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# Cool Water Off-flavor Algae and Water Quality in Four Arkansas Commercial Catfish Farms

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## Abstract

Catfish pond culture during the cool weather period of late fall, winter and early spring have experienced problems related to algal populations. Geosmin tainting by off-flavor cyanobacteria is more abundant at this time. Toxic algae episodes also appear to be more common during cool weather periods, including the devastating new golden algae (*Pyrmesium parvum*) HAB, in adjacent Texas and believed to have occurred in Arkansas. Water and phytoplankton samples were collected weekly at 0800-1000 from November to April for three consecutive years from 12 ponds at 4 commercial catfish farms located in the southeastern Arkansas. *Oscillatoria chalybea* was found from November-January at low levels and *Pseudanabaena limnetica* occurred from January-April. Results from multiple regression show that the values of the expected number of Anabaena circinalis is significantly associated with dissolved oxygen, pH, total ammonia nitrogen, nitrite nitrogen, green algae, Chlorophyll a and Pheophytin a and the expected number of *Oscillatoria chalybea* is associated with total ammonia nitrogen, un-ionized ammonia, Pheophytin a cyanobacteria, and diatoms, whereas the expected number of *Pseudanabaena limnetica* is linked to temperature, total ammonia nitrogen and un-ionized ammonia, respectively.

**Keywords**: Cyanobacteria; Pheophytina; Arkansas; Circinalis; Phytoplankton

## Introduction

Water quality during the warm catfish growing season has attracted a great deal of attention, and less so during the cool season when water quality is judged to be better due to reduced feeding (extensive management) and lower temperatures. However, winter mortalities suggest water quality issues exist, and off-flavor has been found. One of the few studies to have focused on phytoplankton and water quality during the over wintering season for catfish is that of Tucker and van Der Ploeg [1]. They evaluated phytoplankton and water quality on a monthly basis for an annual cycle in 10 commercial ponds on a central Mississippi catfish farm. They noted light was the controlling factor in phytoplankton abundance, although some ponds maintained high algal biomass through the winter. They also stated that poorest water quality occurs during the summer. Their study however did not evaluate pH or UIA, which is the most toxic compound commonly, encountered in catfish ponds. In addition, their study was of one cool weather season and consisted of once monthly sampling. Tucker and Hargreaves [2] have also speculated on the role of sediment overwintering of cyanobacteria in later dominance. Little is known on the stages leading up to massive appearance of cyanobacteria in early summer.

The algal community is affected by manmade nutrient input and herbicides, as well as natural processes. Natural processes include: climate (temperature and sunlight), water movement and turnovers, presence of competing Macrophytes and abundance of plankton-consuming macro and microorganisms. Feeding is often practiced during the warmer interludes during cool weather season, however the impact on the algal population and water quality is not known. Plankton/water quality research has focused on the growing season for fish. This is appropriate as this period is the growth period, which may be influenced by the algae and, directly and indirectly, by water quality. However, survival and health of the overwintered population in ponds is important to the success of the growing season. If the fish are in poor health going to the spring warmup period, disease can be expected, and mortalities during cool season can be substantial. Research is needed on plankton community and water quality levels during the cool weather (fall-winter-spring) season, as influenced by climatic, management and limnological conditions. Plankton populations/communities and dynamics, including those responsible for off-flavours and toxins, need to be known for catfish pond systems. Water quality levels and dynamics research is also needed to assess stress and impacts on the cultured species. Finally, examining the substrate and water samples for cyanobacteria spores and development may provide clues to the progression of cyanobacteria dominance and enable proactive controls. The objectives of this study are: 1) to compare effects of months on water quality parameters and phytoplankton; 2) to study the relationship between water quality parameters and phytoplankton in cool season; 3) to assess relationship between cool water off-flavour algae and water quality parameter and phytoplankton. In addition, this study seeks to expand the Tucker and van Der Ploeg study by adding pH, UIA and plankton of Arkansas commercial catfish ponds.

## Methods

Water and phytoplankton samples were collected from 12 production ponds at 4 commercial catfish farms located in the Mississippi River Alluvial Plain of southeastern Arkansas, and within a 16 km radius of Dumas, AR. The mean total alkalinity and hardness of commercial catfish ponds from this area ranged from 125 to 275 mg/L and 110 to 250 mg/L as $CaCO_3$ [3]. Commercial catfish farms were sampled weekly at 0800-1000 from November to May for three consecutive years. A water column sampler was submerged to 0.9 m at approximately the midpoint along one side of each pond and approximately 1.5 m from the shoreline. A single morning sampling was chosen to reduce sample time intervals between ponds, to coincide with low wind levels and reduce phytoplankton patchiness, and when dissolved oxygen levels were below saturation, Water samples were

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returned to the laboratory in 300 ml opaque Nalgene bottles filled to overflowing in an insulated ice chest, and total dissolved solids (TDS) (mg/L), total ammonia (TAN) (mg/L), dissolved oxygen (DO), and water temperature (Temp) were obtained from a multiprobe water quality instrument. TAN and nitrite nitrogen ($NO_2$-N) were determined with Hach colorimeters un-ionized ammonia (UIA) were later calculated using the formula of Emerson et al. [4]. Chlorophyll a (Chla) corrected for Pheophytin a (Pheo) were determined following APHA [5]. Phytoplankton samples were taken from the column samples and fixed with 10% formalin. Phytoplankters were identified and counted by natural units from unconcentrated samples to the genus level and species level with cyanobacteria following Cocke [6].

Pearson correlation analysis was used to identify and quantify the association between water quality parameters and phytoplankton concentrations and compositions. The influence of month on the water quality and phytoplankton parameters was assessed using one-way ANOVA and multiple linear regression to exam the effect of month, water quality parameters and phytoplankton on cool water off-flavor algae *Anabaena circinalis*, *Oscillatoria chalybea* , and *Pseudanabaena limnetica*, respectively. Cool water off-flavor algae were log 10 transformed prior to analyses. Significance level was set at $P < 0.05$. All analyses were carried out in SAS, version 9.2 (SAS Institute Inc., Cary, North Carolina, USA).

## Results and Discussion

Monthly Variations of water quality and phytoplankton parameters during the three-year combined samples were shown in (Tables 1 and 2). Ranges for water quality and phytoplankton parameters varied considerably over the cool season: DO, 4.09-15.78 mg/L; pH, 7.05-8.60; TAN, 0.01-4.42 mg/L; TDS, 290-1382 mg/L; UIA, 0.0002-0.15 mg/L; $NO_2$-N, 0-0.68; Chla, 0.02-1.832 mg/L; Pheo, 0.001-0.773 mg/L; cyanobacteria (BG), 0-241,560 units/mL; diatoms (D), 0-24,730 units/mL; Euglenoid algae (E), 0-24,430 units/mL; green algae (G), 0-21,130 units/mL.

From Pearson correlation analyses among all 13 parameters and cool seasons, a strong negative correlation (-0.74) was found between DO and Temp. Also, strong positive correlations (0.63 and 0.73) were also observed between UIA and TAN, and BG and Chla (Table 3).

Numbers of *Anabaena circinalis* ranged from 0 to 30,000 units/mL (5/17/07), *Oscillatoria chalybea* from 0 to 200 units/mL, and *Pseudanabaena limnetica* from 0 to 8000 units/mL (4/4/07). *Oscillatoria chalybea* filaments were observed in a shortened condition and in association with thickened one or two cell potential "spores" on 2/27 and 10/19. The prevalence of Anabaena circinalis was greater than *Oscillatoria chalybea* and *Pseudanabaena limnetica* during

| Water Quality Parameters | | | | | | | |
|---|---|---|---|---|---|---|---|
| **Month** | **DO (mg/L)** | **Temp (°C)** | **pH** | **TAN (mg/L)** | **TDS (mg/L)** | **UIA (mg/L)** | **$NO_2$-N (mg/L)** |
| **November** | 10.48 (0.37)bc | 12.16 (0.69)c | 7.71 (0.06)bcd | 0.96 (0.19)abc | 574.44 (56.89)ab | 0.01 (0.004)abc | 0.01 (0.02)a |
| **December** | 10.12 (0.27)c | 12.39 (0.50)c | 7.67 (0.04)cd | 1.21 (0.14)ab | 647.47 (41.39)a | 0.01 (0.003)abc | 0.09 (0.01)a |
| **January** | 11.37 (0.21)a | 7.87 (0.39)e | 7.58 (0.03)d | 1.37 (0.11)a | 601.76 (32.54)ab | 0.01 (0.001)c | 0.04 (0.01)b |
| **February** | 11.19 (0.17)ab | 9.48 (0.30)d | 7.84 (0.04)a | 0.88 (0.09)c | 553.05 (25.16)ab | 0.01 (0.002)bc | 0.02 (0.01)b |
| **March** | 8.32 (0.19)d | 16.89 (0.36)b | 7.80 (0.03)ab | 0.75 (0.10)c | 545.97 (29.49)b | 0.02 (0.002)a | 0.04 (0.01)b |
| **April** | 6.95 (0.28)e | 18.83 (0.51)a | 7.70 ( 0.04)bc | 0.90 (0.15)bc | 587.22 (42.67)ab | 0.02(0.003)ab | 0.05 (0.01)b |

**Table 1:** Mean (SE) monthly water quality parameters in 12 commercial catfish ponds. Means in columns with common letters are not significantly different; *P*>0.05. Temp=Water temperature; TAN=Total ammonia nitrogen; TDS=Total dissolved solid; UIA=Un-ionized ammonia; $NO_2$-N=nitrite-nitrogen.

| Phytoplankton | | | | | | |
|---|---|---|---|---|---|---|
| **Month** | **Chla (mg/L)** | **Pheo (mg/L)** | **BG (units/mL)** | **D (units/mL)** | **E (units/mL)** | **G (units/mL)** |
| **November** | 0.25 (0.05)ab | 0.058 (0.014)a | 3616 (6138)c | 4396 (1273)ab | 548 (506)ab | 1483 (579)a |
| **December** | 0.28 (0.04)ab | 0.067 (0.01)a | 11699 (4466)bc | 3657 (926)ab | 1621 (368)a | 1271 (421)a |
| **January** | 0.24 (0.03)b | 0.062 (0.008)a | 10514 (3543)bc | 2771 (735)b | 507 (292)b | 1054 (334)a |
| **February** | 0.23 (0.02)b | 0.056 (0.007)a | 10057 (2715)c | 3471 (563)b | 702 (224)b | 962 (256)a |
| **March** | 0.34 (0.03)a | 0.070 (0.007)a | 19761 (3182)ab | 5401 (659)a | 114 (263)b | 493 (300)a |
| **April** | 0.34 (0.04)ab | 0.061 (0.011)a | 25717 ( 4604)a | 3546 (954)ab | 237 (379)b | 995 (443)a |

**Table 2**: Mean (SE) monthly phytoplankton parameters in 12 commercial catfish ponds. Means in columns with common letters are not significantly different; *P* > 0.05.Chla = Chlorophyll *a*; Pheo = Pheophytin *a*; BG = Blue green algae; D = Diatom; E = Euglenoid algae; G = Green algae.

| | **Temp** | **DO** | **pH** | **TAN** | **TDS** | **UIA** | **$NO_2$** | **Chla** | **Pheo** | **BG** | **D** | **E** | **G** | **Total P.** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Temp | 1 | | | | | | | | | | | | | |
| DO | **-0.74*** | 1 | | | | | | | | | | | | |
| pH | -0.06 | 0.40* | 1 | | | | | | | | | | | |
| TAN | -0.41* | 0.06 | -0.35 | 1 | | | | | | | | | | |
| TDS | -0.26* | 0.11 | -0.51 | 0.25 | 1 | | | | | | | | | |
| UIA | 0.02 | -0.02 | 0.11* | **0.63*** | -0.03 | 1 | | | | | | | | |
| $NO_2$ | -0.12 | -0.14* | -0.15 | 0.33* | 0.19 | 0.44* | 1 | | | | | | | |
| Chla | 0.14* | 0.01 | 0.32* | 0.14 | 0.05 | 0.26* | 0.28 | 1 | | | | | | |
| Pheo | 0.14* | -0.15 | 0.44 | -0.24 | -0.39 | 0.04 | 0.07 | 0.48* | 1 | | | | | |
| BG | 0.32* | -0.27 | 0.06 | 0.22* | -0.04 | 0.21* | 0.23* | **0.73*** | 0.09* | 1 | | | | |
| D | 0.17* | 0.01 | 0.12 | -0.39 | -0.08 | 0.1 | -0.18* | 0.17 | -0.01 | -0.23 | 1 | | | |
| E | -0.18 | 0.22* | 0.1 | -0.14 | 0.09 | -0.07 | -0.02 | -0.04 | 0.06 | -0.21* | 0.18 | 1 | | |
| G | -0.26* | -0.08 | -0.23 | 0.51 | -0.17* | -0.02 | 0.06 | -0.33* | -0.11 | -0.20* | -0.27* | 0.03 | 1 | |
| Total P | -0.17* | -0.23* | 0.25* | -0.11 | 0.01 | 0.05 | -0.09 | 0.44* | 0.25* | **0.79*** | 0.2 | 0.12 | 0.2 | 1 |

**Table 3:** Correlations coefficients (R) among water quality parameters and plankton. Values ≥0.60 in bold *p =< 0.05. Total. P = Total phytoplankton.

| | *Anabaena circinalis* | *Oscillatoria chalybea* | *Pseudanabaena limnetica* |
|---|---|---|---|
| **Month** | **Mean (count/.mL)** | **Mean (count/mL)** | **Mean (count/mL)** |
| **November** | 50 (100) | 150 (100) | 0 (0) |
| **December** | 1225 (1812) | 50 (93) | 0 (0) |
| **January** | 462 (626) | 29 (72) | 57 (185) |
| **February** | 814 (975) | 0 (0 ) | 15 (53) |
| **March** | 3550 (2863) | 0 (0) | 38 (109) |
| **April** | 3942 (6265) | 0 (0) | 1143 (3023) |

**Table 4**: Mean (SD) of cool water off-flavor algae in four Arkansas commercial catfish farms.

December-April (Table 4). Results from multiple regressions (Table 5) shows that the values of the expected number of *Anabaena circinalis* is significantly different between month and is significantly related to DO, pH, TDS, UIA, Chla and Pheo. The expected number of *Oscillatoria chalybea* is statistically associated with DO, pH, TAN, Chla, Pheo, BD, and D, whereas the expected number of *Pseudanabaena limnetica* is linked to DO, TAN and Chla. The deviance goodness-of-fit test indicated that the multiple regression models fit the data reasonably well. The mean number of *Anabaena circinalis*, *Oscillatoria chalybea* and *Pseudanabaena limnetica* changed dramatically during the cool season. The expected number of *Anabaena circinalis* in November differed significantly from that in April. The expected number of *Oscillatoria chalybea* in November differed significantly from the rest of the cool season. The expected number of *Pseudanabaena limnetica* was significantly different among season, with lower values found in November-March. The coefficients (0.34) for DO indicates that, if every unit increases in DO while holding other variables constant, the mean number of *Anabaena circinalis* would increase by exp (0.34) = 1. While TAN is positively associated with *Oscillatoria chalybea* and negatively associated with *Anabaena circinalis* and *Pseudanabaena limnetica*, *Oscillatoria chalybea* and *Pseudanabaena limnetica* are directly related to UIA. The coefficient of 76.76 for UIA indicates that the magnitude of the effect from UIA on *Pseudanabaena limnetica* is quite large. The coefficient 0.21 for G suggests that, if every unit increases in G, mean number of *Anabaena circinalis* would increase by exp (0.21) = 1.

Water temperature during the cool season directly affected DO by increased solubility at lower temperatures. Few low DO values were observed, and generally ranged from about 6-12 mg/L. DO was higher earlier in January followed by a slight decline in April. Levels of pH were lower early in the cool season, followed by a slight increase in February and March, then a decrease again in the April. The DO and pH values measured in this study were similar to those reported in the study of commercial channel catfish pond effluents in central and west-central Alabama by Schwartz and Boyd [7]. The optimum range of DO and pH for fish and aquatic life is > 4-5 mg/L and 6.0-9.0 [8], respectively. The critical parameter UIA remained relatively low and constant in all ponds during the cool season, reflecting pH values around 7 and the low temperatures. Tucker and Van Der Pleog [1] reported that chlorophyll a can reach concentrations of 450-605 $\mu g.L^{-1}$ in commercial catfish ponds in Mississippi during the summer months. Only 15% of Chla measurements were above 450 mg/L during cool season.

The high correlation between Temp and DO (-0.74), and BG and Chla (0.73) observed in this study was also observed in commercial baitfish ponds (-0.70 and 0.67) [9]. The correlations between Total Phytoplankton (Total P.) and BG, BG and Chla in this study (0.79 and 0.73) was greater than 0.656 and 0.226 reported by Zimba et al. (2001). However, Total P. was less correlated with Cha (0.44) in our study than 0.711 reported by Zimba et al. [10].

Phytoplankton composition was fairly equitable in November (36.0%, 43.8% and 14.8%) BG, D, and G, respectively. However, BG dominated (64.1%, 70.8%, 66.2%, 76.7% and 84.3%) in Dec., Jan., Feb., Mar., and April, respectively. Only during December did E exceed G, and D exceeded G in all months. Tucker and van Der Pleog [1] found BG dominated all year long in three of ten ponds, and from July to September in other seven ponds, and E and small G were more abundant in spring [1]. In this study, the four instances of geosmin/earthy off-flavor odor were noted when Total P. (25,769 units/mL and

| Category | Coeff. | SE | t-value | p-value |
|---|---|---|---|---|
| Anabaena circinalis | | | | |
| Intercept | 32.08 | 9.13 | 3.51 | 0.0009 |
| Month | | | | |
| December | 0.81 | 0.68 | 1.19 | 0.2398 |
| January | 0.36 | 0.66 | 0.54 | 0.5932 |
| February | 0.59 | 0.62 | 0.96 | 0.3426 |
| March | 1.18 | 0.77 | 1.52 | 0.133 |
| April | 2.21 | 0.97 | 2.28 | 0.0266 |
| DO | 0.34 | 0.16 | 2.13 | 0.036 |
| pH | -4.3 | 1.3 | -3.31 | 0.0016 |
| $NO_2$ | -14.17 | 6.61 | -2.14 | 0.0364 |
| TAN | -0.63 | 0.22 | -2.88 | 0.0056 |
| G | 0.21 | 0.092 | 2.24 | 0.029 |
| Chla | 0.0028 | 0.0011 | 2.73 | 0.0044 |
| Phae | -0.013 | 0.0035 | -3.62 | 0.0006 |
| Adjusted $R^2$ | 0.47 | | | |
| *Oscillatoria chalybea* | | | | |
| Intercept | 1.29 | 0.29 | 4.51 | <.0001 |
| Month | | | | |
| December | -1.44 | 0.32 | -4.46 | <.0001 |
| January | -1.75 | 0.31 | -5.69 | <.0001 |
| February | -1.69 | 0.3 | -5.65 | <.0001 |
| March | -1.77 | 0.3 | -5.85 | <.0001 |
| April | -1.97 | 0.34 | -5.84 | <.0001 |
| TAN | 0.76 | 0.16 | 4.79 | <.0001 |
| UIA | -59.97 | 15.63 | -3.84 | 0.0003 |
| Phae | 0.006 | 0.0014 | 4.27 | <.0001 |
| BG | 0.000016 | 0.000006 | 2.86 | 0.0059 |
| D | -0.14 | 0.044 | -3.1 | 0.0029 |
| Adjusted $R^2$ | 0.51 | | | |
| *Pseudanabaena limnetica* | | | | |
| Intercept | 3.68 | 1.046 | 3.52 | 0.0008 |
| Month | | | | |
| December | 0.34 | 0.44 | 0.77 | 0.44 |
| January | 0.74 | 0.41 | 1.82 | 0.073 |
| February | -0.39 | 0.44 | -0.88 | 0.38 |
| March | 1.74 | 0.55 | 3.17 | 0.0024 |
| April | 2.86 | 0.71 | 4.01 | 0.0002 |
| Temp | -0.32 | 0.082 | -3.95 | 0.0002 |
| TAN | -1.01 | 0.19 | -5.12 | <.0001 |
| UIA | 76.76 | 14.29 | 5.37 | <.0001 |
| Adjusted $R^2$ | 0.31 | | | |

**Table 5:** Relative importance of water quality or months for the number of *Anabaena circinalis, Oscillatoria chalybea* and *Pseudanabaena limnetica*, respectively. Reference was November.

30,495 units/mL) and BG percentages (76.7% and 84.3%) were highest in March and April, respectively.

Total alkalinity and hardness in this study were similar to ponds at Stoneville in the study by Hairyadi et al. [11]. Hairiyadi et al. [11] reported BG was the prominent component of the phytoplankton at both locations (ponds at Auburn, Alabama and ponds at Stoneville, Mississippi). However, BG community composition differed between Auburn and Stoneville ponds. *Anabaena circinalis* and *Oscillatoria chalybea* both occurred at ponds at Stoneville and did not occur at ponds at Auburn. The major differences in water quality were higher pH (8.57 vs 8.32 at afternoon and 8.13 vs 7.32 at dawn), total alkalinity (244.2 vs 35.2 mg/L as $CaCO_3$), total hardness (251.8 vs 28.2 mg/L as $CaCO_3$), and dissolved inorganic phosphorus (0.107 vs 0.012 mg/L) at Stoneville and greater TAN (0.47 vs 0.12 mg/L) and nitrite-N concentration (0.016 vs 0.004 mg/L) at Auburn.

Geosmin and 2-methylisoborneol (MIB) are the most common causes of off-flavour in catfish [12,13]. These compounds are produced by species of cyanobacteria (blue-green algae) found in catfish aquaculture ponds [14]. van Der Ploeg et al. [15] found that the presence of geosmin is the main reason of off flavour in soft-water ponds (total alkalinity and hardness 15-30 mg/L as $CaCO_3$) in east Alabama and MIB in hard-water ponds (total alkalinity and hardness 150-300 mg/L as $CaCO_3$) west Mississippi. *Oscillatoria chalybea* was identified responsible for producing 2-methylisoborneol in Mississippi catfish ponds [16,17], whereas, *Anabaena circinalis* and *Pseudanabaena limnetica* were observed in commercial catfish ponds in the Mississippi-Alabama Blackland Prairie region [18]. Geosmin production by *Anabaena circinalis* has been identified in previous studies [19,20] and MIB production by *Pseudanabaena limnetica* in a drinking water reservoir in southern California [21]. No correlation was found between geosmin production and increasing light intensity or temperature [22]. However, Hurlburt et al. [23] reported that weak and positive correlation was found between geosmin concentration and air and soil temperatures at a Louisiana commercial catfish farm. Bowmer et al. [24] showed that, for Anabaena circinalis, the ratio of geosmin to biomass correlated with the change of light intensity. Moreover, geosmin concentration was strongly correlated with increasing ammonium-N nitrogen concentrations [25].

## Conclusions

Thus, although water quality was acceptable, off flavor-causing algae *Anabaena circinalis* reached levels likely to cause geomin-tainting during December, March and April. *Oscillatoria chalybea* was found from Nov-Jan at low levels and *Pseudanabaena limnetica* occurred from Jan-April. More research is needed to be conducted to confirm the findings.

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