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TO LABORATORY PERSONNEL RESPONSIBLE FOR WHITE CELL PREPARATIONS FOR THE DIAGNOSIS AND MONITORING OF CYSTINOSIS

Cystinosis is an inherited disease causing progressive kidney failure, retarded growth, and vision problems. When diagnosed early, these effects can be prevented or delayed by proper management and medication. The metabolic defect is the failure of cellular lysosomes to release cystine. As a consequence the free-cystine in the lysosomes accumulates to many times the normal value. The diagnosis of cystinosis is therefore based in part on the measuremnt of free-cystine in the tissues that accumulate this amino acid. This is most easily done in white (not red) blood cells.

The nature of blood cells and of cystine makes shipment of whole blood to us unreliable for diagnosis. Whole blood contains red cells which are rich in glutathione, a compound which will react with cystine. To prevent this the white cells are separated from the red cells. The white cells are kept cold to slow down reactions, then broken open, acidified, and frozen. This prevents the reaction of cystine with -SH compounds such as glutathione and precipitates the cell protein. These steps stabilize the cystine content of the preparation.

The ACD-Dextran procedure first separates most of the white cells from the red cells by density difference. To remove the rest of the red cells, the solution is made hypotonic for 90 sec. This lyses the RBC's but not the WBC's. Finally the pure white cells are frozen and thawed repeatedly to break open not only the cells but also the lysosomes where cystine is stored in cystinotic patients. The lysate is acidified, frozen, and sent to the Cystine Determination Laboratory for assay. We measure the cystine in the liquid phase and the protein in the precipitate.

Factors which make a good preparation and therefore an accurate cystine/protein value:

1. Prompt processing after blood draw. (If processing must be delayed while other samples are drawn, keep the whole blood at room temperature.)

2. Processing without stopping in the middle.

3. Minimum time between water addition to WBC and SSA addition, especially the thawing time. (During thawing the cystine can react with cell contents, cysteine can oxidize and proteins can hydrolyse.)

4. Minimum red cell contamination.

If you have comments or problems, you can include a note with the samples when you ship, or call (619)543-5260.