Yale University

VP ITC - The Basics

The VP-ITC Unit directly measures heat evolved or absorbed in liquid samples as a result of mixing precise amounts of reactants. A spinning syringe is utilized for injecting and subsequent mixing of reactants. Spin rates are user selectable.

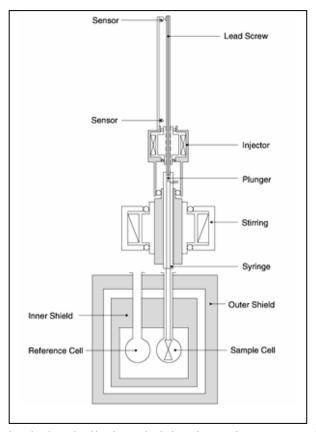
The normal operating range is 2°C to 80°C.

Wetted cell surfaces are Hastelloy® Alloy C 276.

Sample and reference cells are accessible for filling and cleaning through the top of the unit. The sample cell is on the right as one faces the front of the unit.

A pair of identical coin shaped cells is enclosed in an adiabatic Outer Shield (Jacket). Access stems travel from the top exterior of the instrument to the cells. Both the coin shaped cells and the access stems are totally filled with liquid during operation. This requires approximately 1.8 ml. per cell even though the working volume of the cell is only 1.4 ml

Temperature differences between the reference cell and the sample cell are measured, calibrated to power units and displayed to the user as well as saved to disk. The data channel is referred to as the DP signal, or the differential power between the reference cell and the sample cell. This signal is sometimes referred to as the "feedback" power used to maintain temperature



equilibrium. Calibration of this signal is obtained electrically by administering a known quantity of power through a resistive heater element located on the cell.

The syringe containing a "ligand" is titrated (injected) into the cell containing a solution of the "macromolecule". An injection which results in the evolution of heat (exothermic) within the sample cell causes a negative change in the DP power since the heat evolved chemically provides heat that the DP feedback is no longer required to provide. The opposite is true for endothermic reactions. Since the DP has units of power, the time integral of the peak yields a measurement of thermal energy, ΔH . This heat is released or absorbed in direct proportion to the amount of binding that occurs. When the macromolecule in the cell becomes saturated with added ligand, the heat signal diminishes until only the background heat of dilution is observed.

With the VP-ITC system the entire experiment takes place under computer control. The user inputs the experimental parameters (temperature, number of injections, injection volumes) and the computer carries out the experiment. Origin software is then used to analyze the ITC data using fitting models to calculate reaction stoichiometry (n), binding constant (K_b), enthalpy (ΔH) and entropy (ΔS).