Exhibit L

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Understanding and Accounting for Method Variability in Whole Effluent Toxicity Applications Under the National Pollutant Discharge Elimination System

Table 3-7. Number of Laboratories Having a Given Percent of Tests Exceeding the PMSD Upper Bound for the Sublethal Endpoint

Test Method	No. Labs	Endpoints[a]	Number of Labs with Various Percentages of Tests Exceeding the PMSD Upper Bound				
			0%	0%-10%	10%-20%	20%-50%	50%-100%
1000.0 Fathead Minnow	19	G	8	2	7	2	0
1002.0 *Ceriodaphnia dubia*	33	R	15	7	5	6	0
1003.0 Green Alga	9	G	6	1	0	2	0
1004.0 Sheepshead Minnow	5	G	3	1	0	1	0
1006.0 Inland Silverside	16	G	6	5	1	4	0
1007.0 Mysid (growth)	10	G	5	2	0	3	0

[a] G = growth, R = reproduction

3.4 Conclusions about Variability of WET Methods

3.4.1 Variability of EC25, LC50, NOEC

For EC25, the quartiles of the within-laboratory CVs ranged across the promulgated methods from 0.09 to 0.45, and the median CV ranged from 0.13 to 0.38. For LC50, the quartiles of the within-laboratory CVs ranged from 0.07 to 0.35, and the median CV ranged from 0.08 to 0.30. For NOEC, the quartiles of the within-laboratory CVs ranged from 0 to 0.61, and the median CV ranged from 0.10 to 0.46. This summary applies to those methods represented by at least 20 tests and three laboratories.

EPA concludes from Tables 3-2 through 3-4 that point estimates are substantially less variable than the NOEC for the same method and endpoint, and that the LC50 for an acute toxicity test usually is less variable than the LC50 for a chronic toxicity test. The estimated NOEC is more variable than ECp *using current experimental designs* because NOEC can take only those values equal to the concentrations tested, while ECp interpolates between tested concentrations (there may be other, more technical reasons as well). In principle, NOEC could be estimated more accurately and precisely by changing the experimental design to use more concentrations at narrower dilution ratios and by using more replicates. The greater variability of the NOEC underscores the desirability of using point estimates to characterize effluent toxicity.

Tables 3-2 through 3-4 may be used as benchmarks for variability, allowing comparison of one laboratory's CV for reference toxicant testing with CVs reported by experienced laboratories reporting tests that passed the TAC. However, CVs for methods represented by too few laboratories in the table may be atypical.

The CVs in Tables 3-2 through 3-4 may be used as an adjunct to the control chart. If the CV for reference toxicant tests is above the 75th percentile in Tables 3-2 through 3-4, variability likely can be reduced, even if the individual EC25 or LC50 values fall within the control limits. If a control chart is constructed using an unreasonably large standard deviation, the control limits will be unreasonable. If a high CV is not fully explained by an unusually small mean, the standard deviation of EC25 or LC50 should be reduced to bring the CV within the normal range. If the CV exceeds the 90th percentile (Appendix B), there is no question that variability is unacceptably large. Detailed guidance is provided in Chapter 5 (Section 5.3.1.1).

Tables 3-2 through 3-4 indicate the magnitude of the analytical variability that becomes part of the variability of effluent test results under certain conditions. This occurs when effluent test results (NOECs, LC50s, or EC25s) fall between the lowest and highest concentrations tested. Under other conditions, these