

## What is Tacsimate™?

Tacsimate is a unique crystallization reagent developed exclusively by Hampton Research. Tacsimate is composed of a mixture of titrated organic acid salts. Tacsimate contains 1.8305 M Malonic acid, 0.25 M Ammonium citrate tribasic, 0.12 M Succinic acid, 0.3 M DL-Malic acid, 0.4 M Sodium acetate trihydrate, 0.5 M Sodium formate, and 0.16 M Ammonium tartrate dibasic<sup>1</sup>. This mixture is titrated to the appropriate pH using sodium hydroxide and is available in pH 4, 5, 6, 7, 8, or 9 reagent formulations.

Tacsimate was developed by Hampton Research Corp. as an extension to the original work described by Prof. Alexander McPherson in the publication “A comparison of salts for the crystallization of macromolecules”, *Protein Science*, 10: 418-422, 2001. In a subsequent set of experiments, Hampton Research formulated a series of test reagents based on solutions containing each of the seven individual organic acids at six different concentrations and five different pH levels as well as six different combinations of the organic acids at five different pH levels. This screen was then tested against a portfolio of 36 unique proteins and vapor diffusion crystallization. A control set of reagents consisting of Crystal Screen and Grid Screen PEG were also set with this same portfolio of proteins. The experiment was designed to evaluate the effectiveness of using a combination of small molecules in a single crystallization reagent as well as evaluate the usefulness of various organic acids in addition to malonate versus pH. For example, a mixture of seven salts would reduce the number of screen solutions seven fold, reducing sample requirements, reagent cost, set up time, the number of crystallization plates as well as improve efficiency. The experimental results demonstrated one particular combination of titrated organic acids produced more hits per protein and more overall hits than any other single salt or mixture of salts. This mixture of titrated organic acids is the formulation of Tacsimate.

How should I optimize a hit from Tacsimate? The same way one would optimize from any salt based hit. Vary the concentration of Tacsimate as well as the pH. One may choose to either use a buffer with Tacsimate or use one of the Tacsimate solutions titrated to pH 4, 5, 6, 7, 8, or 9. Follow this primary optimization with an evaluation of other crystallization variables such as temperature, additives, seeding, etc.

What cryo reagent should be used with Tacsimate? Tacsimate, like other neutralized organic acids can behave as a cryosalt. One may screen slightly higher concentrations of Tacsimate when pursuing cryo protection conditions. Try a Tacsimate concentration 15 to 20% higher than that used in the reagent well to produce the crystal. Tacsimate is compatible with other cryo reagents, so one may also use glycerol, ethylene glycol and other cryogenic agents in conjunction with Tacsimate when optimizing cryo protection conditions.

What published work is available for further information about the use of neutralize or titrated organic acids such as malonate and Tacsimate?

1. Searching for silver bullets: An alternative strategy for crystallizing macromolecules. Alexander McPherson and Bob Cudney. *Journal of Structural Biology* 156 (2006) 387–406.
2. A novel strategy for the crystallization of proteins: X-ray diffraction validation. Steven B. Larson, John S. Day, Robert Cudney and Alexander McPherson. *Acta Cryst.* (2007). D63, 310–318.
3. A comparison of salts for the crystallization of macromolecules. Alexander McPherson. *Protein Science* (2001), 10:418-422.

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