Exhibit 3

Arid West Water Quality Research Project

EVALUATION OF THE EPA RECALCULATION PROCEDURE IN THE ARID WEST

TECHNICAL REPORT

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prepared by Parametrix, Inc., Albany, Oregon
Chadwick Ecological Consultants, Littleton, Colorado
URS Corporation, Albuquerque, New Mexico and

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cover photo: Santa Cruz River, near Tubac, AZ
Linwood Smith, photographer

number of years studied, and methods used in those studies. Regardless, the fish and invertebrate taxa lists developed provide a list of resident taxa for the recalculation effort described later in this document.

ES.3 ALUMINUM CRITERIA REVIEW AND UPDATE

The 1988 report entitled *Ambient Water Quality Criteria for Aluminum* (EPA 1988) underwent a technical review and update as the initial step for inclusion in the Arid West Water Quality Research Project AWQC Recalculation Project. The speciation and/or complexation of aluminum (Al) is highly dependent on ambient water quality characteristics and ultimately determines the mechanism of toxicity. Concentration of calcium in the water was shown to decrease toxic effects to fish.

A comprehensive literature review resulted in the addition of 36 acute data points from 15 studies to the updated aluminum acute database (Chapter 3). Additionally, 11 chronic data points from nine studies were added to the updated aluminum chronic database. The updated acute database revealed a statistically significant inverse Al toxicity and hardness relationship with a slope of 0.8327. This was not reported in the 1988 Aluminum AWQC.

The updated acute database contains values for 17 genera, while the updated Al chronic toxicity database presents data for six genera of freshwater organisms. Since the revised chronic database did not satisfy the "eight-family rule," the FACR was used to derive a FCV for Al from the acute database. New acute and chronic hardness-based equations were derived from the updated databases (Table ES-1). The updated and revised acute and chronic criteria based on these equations are presented across a wide range of hardness levels (Table ES-1). It is important to understand the boundaries of the reported equation. Since the equation models hardness values that ranged from 1 mg to 220 mg of $CaCO_3$/L, estimations made outside of this range should be treated with caution. Given that arid West EDWs can often exhibit hardness values much greater than 220 mg/L, this represents an uncertainty.

Table ES-1
Updated and Revised Acute and Chronic Al Criteria Values (µg Total Aluminum/L) Across Selected Hardness Values

	Mean Hardness (mg/Las $CaCO_3$)									
Updated/Revised National Standards	25	50	75	100	150	200	250	300	350	400
Acute Al Criterion: $e^{(0.8327\,[\ln(\text{hardness})]+3.8971)}$	719	1,280	1,794	2,280	3,195	4,060	4,889	5,691	6,470	7,231
Chronic Al Criterion: $e^{(0.8327\,[\ln(\text{hardness})]+2.9800)}$	287	512	717	911	1,277	1,623	1,954	2,275	2,586	2,890

NOTE: Current EPA Al criteria: 750 µg/L acute; 87 µg/L chronic (although acute effects could also be observed at acidic pH). Given Al speciation and behavior in complex solutions, the mechanism responsible for toxicity will probably be dependent on pH and calcium concentration of a given solution. Therefore, understanding Al speciation chemistry and its influence on the mechanisms of toxicity to fish and invertebrates are important to interpreting the toxicological studies which form the basis of AWQC development.

3.3 PHASE I- TECHNICAL REVIEW OF 1988 ALUMINUM DOCUMENT

Phase I of the evaluation of the 1988 Aluminum Document consisted of a thorough investigation of the data used to calculate the most recent Al criteria. This document was critically reviewed for relevance of the toxicological data and adherence to EPA methodology (Stephan et al. 1985).

3.3.1 Existing Acute Criteria for Aluminum

The 1988 Aluminum Document (EPA 1988) presents acute data for 14 genera, including seven species of invertebrates and seven species of fish. These 14 species in 11 families satisfy the "eight-family rule" as specified in the 1985 Guidelines. The 1988 Aluminum Document reports a FAV of 1,496 μg/L with a CMC = FAV/2 of 750 μg/L. When referencing reported values used in criteria development, Chadwick Ecological Consultants (CEC) identified an apparent discrepancy regarding the SMAV for *Girardia (= Dugesia) tigrina*. The authors of the toxicity test reported that the greatest Al exposure concentration of for this species was 16,000 μg/L (Brooke 1985) with the ambient acute value of >16,800 μg/L. However, the 1988 Aluminum Document reports >23,000 μg/L for the same species and reference. The implications of this discrepancy could be significant and would result in a *Girardia* GMAV rank change from 6th to 4th. Charles Stephen (pers. comm. to David Moon, December 13, 2004) has since noted that no *G. tigrina* died at 16,000 μg/L, so it was reasonable to assume that the LC_{50} was potentially two times the concentration that caused a low level of acute mortality. The geometric mean of 16,000 μg/L and 32,000 μg/L was reported for *Girardia* to account for the undefined test value. Nonetheless, the undefined value (>16,000 μg/L) is probably more appropriate, when compared to more recent evidence reported by Calevro et al. (1998) that tested Al toxicity in *G. etrusca*. The authors' results showed that this species showed lethality, abnormal mucus production, and decreased regeneration at concentrations near 16,000 μg/L. Therefore, we replaced the existing >23,000 value with the originally reported value of >16,000 μg/L.

3.3.2 Existing Chronic Criteria for Aluminum

The 1988 Aluminum Document presents chronic data for three genera of freshwater organisms, including two species of invertebrates and one fish species. These three species do not satisfy the "eight-family rule" as specified in the 1985 Guidelines. The chronic database assemblage did, however, satisfy the minimal requirements for calculation of an ACR in that one of the invertebrates is an acutely sensitive species.

After calculation of three valid ACRs for the three species, it was evident that the most acutely sensitive species had lower ACRs. Given this relationship, a final ACR (FACR) was calculated using acutely sensitive *Ceriodaphnia dubia*, which resulted in a FACR that was less than 2, which then defaults to 2 according to USEPA guidance (Stephan et al. 1985). A FACR of 2 would result in a chronic criterion equal to the acute. However, additional information on Al toxicity for *Salvelinus fontinalis* and *Morone saxatilis* (Cleveland et al. manuscript and Buckler et al. manuscript) were used by the EPA to modify the FCV to protect these two species (EPA 1988). These two studies were deemed inappropriate for the Al chronic database (i.e., they are listed in Table 5-6, "Other Data on Effects of Aluminum on Aquatic Organisms"), but were still used to reduce the FCV from ~750 to 87 μg/L. Therefore, the 1988 Aluminum Document recommended a Criteria Chronic Concentration (CCC) of 87 μg/L at which no *M. saxatilis* died after a seven-day exposure (Buckler et al. manuscript). In the same toxicity test, 174.4 μg/L killed 58% of the fish. Current practice would be to calculate the chronic value as the geometric mean of these two numbers, or 122 μg/L.

3.4 PHASE II – UPDATE TO THE NATIONAL ALUMINUM CRITERIA DATABASE

A comprehensive literature review was conducted of Al aquatic toxicity related documents used and not used in the 1988 Aluminum Document. This included a review of documents published since the 1988 Aluminum Document, as well as those published prior to 1988 that were not used in criterion derivation. Available Al documents were obtained and reviewed for relevance of toxicological data and adherence to EPA criteria development methodology (Stephen et al. 1985).

A pH range of 6.5 to 9.0 was established as a limit for data used in the update of the Al toxicity databases, because the EPA has established this as an acceptable range for pH in ambient freshwater (EPA 1976). This circumneutral pH gradient was the same range used to derive current criteria in the 1988 Aluminum Document. From the discussion on Al speciation above, we would thus expect that toxic effects of Al in test media of circumneutral pH could be attributed to exposure to non-monomeric Al species. Additionally, reported total Al measurements should be substantially greater than dissolved measurements owing to the poor solubility of Al under these pH conditions. Approximately 120 papers were reviewed, including documents cited in the 1988 Aluminum Document.

Much of the research into Al toxicity in aquatic organisms has been concerned with toxicity of Al in acidic solutions – specifically in research investigating effects of acid rain – with very few studies addressing toxic

Table 3-1
Summary of Reviewed Publications with Reported Acute Aluminum Data that were Deemed Acceptable According to 1985 Guidelines (Stephen et al. 1985) for Addition to the Updated Aluminum Acute Database

Species	Method	Hardness (mg/L $CaCO_3$)	pH	LC_{50} (µg/L)	Reference
Ictalurus punctatus	F, M	23.1	6.5	>400	Palmer et al. 1988
Ictalurus punctatus	F, M	23.1	7.5	>400	Palmer et al. 1988
Oncorhynchus mykiss	S, U	1	7	3,800	Thomsen et al. 1988
Oncorhynchus mykiss	S, U	150	7	71,000	Thomsen et al. 1988
Oncorhynchus mykiss	F, M	25	7.6	<8,000	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	45	7.6	<8,000	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	85	7.6	<8,000	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	125	7.6	<8,000	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	23.2	8.25	6,170	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	35	8.25	6,170	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	83.6	8.29	7,670	Gundersen et al. 1994
Oncorhynchus mykiss	F, M	115.8	8.29	6,930	Gundersen et al. 1994
Pimephales promelas	F, M	21.6	6.5	>400	Palmer et al. 1989
Pimephales promelas	F, M	21.6	7.5	>400	Palmer et al. 1989
Pimephales promelas	F, M	21.6	6.5	>400	Palmer et al. 1989
Pimephales promelas	F, M	21.6	7.5	>400	Palmer et al. 1989
Pimephales promelas	F, M	23.1	6.5	>400	Palmer et al. 1988
Pimephales promelas	F, M	23.1	7.5	>400	Palmer et al. 1988
Pimephales promelas	S, M	26	7.8	1,160	ENSR 1992b
Pimephales promelas	S, M	46	7.6	8,180	ENSR 1992b
Pimephales promelas	S, M	96	8.1	20,300	ENSR 1992b
Pimephales promelas	S, M	194	8.1	44,800	ENSR 1992b
Crangonyx pseudogracilis	S, U	50	6.75	9,190	Martin and Holdich 1986
Asellus aquaticus	S, U	50	6.75	4,370	Martin and Holdich 1986
Gammarus pulex	S, U	--	6.9	>2,698	Storey et al. 1992
Ceriodaphnia dubia	S, M	26	7.5	720	ENSR 1992a
Ceriodaphnia dubia	S, M	46	7.6	1,880	ENSR 1992a

Table 3-1 (Continued)
Summary of Reviewed Publications with Reported Acute Aluminum Data that were Deemed Acceptable According to 1985 Guidelines (Stephen et al. 1985) for Addition to the Updated Aluminum Acute Database

Species	Method	Hardness (mg/L $CaCO_3$)	pH	LC_{50} (µg/L)	Reference
Ceriodaphnia dubia	S, M	96	7.8	2,450	ENSR 1992a
Ceriodaphnia dubia	S, M	194	8.1	>99,600	ENSR 1992a
Ceriodaphnia dubia	S, M	98.5	7.6	2,880	Soucek et al. 2001
Ceriodaphnia sp.	S, M	47.4	7.36	2,300	Call 1984
Cyclops viridis	S, U	--	6.9	>2,698	Storey et al. 1992
Micropterus dolomieu	S, M	12.45	7.5	>1,000	Kane and Rabeni 1987
Salmo salar	S, M	6.8	6.5	599	Hamilton and Haines 1995
Salvelinus fontinalis	F, M	--	6.5	3,600	Decker and Menendez 1974
Hybognathus amarus	S, M	140	8.1	>59,100	Buhl 2002

NOTES:
S = static renewal test exposures
F = flow through test exposure
M = test media aluminum concentration was measured
U = test media aluminum concentration was not measured

Water quality parameters in toxicity tests were added to the updated Al database in addition to test results. Test solution pH and hardness values were needed to determine inclusion of data within the specified circumneutral pH range and to investigate a possible hardness-toxicity relationship. Most of the added studies reported hardness values of test media or reported calcium and magnesium concentrations that were used to calculate water hardness. Of the 35 new acute data points, three provided insufficient information on water quality parameters to determine test media hardness. Unfortunately, each was for a unique species (*Salvelinus fontinalis*, *Cyclops viridis*, and *Gammarus pulex*) found in the updated database that was subsequently removed during FAV derivation (see discussion below).

3.4.2 New Aluminum Chronic Toxicity Data

Following review of these studies, 11 chronic data points from nine studies (Table 3-2) were added to the revised chronic database. Of the nine studies added to the database, seven were published prior to the 1988 Aluminum Document. Three studies published prior to the 1988 Aluminum Document were not cited in either Table 1 ("Chronic Toxicity of Aluminum to Aquatic Animals") or Table 6 ("Other Data on Effects of Aluminum on Aquatic Organisms") of the 1988 Aluminum Document and apparently represent data that were unknown to the EPA at the time.

Table 3-2
Summary of Chronic Aluminum Data that were Deemed Acceptable for Criteria Derivation and Added to the Updated Aluminum Chronic Database

Species	Hardness (mg/L $CaCO_3$)	pH	NOEC- LOEC (Φg/L)	Chronic Value (μg/L)	Reference
Ceriodaphnia dubia	50	7.75	1,100-2,400	1,624	McCauley et al. 1986
Ceriodaphnia dubia	47.4	7.55	6,250-12,100	8,696.26	Call 1984
Daphnia magna	45.3	7.74	--	320[a]	Biesinger and Christenson 1972
Daphnia magna	45.3	7.74	--	1,400[b]	Biesinger and Christenson 1972
Tanytarsus dissimilis	17.43	6.8	10,000-80,000	28,284	Lamb and Bailey 1981
Salvelinus fontinalis	12.5	7.2	>303.9	>303.9	Cleveland 1991
Salvelinus fontinalis	7.5	6.5	169-350	243.21	Cleveland manuscript
Salvelinus fontinalis	12.5	6.5	57-88	70.82	Cleveland manuscript
Salvelinus fontinalis	7.5	6.5	88-169	121	Cleveland et al. 1989
Salvelinus fontinalis	0.567	7.81	0-300	<283	Hunn et al. 1987
Micropterus dolomieu	12.8	7.3	0-250	<250	Kane and Rabeni 1987

NOTES:
[a] EC_{16} for reduced reproduction
[b] 21 day LC_{50}
NOEC = no observable effect concentration
LOEC = lowest observable effect concentration.

Four publications that were found in Table 6 ("Other Data") of the 1988 Aluminum Document were reviewed and deemed appropriate for use in updating the chronic database. Biesinger and Christensen (1972) performed acute and chronic Al toxicity tests with *Daphnia magna.* Acute toxicity results were included in the acute database; yet, no explanation was given as to why chronic data from this study were not included in the chronic database. We reviewed methods used for the chronic toxicity tests, and could not find a reason to exclude these data. Therefore, two chronic values from this study were added to the database. Data from this publication were also deemed suitable for inclusion in the FACR derivation, described later.

In a 55-day Al exposure, Lamb and Bailey (1981) tested acute and chronic toxicity in *Tanytarsus dissimilis.* The authors reported high variability in mortality rates among treatments and provided little information on statistical significance of mortality among treatments. Fortunately, a figure showing the cumulative % mortality was provided and analyzed with text to derive a chronic value of 10,000 μg/L, the treatment level that produced 37% mortality.

The Cleveland manuscript, used to lower the 1988 Aluminum Document FCV, contained additional data for *Salvelinus fontinalis* that were not reported in the EPA chronic databases. CEC added these additional chronic values into the revised chronic database. *S. fontinalis* were exposed to Al in soft water with a pH of 6.5, the lowest pH in the acceptable circumneutral range. The chronic value was determined for a statistical difference in length (growth) and mortality. The growth value was more sensitive than mortality (243 µg/L) and resulted in a chronic value of 70 µg/L. Hunn et al. (1987) investigated influence of pH and Al on early life stages of developing *S. fontinalis*. Only two treatments, the control and 283 µg/L, were used in a 60-day larvae toxicity test using flow through exposure with very soft water. The authors reported a statistical decrease in growth ($p<0.001$) between treatment and control using a least squares deviation linear model with interaction terms representing treatment effects. Since a geometric mean could not be determined, a chronic value of <283 µg/L was added to the revised chronic database.

Five additional studies with appropriate toxicity tests were found that were not listed in the 1988 Aluminum Document. Three of these publications were published after the 1988 Aluminum Document. Cleveland et al. (1991) performed a 56-day Al exposure in *S. fontinalis* to examine effects on bioaccumulation, growth, and mortality. The authors reported 1% mortality in the 7.2 pH treatment at the end of the exposure period at a measured mean Al concentration of 303.88 µg/L, which resulted in an undefined chronic value of >303.88 µg/L. Although test duration was four days short of the recommended 60 days for a chronic test with this species, we decided that test methods were acceptable and suitable for use. In a chapter of a book on environmental chemistry and toxicology of Al, Cleveland (1989) reported another chronic value for *S. fontinalis*. The authors used similar methods as in prior toxicity tests with this species and Al. After a 60-day exposure at a mean pH of 6.5, statistical differences in growth were observed. The result of this partial life cycle test, that started exposures with embryos, was the lowest chronic value added to the chronic database.

The remaining three studies entered into the updated chronic database were published prior to the 1988, but were not cited in the 1988 Aluminum Document. McCauley et al. (1986) performed two acute and chronic toxicity tests using *C. dubia* with different pH exposure media. The 1988 document used only one of the chronic values from a test with a pH of 7.15, but did not report the second test that was conducted at a pH of 7.61. The chronic value that was added to the updated database was from this second test. Extensive acute data were provided by Call (1984) from the University of Wisconsin Center for Lake Superior Environmental Studies laboratory, with addition of a chronic toxicity test using *Ceriodaphnia* sp. After an eight-day Al exposure, statistical differences in survival and reproduction were observed in the 12,100 µg/L treatment (lowest-observed-effect concentration [LOEC]). The updated chronic database value was derived by taking