Update on Dunkard Creek
November 23, 2009

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This is an interim report on the aquatic life kill in Dunkard Creek and the investigation into the cause of the kill. Our findings at this time are preliminary. We are still learning about the ecology of the alga *Prymnesium parvum* in this region and are investigating its distribution. We will continue to make more information available as we learn of it.

Background

The Dunkard Creek watershed drains approximately 180 square miles in Monongalia County in West Virginia and Greene County in Pennsylvania (WVDEP 2009). Dunkard Creek has Forks in both states and forms from the confluence of the West Virginia Fork and the Pennsylvania Fork just upstream of Brave, PA. The stream flows along the Mason Dixon Line crossing back and forth between the states until it leaves West Virginia near Buckeye Church, WV, flowing northeast toward Mount Morris, PA and then further toward its confluence with the Monongahela River.

The kill on Dunkard Creek included fish, salamanders, and mussels and began on or about September 1 (Table 1). In general, the kill has been described as massive and, in terms of mussels, complete. The kill on Dunkard Creek spans approximately 43 miles of stream (different mileages have been seen in different accounts of this kill because early in the kill, the zone was restricted to Prentice, WV, but continued to work its way upstream and downstream from there throughout the kill).

On September 9, we investigated the kill on Dunkard Creek. We collected in situ measurements of pH, dissolved oxygen, conductivity, and temperature at ten sites (D1-D10) and water samples for metals, nutrients and mining constituents at 4 sites (D4, and D8-10). Our investigation was centered on the Blacksville #2 discharge in the WV Fork of Dunkard Creek.

During the rest of September the West Virginia Department of Environmental Protection (WVDEP), West Virginia Department of Natural Resources (WVDNR), Pennsylvania Department of Environmental Protection (PADEP), and Pennsylvania Fish and Boat Commission (PAFBC) continued to sample the creek. We are currently compiling this data in a central database and hope to construct a more complete timeline of the kill.

Table 1. **Rough timeline of Dunkard Creek kill.** This kill has been investigated by WVDNR, WVDEP Regional and Charleston offices, PAFBC, PADEP Southwest Regional Office, and USEPA Wheeling Freshwater Biology Team.

August 28 –WVDNR reports high conductivity in Dunkard Creek. The conductivity may or may not have been higher at an earlier point in time. Also, we don't know how long it was at 50,000 uS
Sept 4 – Preliminary investigations by WVDNR on mussel and fish kills in Pentress, WV
Sept 4 – October 1 – WVDNR investigates kill at 44 sites and 22 observation days
Sept 4 - WVDEP samples water at 4 sites and conductivity at 31 sites
Sept 8 - 11 – WVDNR and PAFBC on site evaluating fish and mussel kill
Sept 9 – USEPA samples in situ water chemistry at 10 sites and collects water samples at 4 sites
Sept 9-18 – PFBC samples fish kill at numerous sites in PA. USEPA assisted on Sept 10
Sept 10 – PADEP samples at five sites in Dunkard Creek
Sept 13 -14 – WVDEP (Brad Swiger) sampling in Dunkard Creek
Sept 15 – PADEP samples five sites in Dunkard Creek
Sept 18 – WVDEP fly over in helicopter investigating kill
Sept 20 – USEPA and WVDEP sample algae at 6 sites in watershed
Sept 23 – WVDEP samples algae at 6 sites in watershed
Sept 30 – USEPA meets with WVU, PADEP, WVDEP
Oct 19 – WVDNR electrofishing survey at selected sites in basin
Oct 26 – CONSOL Energy, Inc. (CONSOL) reports finding golden algae in a sample collected from Whitely Creek on September 29, 2009.

Cause of the Fish Kill

We now know that a substantial bloom of the golden algae *Prymnesium parvum* was present in Dunkard Creek at the time of the kill. This identification has been confirmed by experts from North Carolina, South Carolina, Florida, and Oklahoma. This saltwater alga produces a potent toxin that is capable of killing fish, mussels, and salamanders. This toxin affects gill breathing organisms and is not toxic to humans, waterfowl, or livestock (Sager et al. 2008).

P. parvum is found worldwide and is most common in saltwater (Sager et al. 2008). It is an invasive saltwater alga now being found in brackish (both natural and anthropogenic) inland waters and has been documented in many states (Figure 1). Since its discovery in Texas in 2001, *P. parvum* blooms have killed over 30 million fish in 33 water bodies (Sager et al. 2008).

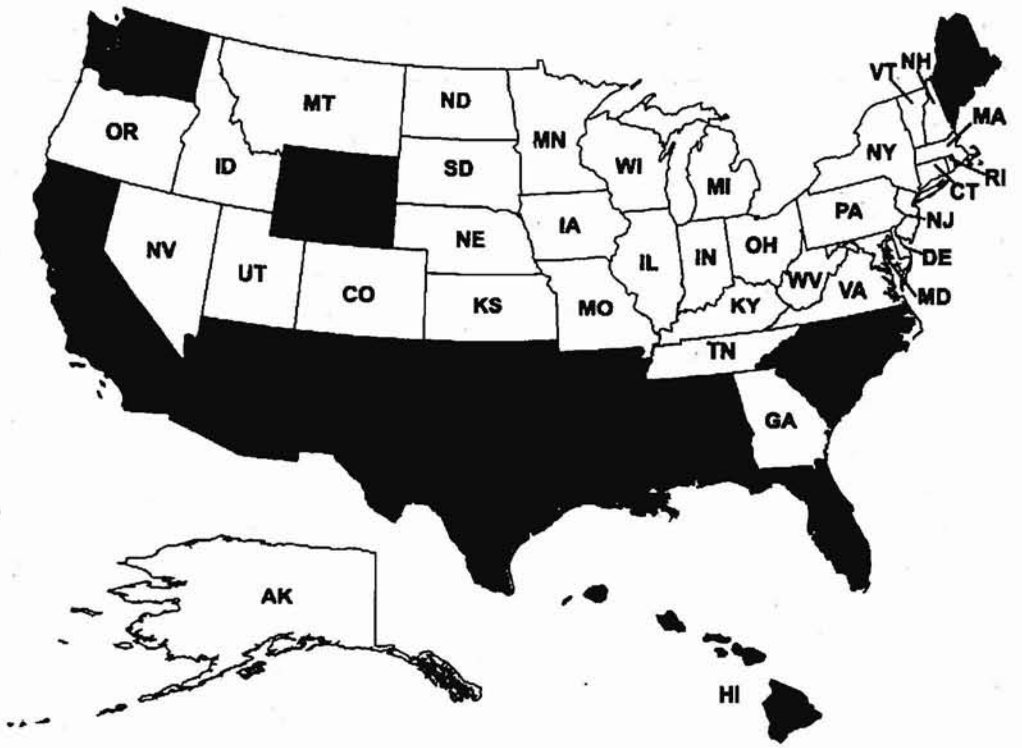

Figure 2. The states of the United States with golden alga presence reported (in dark shading).

Figure 1. **Distribution of *P. parvum* in the United States** (Sager et al 2008).

There are a number of factors that influence blooms of *P. parvum*:

1. *P. parvum* is a saltwater algae and blooms are associated with increased salinity (Baker et al. 2009, Sager et al. 2008, Rodgers, In Press). Blooms in Texas are limited to Central and Western Texas where natural conditions and brines associated with oil production produce saline water bodies.
2. Research has shown that the toxin produced by this alga is dependant upon the availability of cations (e.g., Ca^{2+} and Mg^{2+}) in the surrounding water. At higher pH, more cations are available for the formation of toxin. It has been noted in Texas that in waterbodies with a pH <7, fish kills do not occur despite the presence of the algae (Sager et al., 2008).
3. *P. parvum* is a mixotroph and can get its energy through photosynthesis when nutrients are sufficient. When nutrients are limited, however, it can produce toxins to kill other organisms and feed from their nutrients.
4. *P. parvum* has been found in a range of waters with TDS levels from 1000 – 100,000 mg/l TDS, and experts believe an optimum TDS range is 3000-60,000 mg/l. So, although it is a brackish water alga, it can survive in waters with relatively low TDS levels.
5. *P. parvum* competes with native algae and the saline conditions that favor P. parvum are stressful for its freshwater competitors.

The bloom on Dunkard Creek was noted first by a WVDEP fly-over on September 18, more than two weeks after the fish kill was discovered. Inspectors from WVDEP noticed the water was discolored and stained over the entire length of Dunkard Creek and this staining originated at a beaver dam in the headwaters of the West Virginia Fork of Dunkard Creek. This beaver pond is upstream of the Blacksville #2 mine, but downstream of another outfall from CONSOL's St. Leo Mine. WVDEP staff suspected the coloration was caused by an algal bloom. We, along with WVDEP, sampled six sites on Dunkard for algae on September 20, 2009. WVDEP subsequently sampled the week following.

Preliminary results (Table 2) show that the algae were found in sufficient numbers to produce toxin (Dr. Carmello Tomas, associate professor of biological sciences at the University of North Carolina-Wilmington). Dr. Tomas ran an Erythrocyte Lysis assay to assess the toxicity of the samples. This assay measures the percent hemolysis of erythrocytes as a measure of the toxicity of the algae (the algae produces a hemotoxin). In general he found that the areas with the highest conductivity were most impacted. Dr. Tomas reported the results to WVDEP in an email dated September 29, 2009.

Table 2. **Preliminary Results from University of North Carolina-Wilmington Laboratory.** Cells/ml is a measure of the number of algal cells in a ml. of sample. Percent hemolysis is a measure of the percent of lysed blood cells when compared to a control.

Sample	Cells/mL	% Hemolysis Cells
WANA	345,320	95.9
MDP	242,300	91.1
WTL	304,600	93.6
UMR	102.200	____
DBP	94.600	____
UBD	460	____

According to counts the densities were

1. WANA *(bridge at Wana, WV ~RM38)*
2. WTL *(beaver dam wetland ~RM20)*
3. MDP *(Mason Dixon Park ~RM44)*
4. UMR *(Upstream of Miracle Run ~RM34)*
5. DBP *(Downstream of beaver dam ~RM43)*
6. UBD *(upstream of beaver dam ~RM45)*

For hemolytic analyses the top three are in order.

Algal cell densities found by Dr. Tomas at WANA and at MDP were high enough to produce a toxic effect (as evidenced by the assay). These cell densities are high compared to other blooms that have been noted as toxic (Rodgers, In Press).

We have also been working with researchers at the University of Oklahoma Biological Station Plankton Ecology Lab. They are currently doing a genetic analysis that may determine the source of the algae and are also assessing the toxin levels.

WVDEP sent fish organs to a fish pathologist with the USGS (Dr. Vicki Blazer). Her preliminary findings report organ damage consistent with a toxin.

Given what has been seen in other states and the etiology of this kill, we believe the toxin from this algae bloom led to the kill of fish, mussels, and salamanders on Dunkard Creek. At this time, we do not know where the algae originated. The elevated conductivity in the creek likely created favorable conditions for this alga to grow and produce toxin. This alga is not known to grow or produce toxin at the natural levels of TDS in Dunkard Creek (<280 mg/l).

Stressors in Dunkard Creek

WVDEP's 303(d) List and TMDLs

Elevated TDS and component ions (e.g., chloride, sulfate, magnesium, bicarbonate) are toxic to aquatic life and chronic exposure to high TDS leads to aquatic life use impairment (Pond et al. 2008). The level of TDS in Dunkard Creek during the time of the kill was many times higher than levels known to cause aquatic life use impairment. A major component of the TDS in Dunkard Creek is chloride, which is a pollutant with an EPA chronic criterion of 230 mg/L and an acute criterion of 860 mg/L (published in 1988). WVDEP adopted these criteria into its water quality standards. Many stream reaches in the Dunkard Creek watershed (Figures 2 and 3) are currently on the 303(d) list for impairments to aquatic life (Table 3), and EPA recently approved WVDEP's Total Maximum Daily Loads (TMDLs) for Dunkard Creek for some stressors (WVDEP 2009).

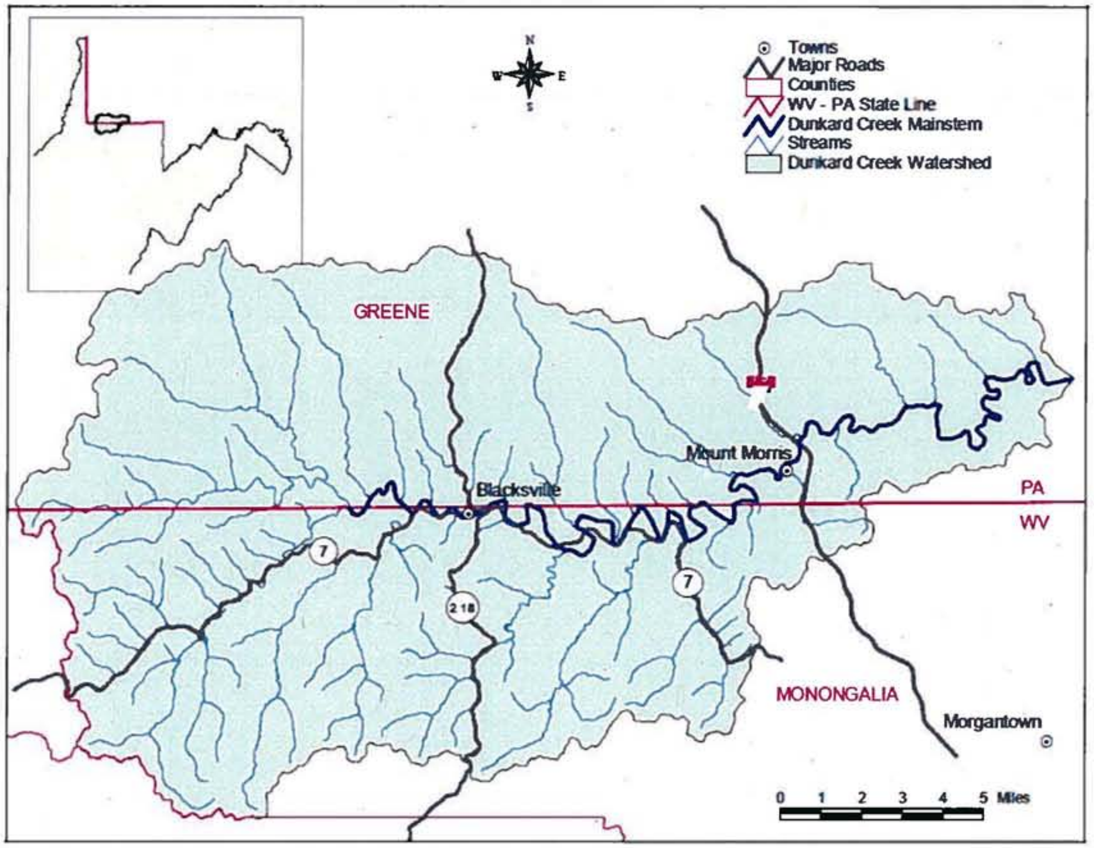

Figure 3-1. Location of the Dunkard Creek watershed

Figure 2. **General Location of Dunkard Creek Watershed (WVDEP 2009).**

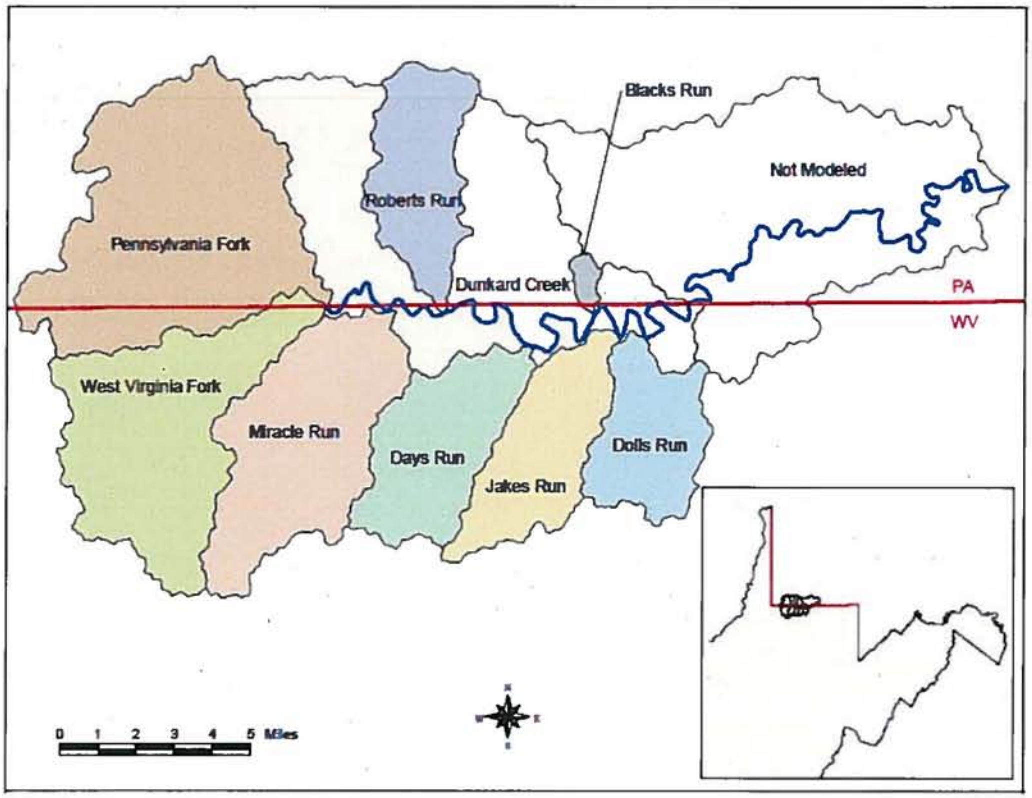

Figure 3-2. Dunkard Creek TMDL watersheds

Figure 3. **TMDL Watersheds of Dunkard Creek (WVDEP 2009).**

Table 3. **Reach impairments and stressors that have TMDLs developed for Dunkard Creek and its tributaries (WVDEP 2009).**

Table 3-3. Waterbodies and impairments for which TMDLs have been developed

Subwatershed	Stream Name	NHD_Code	Fe	Cl	FC	BIO
Dunkard Creek	Dunkard Creek	WV-ML-128	x		x	x
Dolls Run	Dolls Run	WV-ML-128-AC	X		x	x
Dolls Run	Pedlar Run	WV-ML-128-AC-4	X		x	x
Dolls Run	UNT/Pedlar Run RM 1.20	WV-ML-128-AC-4-B	X		x	
Dolls Run	Smoky Drain	WV-ML-128-AC-5	X		x	x
Dolls Run	Berry Hollow	WV-ML-128-AC-6	X			
Jakes Run	Jakes Run	WV-ML-128-AE	X		x	x
Jakes Run	UNT/Jakes Run RM 5.5	WV-ML-128-AE-12	X		x	
Jakes Run	UNT/Jakes Run RM 2.33	WV-ML-128-AE-4			x	
Blacks Run	Blacks Run	WV-ML-128-AF	X			x
Dunkard Creek	Hackelbender Run	WV-ML-128-AG	X			
Days Run	Days Run	WV-ML-128-AJ	X		x	x
Dunkard Creek	UNT/Days Run RM 6.2	WV-ML-128-AJ-10	X			
Dunkard Creek	UNT/Days Run RM 7.3	WV-ML-128-AJ-12	X			
Dunkard Creek	Indian Camp Run	WV-ML-128-AJ-4	X			
Days Run	Shriver Run (ML-128-AJ-8)	WV-ML-128-AJ-8	x		x	x
Days Run	Building Run (ML-128-AJ-8-C)	WV-ML-128-AJ-8-C	X		x	
Days Run	UNT/Days Run RM 5.8	WV-ML-128-AJ-9	X		x	x
Dunkard Creek	UNT/UNT RM 0.89/Days Run RM 5.8	WV-ML-128-AJ-9-C	X			
Dunkard Creek	Kings Run	WV-ML-128-AP	X			
Roberts Run	Roberts Run	WV-ML-128-AR	X		x	
Miracle Run	Miracle Run	WV-ML-128-AV	X		x	
Miracle Run	Thomas Run	WV-ML-128-AV-1			x	
Miracle Run	Scott Run	WV-ML-128-AV-11			x	
Miracle Run	UNT/Miracle Run RM 5.50	WV-ML-128-AV-16	X			
Miracle Run	UNT/Miracle Run RM 6.55	WV-ML-128-AV-18	X			
Miracle Run	Right Branch/Miracle Run	WV-ML-128-AV-3	X		x	x
PA Fork Dunkard Creek	Pennsylvania Fork/Dunkard Creek	WV-ML-128-BA	X		x	
PA Fork Dunkard Creek	Brushy Fork	WV-ML-128-BA-12	X			
PA Fork Dunkard Creek	UNT/Pennsylvania Fork RM 8.2	WV-ML-128-BA-15	X			
PA Fork Dunkard Creek	UNT/Pennsylvania Fork RM 9.55	WV-ML-128-BA-18	X			
PA Fork Dunkard Creek	Pumpkin Run	WV-ML-128-BA-4	X			
WV Fork Dunkard Creek	West Virginia Fork/Dunkard Creek	WV-ML-128-BB	x	x	x	
WV Fork Dunkard Creek	Shriver Run (ML-128-BB-10)	WV-ML-128-BB-10	X			
WV Fork Dunkard Creek	Range Run	WV-ML-128-BB-13	X		x	x

Table 3. continued. **Reach impairments and stressors that have TMDLs developed for Dunkard Creek and its tributaries (WVDEP 2009).**

Subwatershed	Stream Name	NHD_Code	Fe	Cl	FC	BIO
WV Fork Dunkard Creek	South Fork/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-14	x	x	x	
WV Fork Dunkard Creek	Middle Fork/South Fork/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-14-A			x	
WV Fork Dunkard Creek	UNT/South Fork RM 3.0/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-14-F	X	x		
WV Fork Dunkard Creek	North Fork/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-15	X		x	x
WV Fork Dunkard Creek	Camp Run	WV-ML-128-BB-15-B	X		x	x
WV Fork Dunkard Creek	Browns Run	WV-ML-128-BB-15-B-1	X			
WV Fork Dunkard Creek	Joy Run	WV-ML-128-BB-15-B-2	X			
WV Fork Dunkard Creek	Briar Run	WV-ML-128-BB-15-B-4	X			
WV Fork Dunkard Creek	Hughes Run	WV-ML-128-BB-3	X			
WV Fork Dunkard Creek	Wise Run	WV-ML-128-BB-9	X		x	x

Note:

UNT = unnamed tributary; RM = river mile.

CL indicates chloride impairment

FC indicates fecal coliform bacteria impairment

BIO indicates a biological impairment

Table 4. **Stressors on biologically impaired reaches of Dunkard Creek. WVDEP identified ionic stress as a stressor in some reaches of Miracle Run and the WV Fork of Dunkard (WVDEP 2009).**

Table 4-1. Significant stressors of biologically impaired streams

TMDL Watershed	Stream Name	NHD_Code	Biological Stressors	TMDLs Developed
Dunkard Creek	Dunkard Creek	WV-ML-128	Sedimentation Organic Enrichment	Total Iron Fecal Coliform
Dolls Run	Dolls Run	WV-ML-128-AC	Sedimentation Organic Enrichment	Total Iron Fecal Coliform
Dolls Run	Pedlar Run	WV-ML-128-AC-4	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Dolls Run	Smoky Drain	WV-ML-128-AC-5	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Jakes Run	Jakes Run	WV-ML-128-AE	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Blacks Run	Blacks Run	WV-ML-128-AF	Sedimentation	Total Iron
Days Run	Days Run	WV-ML-128-AJ	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Days Run	Shriver Run	WV-ML-128-AJ-8	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Days Run	UNT/Days Run RM 5.8	WV-ML-128-AJ-9	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
Miracle Run	Miracle Run	WV-ML-128-AV	Organic Enrichment Sedimentation Ionic Stress	Fecal Coliform Total Iron Ionic Strength (To remain on the 303d List)
Miracle Run	Building Run	WV-ML-128-AV-15	Ionic Stress	Ionic Strength (To remain on the 303d List)
Miracle Run	Right Branch/Miracle Run	WV-ML-128-AV-3	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
WV Fork	West Virginia Fork/Dunkard Creek	WV-ML-128-BB	Sedimentation Organic Enrichment Ionic Stress	Total Iron Fecal Coliform Ionic Strength (To remain on the 303d List)
WV Fork	Range Run	WV-ML-128-BB-13	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
WV Fork	South Fork/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-14	Ionic Stress Organic Enrichment Sedimentation	Ionic Strength (To remain on the 303d List) Fecal Coliform Total Iron
WV Fork	North Fork/West Virginia Fork/Dunkard Creek	WV-ML-128-BB-15	Organic Enrichment Sedimentation	Fecal Coliform Total Iron
WV Fork	Camp Run	WV-ML-128-BB-15-B	Organic Enrichment	Fecal Coliform
WV Fork	Wise Run	WV-ML-128-BB-9	Organic Enrichment Sedimentation	Fecal Coliform Total Iron

We also reviewed chloride toxicity information compiled by the State of Iowa for development of its chloride criterion as well as results from Canada's Ministry of the Environment. The following data are from Canada's Ministry of Environment (http://www.env.gov.bc.ca/wat/wq/BCguidelines/chloride/chloride.html).

Figure 4 shows the percentage of aquatic organisms affected at certain chloride concentrations. Figure 5 shows acute and chronic data for all affected species, as well as a modeled response to longer term chronic exposures. Table 5 shows the 96 hr LC 50 (concentration that kills 50% of test organisms for tested species). An LC50 represents an acute endpoint, so these levels would not be protective to longer term chronic exposures nor do they reflect effects on chronic endpoints, such as biotic growth or reproduction. Chloride LC50 levels shown in Table 4 vary widely by species and in general, fish can, in the short term, tolerate high levels. In general, invertebrates tend to be more sensitive to elevated TDS than are vertebrates.

The situation in Dunkard Creek should be considered a chronic exposure since chloride levels were elevated above the criteria for long periods of time. Tables 6 and 7 report our field and laboratory chemistry results from our field visit on September 9, 2009. The chloride levels that WVDEP, PADEP, and USEPA sampled during the kill in the area of the kill were in the range of 4000 mg/L in the West Virginia Fork of Dunkard Creek below the Blacksville #2 discharge to 400 mg/L further downstream in mainstem Dunkard, and upstream of the discharge.

Other ions (sulfate and magnesium) and metals (selenium) were also found to be elevated instream on our September 9 field visit. These other ions are also contributing to the high dissolved solids load, ionic stress, and total ion toxicity. EPA does not have aquatic life criteria for sulfate and magnesium, or for ion mixtures, but does recognize the toxicity of these ions, both alone and in combination with other ions.

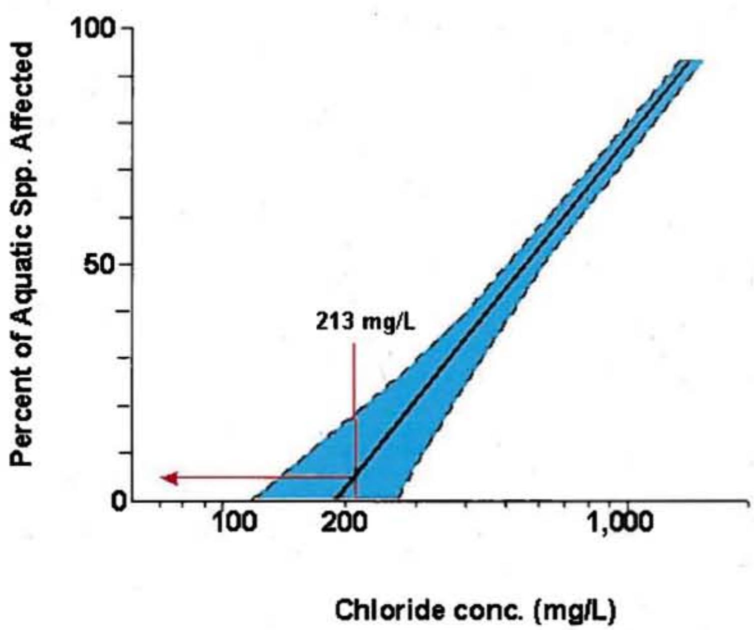

Figure 4. **From (http://www.env.gov.bc.ca/wat/wq/BCguidelines/chloride/chloride.html) Aquatic life chronic species sensitivity distribution for chloride ion based on laboratory toxicity test data (adapted from Evans and Frick, 2000). The upper and lower 95% confidence interval are also shown. Source: Bright and Addison (2002).**

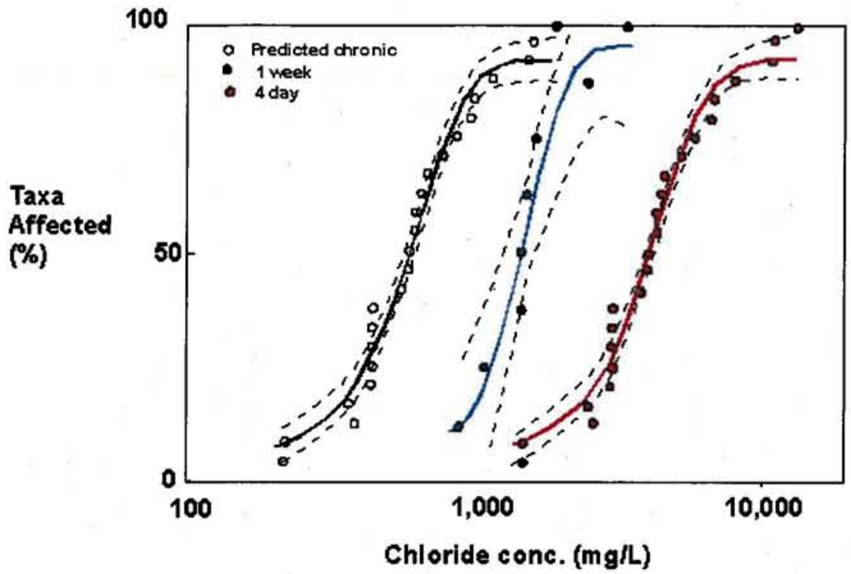

Figure 5. **From (http://www.env.gov.bc.ca/wat/wq/BCguidelines/chloride/chloride.html) Predicted chronic and actual (4 day and one week) toxicity levels for aquatic life exposed to NaCl. (upper and lower 95% confidence intervals based on a log-logistic fit are shown). Source: Bright and Addison (2002).**

Table 5. From http://www.env.gov.bc.ca/wat/wq/BCguidelines/chloride/chloride.html. **Four-day LC_{50}s of various taxa exposed to sodium chloride (adapted from Table 7-5 in Evans and Frick 2001 and Table B.6 in Bright and Addison 2002).**

Species	Common Name	96 h LC_{50} (mg Cl/L)	References
Tubifex tubifex	Tubificid worm	1 204	Khangarot, 1995
Ceriodaphnia dubia	Cladoceran	1 400	Cowgill and Milazzo, 1990
Daphnia pulex	Cladoceran	1 470	Birge et al., 1985
Ceriodaphnia dubia	Cladoceran	1 596	WI SLOH, 1995
Daphnia magna	Cladoceran	1 853	Anderson, 1948
Daphnia magna	Cladoceran	2 390	Arambasic et al., 1995
Physa gyrina	Snail	2 480	Birge et al., 1985
Lirceus fontinalis	Isopod	2 970	Birge et al., 1985
Cirrhinius mrigalo	Indian carp fry	3 021	Gosh and Pal, 1969
Labeo rohoto	Indian carp fry	3 021	Gosh and Pal, 1969
Catla catla	Indian carp fry	3 021	Gosh and Pal, 1969
Daphnia magna	Cladoceran	3 658	Cowgill and Milazzo, 1990
Cricotopus trifascia	Chironomid	3 795	Hamilton et al., 1975
Chironomus attenatus	Chironomid	4 026	Thorton and Sauer, 1972
Hydroptila angusta	Caddisfly	4 039	Hamilton et al., 1975
Daphnia magna	Cladoceran	4 071	WI SLOH, 1995

Limnephilus stigma	Caddisfly	4 255	Sutcliffe, 1961
Anaobolia nervosa	Caddisfly	4 255	Sutcliffe, 1961
Carassius auratus	Goldfish	4 453	Adelman et al., 1976
Pimephales promelas	Fathead minnow	4 600	WI SLOH, 1995
Pimephales promelas	Fathead minnow	4 640	Adelman et al., 1976
Lepomis macrochirus	Bluegill	5 840	Birge et al., 1985
Culex sp.	Mosquito	6 222	Dowden and Bennett, 1965
Pimephales promelas	Fathead minnow	6 570	Birge et al., 1985
Lepomis macrochirus	Bluegill	7 864	Trama, 1954
Gambusia affinis	Mosquito fish	10 616	Wallen et al., 1957
Anguilla rostrata	American eel	10 900	Hinton and Eversole, 1978
Anguilla rostrata	American eel	13 085	Hinton and Eversole, 1978

Table 6. **Field meter readings from USEPA sampling of Dunkard Creek on 9/9/09.**

Site #	Location	Lat (WGS83)	Long (WGS83)	Temp (C)	Sp. Cond (us/cm)	DO (mg/l)	DO sat %	pH
D1	Dunkard Creek us Dolls Run	39.71386	80.11665	19.7	2257	8.67	95.3	8.28
D2	Dunkard Creek in Pentress, WV	39.71237	80.16134	20.1	2714	13.93	154.8	8.37
D3	Dunkard Creek in Blacksville, WV	39.72027	80.2084	19.5	3259	9.52	104.7	8.2
D4	Dunkard Creek ds Miracle Run	39.71949	80.24094	19.4	3911	8.85	97.3	8.13
D5	Dunkard Creek us Morris Run	39.73042	80.25139	20.67	5085	10.36	117.4	8.39
D6	Hoovers Run (trib to Dunkard)	39.72999	80.26601	18.8	770	8.64	92.8	8.45
D7	PA Fork Dunkard at T309 Bridge	39.722	80.27048	18.95	672	7.88	84.7	8.02
D8	WV Fork Dunkard ds Consol Outfall	39.72102	80.27453	21.93	18,570	13.45	165.8	8.17
D9	Consol Outfall 005 WV 0064602	39.71864	80.27777	22.64	25,250	8.34	105.3	8.55
D10	WV Fork Dunkard us Consol Outfall	39.71863	80.27785	20.62	4957	11.54	130.7	8.13

Table 7. **Water chemistry parameters for 4 sites on Dunkard Creek.**

		Upstream of Miricle Run	Downstream of outfall 005	**OUTFALL 005**	Upstream of outfall 005	
		D4	D8	**D9**	D10	
Analyte	units					*Detection*
Aluminum	ug/L	47.6	45.4	**57.6**	135	*30*
Antimony	ug/L	U	U	**U**	U	*2*
Arsenic	ug/L	5	31.5	**42.2**	5.9	*1*
Barium	ug/L	80.2	47.2	**U**	93.3	*10*
Beryllium	ug/L	U	U	**U**	U	*1*
Cadmium	ug/L	U	U	**U**	U	*1*
Calcium	ug/L	111000	473000	**71800**	99500	*5000*
Chromium	ug/L	U	U	**U**	U	*2*
Cobalt	ug/L	1.2	2.2	**3.4**	U	*1*
Copper	ug/L	13.1	57.5	**85.7**	11.9	*2*
Iron	ug/L	205	451	**2700**	652	*100*
Lead	ug/L	U	U	**U**	U	*1*
Magnesium	ug/L	48500	229000	**37700**	32700	*500*
Manganese	ug/L	176	643	**1290**	601	*15*
Nickel	ug/L	9.2	24.7	**32**	7.3	*1*
Potassium	ug/L	8040	35800	**55300**	9550	*2000*
Selenium	ug/L	15.1	107	**146**	15.8	*5*
Silver	ug/L	U	U	**U**	U	*1*
Sodium	ug/L	786000	4040000	**5780000**	697000	*10000*
Thallium	ug/L	U	U	**U**	U	*1*
Hardness	ug/L	475000	2080000	**3000000**	383000	*3300*
Vanadium	ug/L	U	U	**U**	U	*5*
Zinc	ug/L	5.5	10.6	**11.6**	8.2	*2*
Chloride	mg/L	447	3740	**6120**	444	
Sulfate	mg/L	1360	6730	**10800**	1070	
Total Alkalinity	mg/L	162	86.2	**41.6**	180	*20*
Bicarb Alkalinity	mg/L	162	79.9	**28.7**	180	*20*
Carbonate Alkalinity	mg/L	U	U	**U**	U	*20*
Nitrite+Nitrate N	mg/L	1.8	1.08	**1.07**	U	*0.01*
TP Result	mg/L	0.076	<0.050	<0.050	0.092	

Controlling the Algae Bloom

Once *P. parvum* is established in a watershed, it is difficult to eradicate and is essentially there to stay (Karen Glenn, personal comm., September 2009).
In freshwater systems, *P. parvum* toxicity is likely affected by TDS, specific cations (e.g. calcium and magnesium have been positively correlated to toxicity), temperature, nutrients, and freshwater algae, which compete with *P. parvum* for resources. Part of the problem with increasing TDS is that native algae are stressed and cannot compete with the growth of *P. parvum*.

Laboratory studies of *P. parvum* growth corroborate these correlations and interactions. An unpublished study in Texas (http://www.tpwd.state.tx.us/landwater/water/environconcerns/hab/ga/workshop/media/kugrens.pdf) saw a decrease in growth with decreases in salinity. Baker et al. (2009) model an interaction of *Prymnesium* toxicity and phosphorous, temperature, and salinity, but conclude that these relationships may not hold at the lower salinities – the edge of the niche for *P. parvum*. The authors suggest that "(a) lower limit of salinity for population increase appears to lie between 0.5 and 1 g/L for (*P. parvum*)".

While there has been some success in controlling blooms of *P. parvum* in aquaculture situations (Rodgers, In Press) using algaecides or nutrient additions, there has been no success in controlling them in large reservoirs or rivers and streams (Karen Glenn, personal communication, September 2009). Algaecides would be toxic to a large range of resident algae and other organisms and native algae. And under non-saline conditions, native algae can compete with *P. parvum*. Adding nutrients to ambient waters during low flows in the fall could likely result in depletion of dissolved oxygen and increase ammonia levels as well as export of nutrients to downstream waters, possibly causing or contributing to water quality standards violations downstream.

Because control of TDS is not an option in most of the affected areas in Oklahoma and Texas (as many of the affected waters are naturally brackish), controlling *P. parvum* blooms through the control of TDS has not been attempted there. We believe control of TDS on Dunkard Creek and other watersheds is the best solution to control *P. parvum* blooms. Lowering TDS and chlorides in the stream would also make it easier to restore the native fauna of Dunkard Creek and decrease the loading of TDS to the Monongahela River. A water quality criterion for TDS could be developed to protect aquatic life uses. We are currently working with USEPA HQ OST to develop an aquatic life advisory level for conductivity representative of the ion matrix in alkaline mine drainage (dominated by calcium, magnesium, sulfate, and bicarbonate). We hope these efforts can be extended to consider other ion matrices like Marcellus shale brines and coalbed methane brines that contain more chloride.

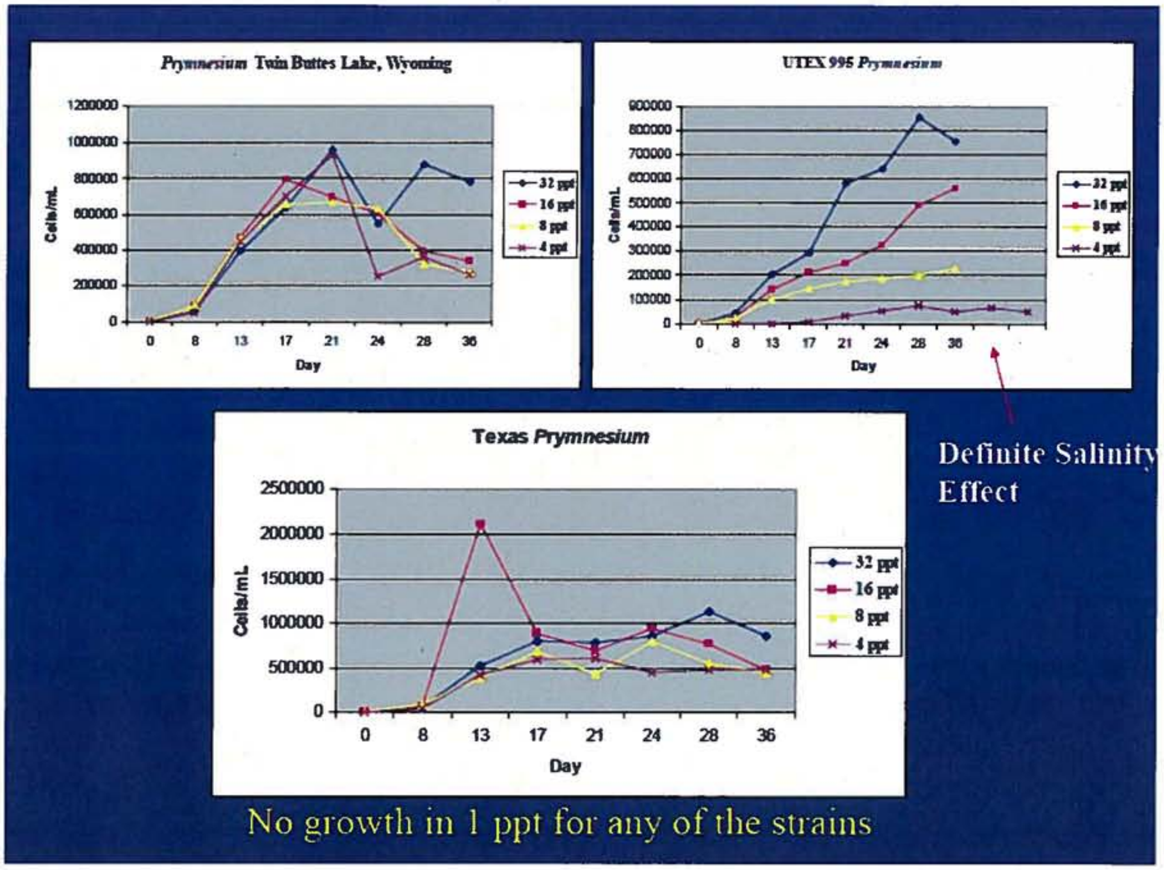

Figure 6. **From http://www.tpwd.state.tx.us/landwater/water/environconcerns/hab/ga/workshop/media/kugrens.pdf showing decreased growth with decreased salinity.**

Implications for Other Waterbodies

The map presented in the beginning of this report shows that *P. parvum* has now increased its range and Dunkard Creek will serve as a source of *P. parvum* to other freshwater bodies in the region. On October 26, CONSOL reported finding golden algae in a sample collected from Whitely Creek (the adjacent watershed to the North of Dunkard Creek) on September 29, 2009. Many natural and anthropogenic vectors can spread the algae (birds, fishermen, industrial equipment, etc.) As the algae spreads, any stream with high ionic strength in excess of 750 uS could be at risk for a *P. parvum* bloom and associated fish kill.

WVDEP has since sampled P. parvum is 32 streams in WV with high TDS levels and we, along with the PADEP, sampled for P. parvum in 9 streams and 4 mainstem Monongahela River sites.

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