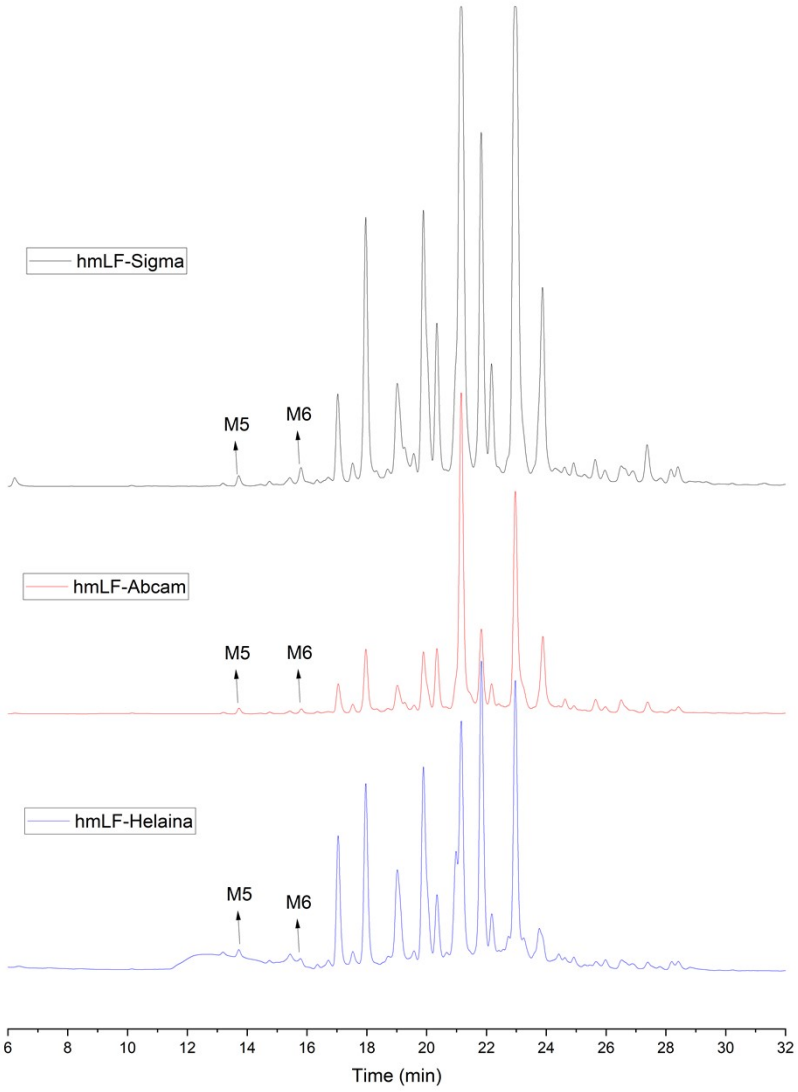
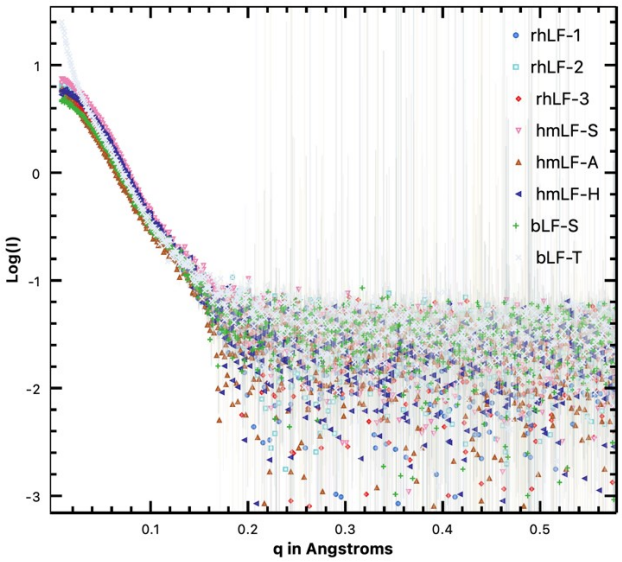


Fig. S2. Comparison of the released N-glycan profiles of native hmLF from three different sources including MilliporeSigma (hmLF-Sigma), Abcam (hmLF-Abcam) and Helaina (hmLF-Helaina). The N-glycans having 5 and 6 mannoses (M5 and M6) were indicated by arrows. The glycans were released, labeled by InstantPC and measured by UHPLC-Fluoresence detection as described in the Material and Methods section.

Supplementary Data

Fig. S3

(A)



(B)

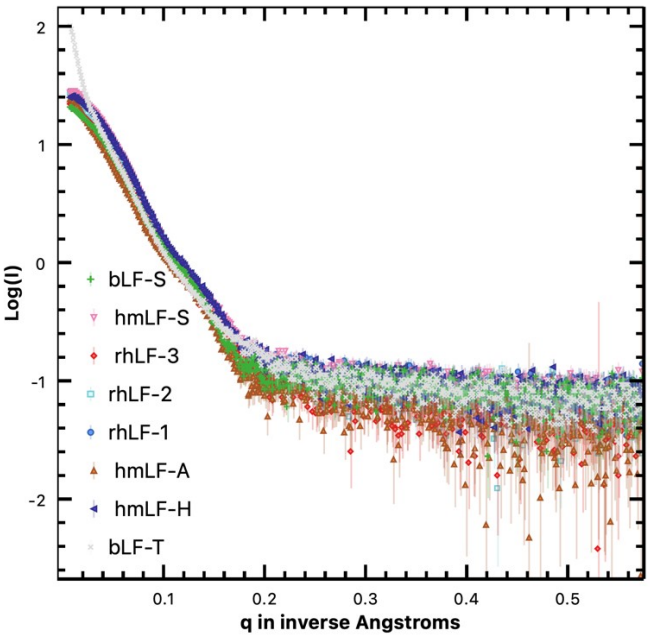


Fig. S3. SAXS raw data of Helaina rhLF (three batches), native hmLF (three sources), and native bLF (two sources) acquired at 1mg/ml (A, top) and 4mg/ml (B, bottom). Helaina rhLF-1, -2, and -3 indicate three batches of Helaina rhLF. hmLF-A, -S, and -H are native hmLF from Abcam, MilliporeSigma, and Helaina, respectively. bLF-S and -T are native bLF supplied by MilliporeSigma and The Lactoferrin Company. Conditions for data acquisition were detailed in the Material and Methods.

Supplementary Data

Fig. S4.

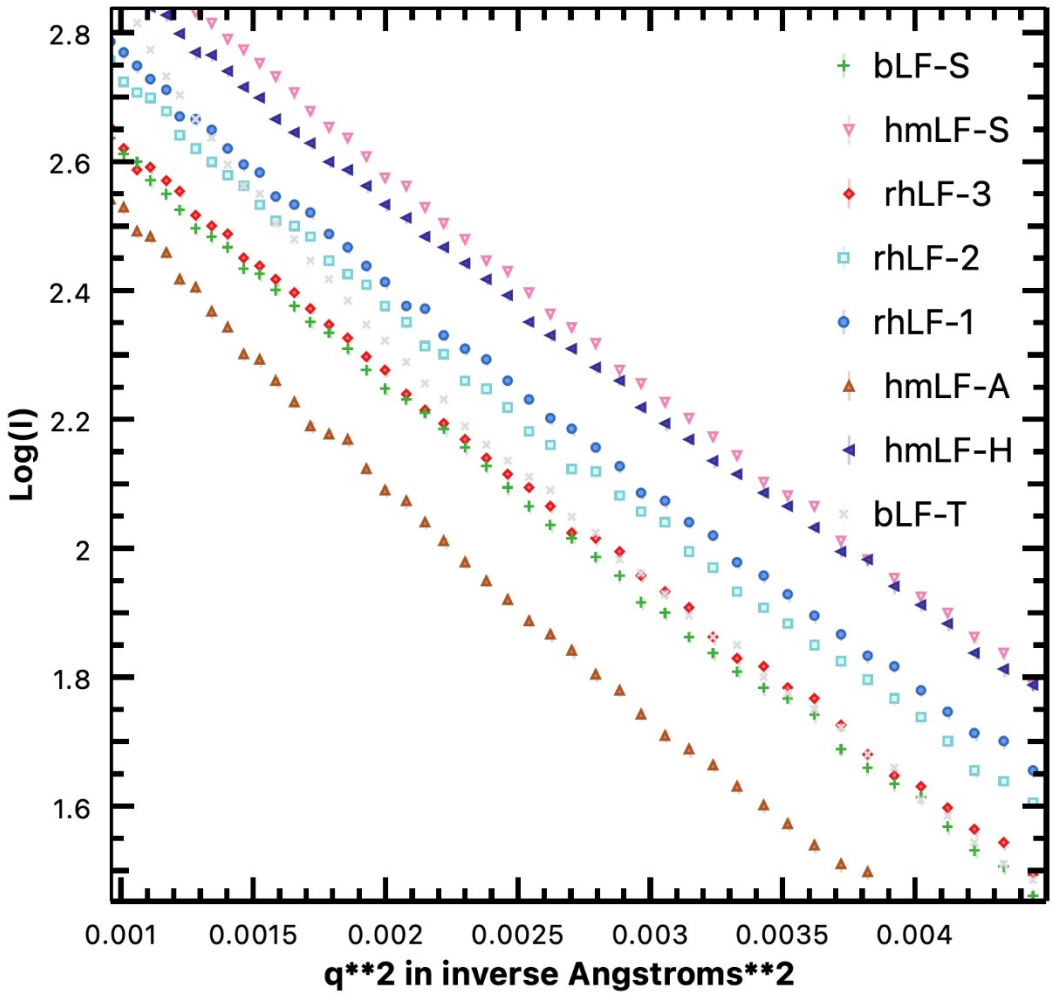


Fig. S4. Guinear plots of Helaina rhLF (three batches), native hmLF (three sources), and native bLF (two sources) derived from the SAXS raw data acquired at 4 mg/mL (Fig. 3S). The sample labels are the same as in Fig. S3. Conditions for data acquisition were detailed in the Material and Methods.

Supplementary Data

Fig. S5.

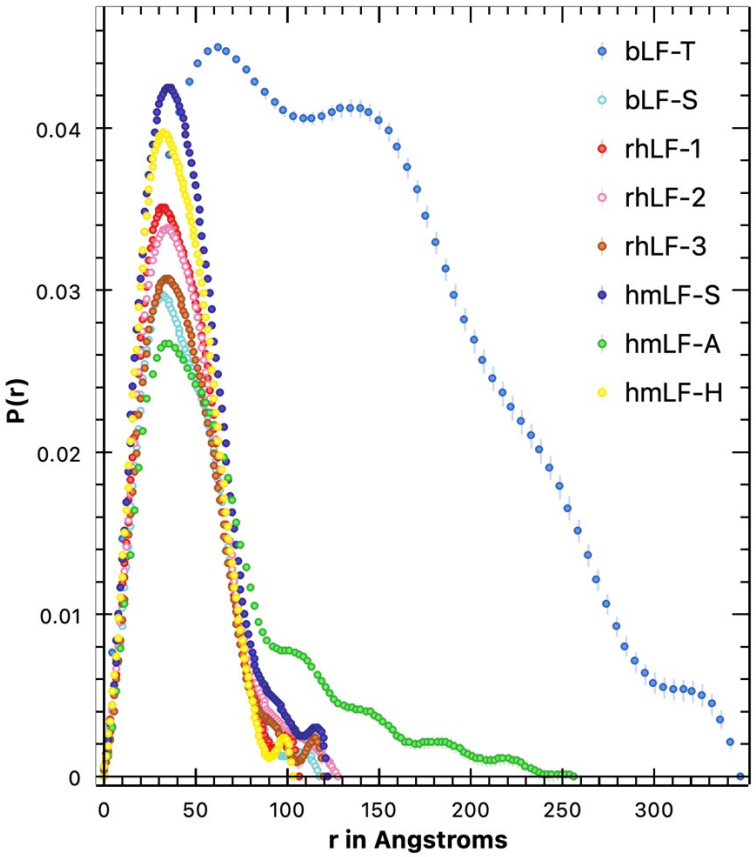


Fig S5, Pair distance distribution function P(r) plot of Helaina rhLF (three batches), native hmLF (three sources), and native bLF (two sources) derived from the SAXS raw data acquired at 4 mg/mL (Fig. 3S). The sample labels are the same as in Fig. S3. Conditions for data acquisition were detailed in the Material and Methods.

Supplementary Data

Fig. S6.

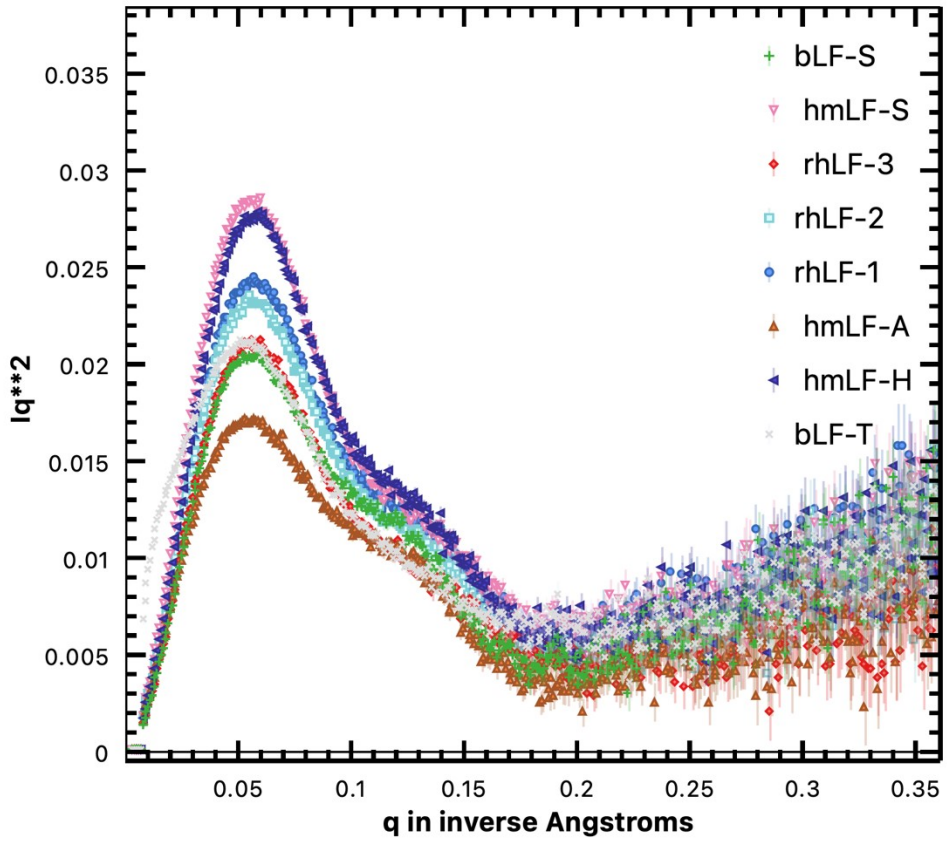
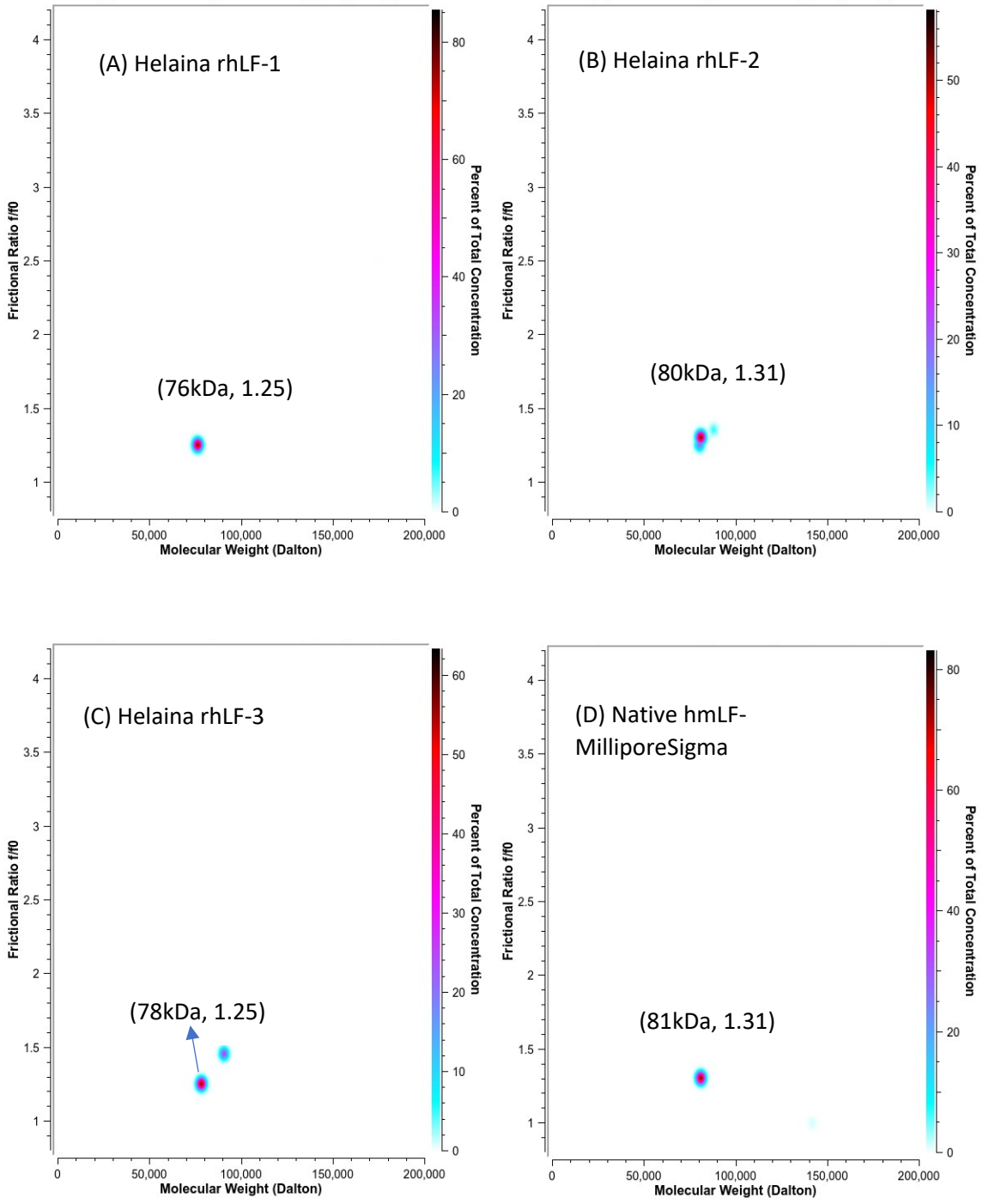


Fig S6, Kratky plots of Helaina rhLF (three batches), native hmLF (three sources), and native bLF (two sources) derived from the SAXS raw data acquired at 4 mg/mL (Fig. 3S). The sample labels are the same as in Fig. S3. Conditions for data acquisition were detailed in the Material and Methods.

Supplementary Data

Fig. S7.



Supplementary Data

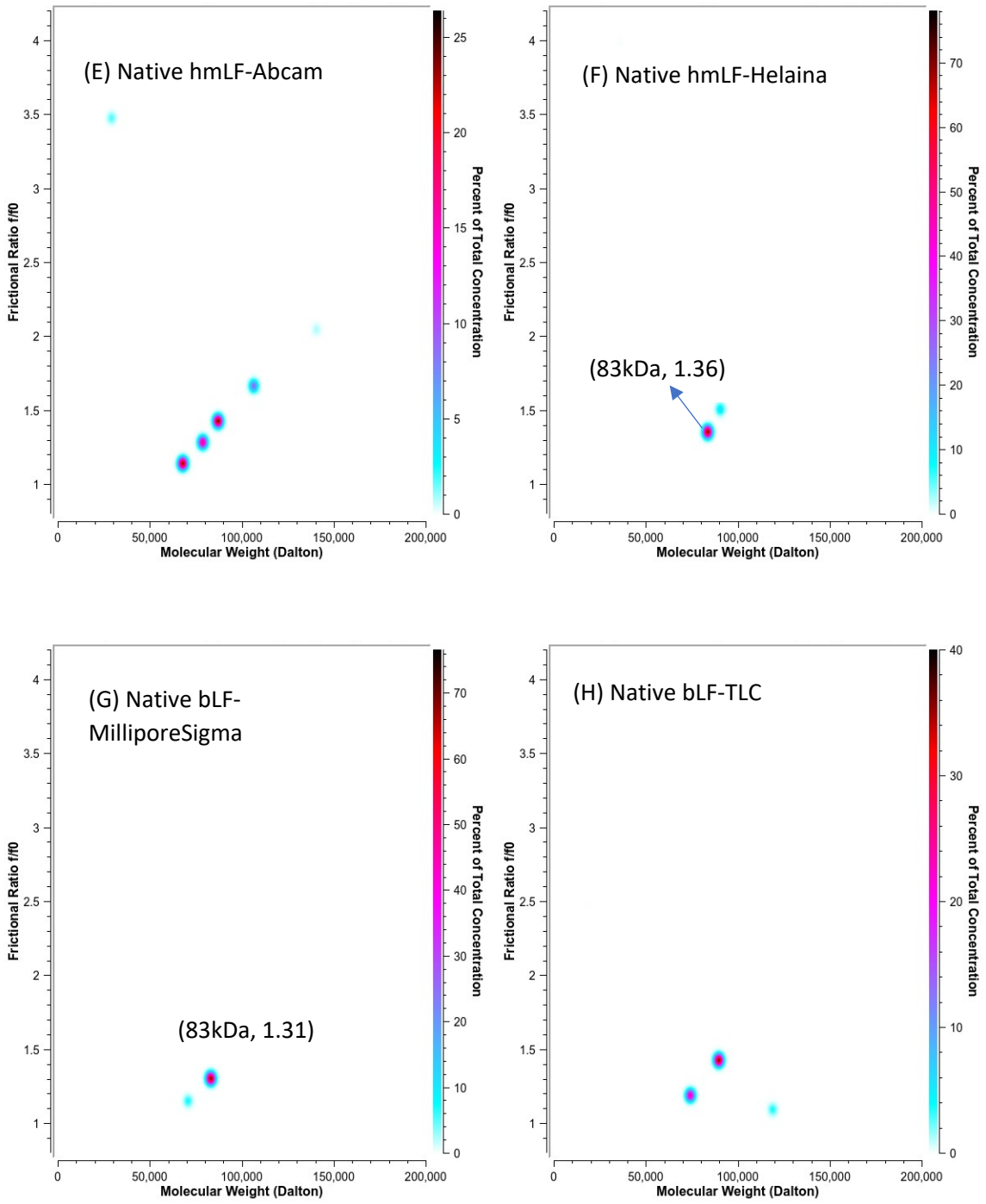


Fig. S7, AUC data of LF protein samples studied at 4 mg/mL concentration. Conditions for data acquisition and processing were detailed in the Material and Methods. The data presented a pseudo-three-dimensional distribution of the observed protein species, with the calculated molecular weight on the x-axis, the calculated frictional ratio on the y-axis, and the % concentration in the Z-plane, with the heat map on the right axis. Conditions for data acquisition and processing were detailed in the Material and Methods.