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Research Article

Microcystins and Daily Sunlight: Predictors of Chronic Liver Disease and Cirrhosis Mortality

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Abstract

Cyanobacteria (blue-green algae) may rapidly propagate under favorable conditions, forming dense blooms. As blooms deteriorate, blue-green algae can generate potent toxins, potentially harmful to companion animals, wildlife, and humans. Microcystin is a widely studied toxin, and ingestion of contaminated drinking water is a frequent route of human exposure. The algae toxin has been detected in global drinking water supplies, particularly in regions plagued by liver disease. Microcystin production is dependent on environmental factors driven by changes in weather, including nutrient levels, pH, and water temperature. No prior study examined the ecological association between microcystins and liver disease mortality, accounting for meteorological factors. The purpose of the ecological study was to determine if meteorological factors and microcystins predicted liver disease mortality rates in the United States. Environmental data (CDC WONDER) and toxin data (USEPA) were used in multiple linear regression analyses. Mean daily sunlight and mean total microcystins significantly predicted age-adjusted chronic liver disease and cirrhosis death rates ($p < 0.05$). Mean annual precipitation ($p = 0.156$) and mean daily maximum temperature ($p = 0.149$) non-significantly predicted age-adjusted chronic liver disease and cirrhosis death rates. The study demonstrated that meteorological factors and concurrent microcystin concentrations might contribute to an increase in liver disease mortality across the United States. The results can prompt others to study environmental exposures of chronic liver diseases, guiding environmental health and the water industry of human survival needs.

Keywords: Chronic liver disease and cirrhosis; Daily sunlight; Meteorologic factors; Microcystins

Abbreviations

CDC WONDER-Centers for Disease Control and Prevention Wide-ranging Online Data for Epidemiologic Research; ELISA-Enzyme-linked immunosorbent assay; ICD-10-International Classification of Disease, Tenth Revision; NLDAS-North America Land Data Assimilation System; SPSS-Statistical Package for the Social Sciences; USEPA-United States Environmental Protection Agency; WHO-World Health Organization

1. Introduction

In the United States (US), chronic liver disease (CLD) and cirrhosis represent a significant cause of mortality. CLD and cirrhosis was the 12th leading cause of death in 2013, resulting in more than 36,000 deaths [1]. The figure has been underestimated for over two decades as researchers have indicated an annual mortality near 66,000 deaths [2]. CLD is a debilitating condition in which the liver progressively worsens for six months or greater, terminating with cirrhosis. Major causes of CLD in the US include alcoholic liver disease and hepatitis C, though non-alcoholic fatty liver disease has become the most common etiology [3, 4]. Racial/ethnic differences in CLD and cirrhosis prevalence are evident in the US, despite limited and restricted data on few racial/ethnic groups [5]. In developing countries, varied etiologies for cirrhosis have been reported, such as malnutrition, tropical infections, and toxins [6]. While certain risk factors may be region-specific, identifying CLD and cirrhosis risk factors constitutes an essential step in exposure prevention.

Microcystins are cyclic heptapeptide structures produced by cyanobacteria in aquatic environments [7, 8]. They may be released into the water column via cell rupture or after bloom senescence [9-11]. Colonial Microcystis primarily manufactures microcystin, although other freshwater cyanobacterial genera can synthesize the biotoxin [12-15]. Microcystin is a contaminant of water sources used for agriculture, drinking water, and recreation [16, 17]. Additionally, microcystin fatalities are common in animals, livestock, and pets [18, 19].

Though rare, the largest microcystin episode in humans occurred in Caruaru, Brazil, killing 52 hemodialysis patients. Cylindrospermopsins were also considered a contributing factor in hemodialysis deaths [20-24]. However, the most common exposure route for microcystin is the consumption of contaminated drinking water [25, 26]. Following oral ingestion, microcystin is transported by a bile acid transport system into the liver. Organic anion transporting polypeptides (OATP) 1A2, 1B1, and 1B3 transport microcystin into hepatocytes [27]. Herein, microcystin covalently binds protein phosphatases PP1 and PP2A, inactivating dephosphorylation capabilities. High microcystin concentrations, therefore, causes hyperphosphorylation among cytoskeletal proteins, subsequently disrupting hepatocyte structure and function [28, 29]. For example, microcystin-leucine arginine was found to stimulate hyperphosphorylation of cytokeratin 8 and 18 in primary cultured rat hepatocytes, a process associated with liver tumor promotion [30].

Furthermore, several epidemiologic investigations linked microcystin exposure to increased liver disease. Two surveys in China identified hepatotoxic blue-green algae toxins in drinking water sources as one potential risk factor for primary liver cancer [31]. In Florida, an increased risk for primary hepatocellular carcinoma occurred among persons within a serviced area of a surface water treatment plant [32]. Chronic exposure to freshwater microcystin in Three Gorges Region, China, may have induced liver damage in children [33]. On the contrary, one study determined that liver cancer was not associated with toxic cyanobacterial exposure [34]. These studies support a probable association between microcystin and liver disease, but none considered meteorological factors in their assessment.

Apart from anthropogenic eutrophication, global climate change is a key driver of cyanobacterial expansion worldwide [35]. Combustion of fossil fuels and concomitant air temperatures may enhance algae productivity. Similarly, variations in weather patterns, resulting in severe droughts and rainfall, can leach nitrates and phosphate into eutrophic waters [36]. Research has indicated a synergistic interaction between climate-related changes and increased nutrients [37]. Thus, climatic factors and nutrient levels may enhance the frequency and severity of potentially toxic blooms in freshwater ecosystems. Since climate is long-term and can trigger bloom formation and ensuing health risks, little is known about short-term meteorologic factors on liver disease. Furthermore, the influence of meteorological patterns on liver diseases has not been extensively studied in the field [38]. A population-based study determined that acute-on-chronic liver disease prevalence was influenced by lower temperatures [39]. Therefore, we conducted an ecological study to examine whether meteorological factors, including daily sunlight, daily maximum temperature, and daily precipitation, in conjunction with microcystin, associated with liver disease mortality. Understanding if meteorological factors increase a secondary factor of liver disease mortality, such as microcystin, further warranted examination.

2. Materials and Methods

Secondary data on total microcystins were collected from the 2007 United States Environmental Protection Agency (USEPA) National Lakes Assessment. An enzyme-linked immunosorbent assay (ELISA) (Abraxis, LLC, Warminster, PA) analyzed total microcystins in lake samples (limit of detection < 0.10 µg/L). A composite average of total microcystins was computed for each state by averaging two or more repeated measurements from the same county with individual measurements from different counties. Non-detectable levels of total microcystins or lack of toxin data in the dataset resulted in the exclusion of seven states (Alaska, Hawaii, New Hampshire, New Mexico, South Carolina, Vermont, Wyoming). Mean total microcystins in each state were compared against the WHO relative probable health risk due to microcystins (low, moderate, high).

Environmental data on annual precipitation, average daily max temperature, daily precipitation, and daily sunlight, derived from the North America Land Data Assimilation System (NLDAS) (1979-2011), were gathered from the Centers for Disease Control and Prevention Wide-ranging Online Data for Epidemiologic Research (CDC WONDER). Data from 2007 were used to coincide with concentrations of total microcystins. The Underlying Cause of Death database was used to retrieve age-adjusted CLD and cirrhosis death rates of the United States between

2003 and 2007. The International Classification of Disease, Tenth Revision (ICD-10) 113 Cause List was utilized to examine records of age-adjusted CLD and cirrhosis death rates (K70, K73-K74). All ages, genders, origins, and races were selected in the demographics of age-adjusted CLD and cirrhosis death rates (Table A1). Multiple linear regressions were performed in Statistical Package for the Social Sciences (SPSS) version 25. Normality was achieved by log-transforming (base 10) all variables in the analysis.

Further examination identified extraneous variables within the dataset. The removal of outliers resulted in 35 states in the final analysis (Table A2). Statistical significance was based on $p < 0.05$. Descriptive statistics on mean total microcystins and mean meteorological factors were grouped by census region and state. Inferential statistics were applied to aggregate national data to assess the ecological association between total microcystins, meteorological factors, and age-adjusted CLD and cirrhosis death rates.

3. Results

3.1 Mean total microcystins and meteorological factors by census region

Table 1 displays a summary of mean total microcystins and meteorological factors by census region in 2007. Mean total microcystins was highest in the Midwest, with a concentration of 3.90 µg/L. The South and West had mean total microcystins of 0.858 µg/L and 1.99 µg/L, respectively. Mean total microcystins was lowest in the Northeast, at 0.688 µg/L. Mean daily maximum temperature ranged between 15.44 C in the Midwest to 22.10 C in the South. The West received the least mean daily precipitation at 1.28 mm, while the Northeast received the most at 3.06 mm. Mean daily sunlight ranged from 15064.91 KJ/m^2 in the Northeast to 17635.69 KJ/m^2 in the West.

Census region	**Mean total microcystins (µg/L)**	**WHO relative probable health risk**	**Mean daily maximum temperature (C)**	**Mean daily precipitation (mm)**	**Mean daily sunlight (KJ/m^2)**
South *n* =15	0.851 σ = 0.60	Low	22.10	2.71	17171.66
Northeast *n* = 7	0.688 σ = 0.29	Low	15.98	3.06	15064.91
Midwest *n* = 12	3.90 σ = 5.60	Low	15.44	2.33	15409.55
West *n* = 9	2.00 σ = 1.95	Low	15.52	1.28	17635.69

Table 1: Summary of mean total microcystins and mean meteorological factors by census region in 2007. *n* = number of states, σ = standard deviation, WHO = World Health Organization, Low = 0.10 µg/L ≤ 10 µg/L. C = Celsius, mm = millimeters, KJ/m^2 = Kilojoule per square meter.

State	Mean total microcystins (µg/L)	WHO relative probable health risk	Mean daily maximum temperature (C)	Mean daily precipitation (mm)	Mean daily sunlight (KJ/m^2)
Alabama	0.33 *n* = 1	Low	24.83	2.43	17761.61
Arizona	1.0 *n* = 1	Low	22.62	0.85	19804.18
Arkansas	0.885 *n* = 2	Low	22.93	3.19	16681.82
California	0.22 *n* = 4	Low	21.0	0.99	19698.04
Colorado	2.73 *n* = 3	Low	13.66	1.36	17497.51
Connecticut	0.343 *n* = 6	Low	14.22	3.11	15452.60
Delaware	0.58 *n* = 6	Low	17.63	2.48	16249.63
Florida	1.62 *n* = 12	Low	27.28	3.09	18945.54
Georgia	0.31 *n* = 7	Low	24.80	2.47	18231.50
Idaho	3.04 *n* = 5	Low	12.30	1.35	16188.47
Illinois	1.47 *n* = 15	Low	17.77	2.56	15591.87
Indiana	0.55 *n* = 32	Low	17.36	2.93	15603.23
Iowa	0.69 *n* = 14	Low	15.06	2.82	15311.84
Kansas	0.98 *n* = 5	Low	19.16	2.57	16770.71
Kentucky	0.76 *n* = 2	Low	20.20	2.89	16220.59
Louisiana	0.631 *n* = 8	Low	25.37	3.71	17654.09
Maine	0.845 *n* = 5	Low	9.41	3.25	14242.49
Maryland	0.267 *n* = 3	Low	17.55	2.48	16034.71
Massachusetts	0.903 *n* = 2	Low	31.12	3.06	15315.42
Michigan	1.26 *n* = 23	Low	12.65	2.09	14985.34
Minnesota	1.79 *n* = 39	Low	11.77	1.83	14622.10
Mississippi	0.465 *n* = 2	Low	24.88	2.87	17554.24
Missouri	0.20 *n* = 11	Low	19.21	2.92	15957.14
Montana	1.27 *n* = 15	Low	12.22	1.30	15080.89
Nebraska	4.52 *n* = 28	Low	16.62	2.11	16054.05
Nevada	0.53 *n* = 1	Low	16.08	0.54	18346.94
New Jersey	0.703 *n* = 3	Low	16.28	3.25	15758.56
New York	0.593 *n* = 4	Low	11.92	3.06	14393.31
North Carolina	0.266 *n* = 12	Low	21.55	2.34	17402.86
North Dakota	18.18 *n* = 38	Moderate	12.02	1.40	14816.28
Ohio	13.91 *n* = 6	Moderate	16.41	2.75	15197.93
Oklahoma	1.03 *n* = 15	Low	21.59	3.09	16921.44
Oregon	1.18 *n* = 6	Low	13.71	1.84	16404.71
Pennsylvania	1.17 *n* = 7	Low	14.30	2.91	14594.54
Rhode Island	0.26 *n* = 4	Low	14.65	2.81	15697.50
South Dakota	2.53 *n* = 28	Low	14.88	1.61	15374.59
Tennessee	0.75 *n* = 4	Low	21.84	2.40	16648.09
Texas	2.48 *n* = 13	Low	24.95	2.52	17999.03
Utah	6.94 *n* = 4	Low	15.35	0.85	17701.46
Virginia	0.691 *n* = 6	Low	19.27	2.35	16634.94
Washington	1.14 *n* = 5	Low	12.76	2.52	17999.03
West Virginia	1.70 *n* = 1	Low	16.87	2.35	16634.94
Wisconsin	0.735 *n* = 16	Low	12.38	2.39	14629.55

Table 2: Summary of mean total microcystins above 0.10 µg/L and mean meteorological factors by state in 2007. *n* = number of measurements ≥ 0.10 µg/L, WHO = World Health Organization, Low = 0.10 µg/L ≤ 10 µg/L, Moderate = 10 ug/L ≤ 20 µg/L. C = Celsius, mm = millimeters, KJ/m^2 = Kilojoule per square meter.

3.2 Mean total microcystins and meteorological factors by census region

Mean total microcystins and mean meteorological factors by state in 2007 are depicted in Table 2. The mean total microcystins for all 43 states was 1.91 µg/L. The lowest mean total microcystins occurred in Missouri (0.20 µg/L), and the highest mean total microcystins occurred in North Dakota (18.18 µg/L). 41 states (95.35%) had a low relative probable health risk, while 2 states (4.65%) had a moderate relative probable health risk. For meteorological factors, mean daily maximum temperature reached 17.87 C, mean daily precipitation 2.36 mm, and mean daily sunlight was 16434.07 KJ/m^2.

State	Region	Age-adjusted chronic liver disease and cirrhosis death rates per 100,000
Alabama	South	9.6
Arizona	West	11.9
Arkansas	South	8.0
California	West	11.2
Colorado	West	9.9
Connecticut	Northeast	7.5
Delaware	South	8.6
Florida	South	10.5
Georgia	South	8.0
Idaho	West	9.1
Illinois	Midwest	8.2
Indiana	Midwest	7.6
Iowa	Midwest	6.2
Kansas	Midwest	7.4
Kentucky	South	8.3
Louisiana	South	7.9
Maine	Northeast	8.4
Maryland	South	7.5
Massachusetts	Northeast	7.8
Michigan	Midwest	9.4
Minnesota	Midwest	6.4
Mississippi	South	8.7
Missouri	Midwest	7.0
Montana	West	11.0
Nevada	West	11.1
New Jersey	Northeast	7.6
New York	Northeast	6.3
North Carolina	South	8.7
Oklahoma	South	11.2
Oregon	West	10.3
Pennsylvania	Northeast	7.6
Rhode Island	Northeast	9.4
South Dakota	Midwest	10.7
Tennessee	South	10.0
Texas	South	11.4
Virginia	South	7.4
Washington	West	9.0

Table 3: Age-adjusted chronic liver disease and cirrhosis death rates per 100,000 from 2003 to 2007 by state.

3.3 Regression models

Multiple linear regressions were run to assess the predictive function of meteorological factors and total microcystins on age-adjusted CLD and cirrhosis death rates. All predictors were initially merged into the model. A positive association was observed between mean total microcystins, meteorological factors, and liver disease mortality (R = 0.726). Approximately 46.4% (R^2 = 0.464) of variance in age-adjusted CLD was explained by the predictors. The simultaneous model partially supported the hypothesis that meteorological factors in concurrence with total microcystins predict liver disease mortality (Table 4). The stepwise method was selected to determine which explanatory variables fitted the regression model. In Table 4, the final model revealed a positive association between mean daily sunlight, total microcystins, and age-adjusted CLD and cirrhosis death rates (R = 0.676 and R^2 = 0.423). Mean daily maximum temperature and mean daily precipitation were not statistically significant predictors ($p > 0.05$) (Table 5).

Model	R	R^2	***F*-change**
Simultaneous	0.726	0.464	0.000117
Stepwise	0.676	0.423	0.009

Table 4: Multiple linear regressions of exposure correlates and liver disease mortality.

Variables	**β**	***p***
Total Microcystins	0.365	0.009
Daily Sunlight	0.621	0.000044
Daily Maximum Temperature	-0.290	0.149
Daily Annual Precipitation	-0.188	0.156

Table 5: Coefficients of predictors for liver disease mortality.

4. Discussion

Liver disease is a serious health problem in the United States. In 2013, CLD and cirrhosis claimed over 33,000 lives, making it the 12th leading cause of death [1]. Liver disease mortality estimates remain conservative, although research suggests that nearly 66,000 individuals die from CLD and cirrhosis each year [2]. The condition is largely preventable, with alcohol, obesity, and viral hepatitis being three major risk factors [40]. Other risk factors, such as toxins, tropical infections, and malnutrition occur in developing countries [6]. These environmental factors may increase in developed countries due to constant lifestyle and weather changes.

Microcystin is a blue-green algal hepatotoxin secreted by freshwater cyanobacteria. When favorable environmental conditions persist within stagnant waters, cyanobacteria multiply to create thick blooms. Bloom senescence can promote microcystin release as cells lyse in water. Microcystin is hepatotoxic since it targets protein phosphates in hepatocytes upon oral ingestion of contaminated drinking water. Inactivation of protein phosphatases PP1 and PP2A stimulates hyperphosphorylation, which can drastically affect liver function [28-30].

Moreover, epidemiological studies indicate microcystin exposure may associate with liver disease. Yet, limited knowledge exists on meteorological factors and chronic liver disease. For instance, average humidity and temperature were shown to correlate with acute-on-chronic liver failure positively and negatively, respectively [39]. The present study explored the ecological association between microcystins, meteorological factors, and liver disease mortality. Study results demonstrated a positive association between daily sunlight, total microcystins, and age-adjusted CLD and cirrhosis death rates. The observed association mirrored previous epidemiological investigations connecting freshwater microcystins with liver disease [31-33]. Daily sunlight exposure was a strong predictor of age-adjusted CLD and cirrhosis death rates. In a different study examining sunlight exposure and end stage renal disease, low amounts of daily sunlight increased the risk of all-cause mortality in dialysis patients, especially among diabetics and individuals age 75 and older [41].

There were a few limitations inherent within the study. First, the study was ecological in design, meaning interpreted results strictly concerned populations, not individuals. The ecological fallacy is a major limitation of an ecological study in which inferences from group data generalize among individuals. In the study context, people who consume drinking water contaminated with microcystin and receive sunlight exposure over their lifetime may die from liver disease. However, one should note that people drink treated water sourced from potable supplies, and CLD and cirrhosis may result from a combination of environmental and health risk factors. Another limitation involves confounding bias, which is not uncommon in ecological studies. The current study lacked known risk factors for CLD and cirrhosis, such as alcohol consumption and obesity. The variables were omitted due to incomplete data of risk factors in the collected secondary data sources. Missing pertinent variables in regression models can increase or decrease the predictivity of exposure variables on a health outcome. Hence, the effect of total microcystin and daily sunlight exposure is perhaps higher than expected if other risk factors were embedded in the model. A third limitation rests with ELISA for total microcystins quantitation. The method is subject to cross-reactivity, matrix interference, and low specificity despite screening multiple samples concurrently. Consequently, the identity of specific congeners and their associated concentrations in lake samples is unknown. Microcystin detection via liquid chromatography mass spectrometry can offer valuable information by distinguishing various congeners in the environment [42].

Liver disease is a major public health problem and continues to grow as population growth increases and people age. The study identified a positive association between mean total microcystins, daily sunlight exposure, and age-adjusted CLD and cirrhosis death rates. The association, however, does not imply causation. Future research should consider behavioral and environmental lifestyle factors when exploring associations between meteorological factors, hepatotoxins, and liver disease mortality. Findings may encourage routine biomonitoring practices in endemic areas of liver disease where lakes bloom. Health and medical professionals can use the results to aid in the prevention, diagnosis, and treatment of liver diseases.

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Conflict of Interest

The author declares no conflict of interest.

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Appendix

State	Race	Gender	Deaths	Population	Age-adjusted chronic liver disease and cirrhosis death rates rate per 100,000
Alabama	Black or African American	Female	136	3248829	4.4
Alabama	Black or African American	Male	282	2825850	12
Alabama	White	Female	721	8359324	7
Alabama	White	Male	1238	8072164	13.7
Arizona	American Indian or Alaska Native	Female	204	804525	33.7
Arizona	American Indian or Alaska Native	Male	303	780419	55
Arizona	Black or African American	Female	17	586581	4.0 (Unreliable)
Arizona	Black or African American	Male	33	638129	9.7
Arizona	White	Female	995	12817699	7
Arizona	White	Male	1957	12724498	15.1
Arkansas	Black or African American	Female	48	1167578	4.7
Arkansas	Black or African American	Male	69	1052648	7.9
Arkansas	White	Female	365	5782643	5.2
Arkansas	White	Male	717	5626527	11.6
California	American Indian or Alaska Native	Female	118	1450691	11.2
California	American Indian or Alaska Native	Male	205	1467249	19.9
California	Asian or Pacific Islander	Female	284	12721237	2.4
California	Asian or Pacific Islander	Male	525	11690059	5.3
California	Black or African American	Female	363	6642153	6
California	Black or African American	Male	701	6428279	13.4
California	White	Female	5512	69075470	7.8
California	White	Male	11555	69452053	17.7
Colorado	American Indian or Alaska Native	Female	20	178431	17.1
Colorado	American Indian or Alaska Native	Male	27	183167	17.9
Colorado	Black or African American	Female	12	511727	2.8 (Unreliable)
Colorado	Black or African American	Male	46	554676	11.5
Colorado	White	Female	773	10516402	7.1
Colorado	White	Male	1384	10603678	13.3
Connecticut	Black or African American	Female	41	999520	4.9
Connecticut	Black or African American	Male	53	911084	8
Connecticut	White	Female	478	7656792	4.9
Connecticut	White	Male	870	7261770	10.9
Delaware	Black or African American	Female	13	478336	3.2 (Unreliable)
Delaware	Black or African American	Male	36	429534	10.5
Delaware	White	Female	133	1619511	6.7
Delaware	White	Male	204	1546378	11.9
Florida	American Indian or Alaska Native	Male	16	222951	8.7 (Unreliable)
Florida	Asian or Pacific Islander	Female	10	1179080	1.3 (Unreliable)
Florida	Asian or Pacific Islander	Male	32	1037038	4.3
Florida	Black or African American	Female	275	7452040	4.4
Florida	Black or African American	Male	477	6914609	9.1
Florida	White	Female	3505	36539331	7.2
Florida	White	Male	6627	35245038	15.6
Georgia	Asian or Pacific Islander	Male	15	671654	4.0 (Unreliable)
Georgia	Black or African American	Female	273	7211431	4.6
Georgia	Black or African American	Male	386	6395644	8.4
Georgia	White	Female	977	14854398	6
Georgia	White	Male	1728	14791423	11.9
Idaho	American Indian or Alaska Native	Female	24	61511	54.7
Idaho	American Indian or Alaska Native	Male	20	61828	38.1
Idaho	White	Female	209	3424909	5.9
Idaho	White	Male	390	3447041	11.6
Illinois	American Indian or Alaska Native	Male	13	160226	11.8 (Unreliable)
Illinois	Asian or Pacific Islander	Female	26	1426761	2.9
Illinois	Asian or Pacific Islander	Male	33	1356464	3
Illinois	Black or African American	Female	269	5161785	5.7

Illinois	Black or African American	Male	472	4568874	12.6
Illinois	White	Female	1618	25402857	5.6
Illinois	White	Male	2768	24861441	11.1
Indiana	Black or African American	Female	64	1526741	5
Indiana	Black or African American	Male	129	1414048	12
Indiana	White	Female	814	14149157	5.1
Indiana	White	Male	1466	13743938	10.5
Iowa	American Indian or Alaska Native	Female	11	32209	55.4 (Unreliable)
Iowa	American Indian or Alaska Native	Male	12	31776	55.8 (Unreliable)
Iowa	Black or African American	Male	14	225798	9.5 (Unreliable)
Iowa	White	Female	365	7163517	4.2
Iowa	White	Male	599	6937325	8
Kansas	American Indian or Alaska Native	Male	14	87936	20.4 (Unreliable)
Kansas	Black or African American	Female	15	439847	4.1 (Unreliable)
Kansas	Black or African American	Male	29	449629	8.7
Kansas	White	Female	350	6250696	4.9
Kansas	White	Male	643	6103273	10.2
Kentucky	Black or African American	Female	28	849334	3.7
Kentucky	Black or African American	Male	67	822665	11
Kentucky	White	Female	596	9661234	5.3
Kentucky	White	Male	1142	9301818	11.8
Louisiana	Black or African American	Female	135	3843199	3.9
Louisiana	Black or African American	Male	303	3465126	10.6
Louisiana	White	Female	465	7362405	5.3
Louisiana	White	Male	880	7151226	11.5
Maine	White	Female	243	3275144	5.9
Maine	White	Male	412	3126808	11.4
Maryland	Asian or Pacific Islander	Female	14	769240	2.5 (Unreliable)
Maryland	Asian or Pacific Islander	Male	14	706625	2.9 (Unreliable)
Maryland	Black or African American	Female	162	4476007	3.9
Maryland	Black or African American	Male	353	3908074	10.4
Maryland	White	Female	586	9119527	5.4
Maryland	White	Male	1032	8797543	10.9
Massachusetts	Black or African American	Female	35	1251232	3.4
Massachusetts	Black or African American	Male	76	1163882	9.4
Massachusetts	White	Female	905	14423604	5.1
Massachusetts	White	Male	1692	13483281	11.7
Michigan	American Indian or Alaska Native	Female	34	200968	21.8
Michigan	American Indian or Alaska Native	Male	35	195793	21.1
Michigan	Asian or Pacific Islander	Male	19	604778	5.2 (Unreliable)
Michigan	Black or African American	Female	226	3916102	6.2
Michigan	Black or African American	Male	441	3546973	14.8
Michigan	White	Female	1442	20806241	6
Michigan	White	Male	2776	20296999	12.8
Minnesota	American Indian or Alaska Native	Female	47	175152	36.5
Minnesota	American Indian or Alaska Native	Male	48	175077	36.4
Minnesota	Asian or Pacific Islander	Male	12	476958	5.8 (Unreliable)
Minnesota	Black or African American	Female	20	622804	5.7
Minnesota	Black or African American	Male	22	656943	6.1
Minnesota	White	Female	567	11618936	4.3
Minnesota	White	Male	974	11408579	8.2
Mississippi	American Indian or Alaska Native	Male	14	37529	43.6 (Unreliable)
Mississippi	Black or African American	Female	89	2838543	3.7
Mississippi	Black or African American	Male	192	2521161	9.8
Mississippi	White	Female	356	4527502	6.5
Mississippi	White	Male	625	4405730	13
Missouri	Black or African American	Female	71	1830161	4.4
Missouri	Black or African American	Male	113	1643300	9.2
Missouri	White	Female	678	12661755	4.5

Missouri	White	Male	1296	12190948	10
Montana	American Indian or Alaska Native	Female	78	161583	60.1
Montana	American Indian or Alaska Native	Male	56	159717	45
Montana	White	Female	152	2157596	5.9
Montana	White	Male	290	2163816	11.5
Nevada	American Indian or Alaska Native	Female	17	103775	22.1 (Unreliable)
Nevada	American Indian or Alaska Native	Male	28	102707	32.8
Nevada	Asian or Pacific Islander	Female	12	489734	3.1 (Unreliable)
Nevada	Asian or Pacific Islander	Male	14	418091	4.6 (Unreliable)
Nevada	Black or African American	Female	19	513466	4.0 (Unreliable)
Nevada	Black or African American	Male	43	524606	10.9
Nevada	White	Female	389	4884637	7.5
Nevada	White	Male	870	5113929	16.1
New Jersey	Asian or Pacific Islander	Female	28	1682503	2.8
New Jersey	Asian or Pacific Islander	Male	45	1620193	3.7
New Jersey	Black or African American	Female	148	3415394	4.6
New Jersey	Black or African American	Male	244	3049039	10.2
New Jersey	White	Female	1090	17003539	5.2
New Jersey	White	Male	1968	16244174	11.2
New York	American Indian or Alaska Native	Female	17	418514	5.6 (Unreliable)
New York	American Indian or Alaska Native	Male	26	410289	9.5
New York	Asian or Pacific Islander	Female	34	3493849	1.3
New York	Asian or Pacific Islander	Male	109	3328806	4
New York	Black or African American	Female	274	9298457	3
New York	Black or African American	Male	581	8005171	8.9
New York	White	Female	1855	36260895	4.2
New York	White	Male	3529	34501101	9.6

Table A1: Demographics of age-adjusted chronic liver disease and cirrhosis death rates per 100,000 in the United States from 2003 to 2007. Death rates with a numerator of 20 or less are flagged as unreliable.

State	Mean Total Microcystins (µg/L)	Mean Daily Maximum Temperature (C)	Mean Daily Precipitation (mm)	Mean Daily Sunlight (KJ/m^2)	Age-Adjusted Chronic Liver Disease and Cirrhosis Death Rates Per 100,000
Alabama	0.33	24.83	2.43	17761.61	9.6
Arizona	1.00	22.62	0.85	19804.18	11.9
Arkansas	0.885	22.93	3.19	16681.82	8.0
California	0.22	21.0	0.99	19698.04	11.2
Colorado	2.73	13.66	1.36	17497.51	9.9
Connecticut	0.343	14.22	3.11	15452.60	7.5
Delaware	0.58	17.63	2.48	16249.63	8.6
Florida	1.62	27.28	3.09	18945.54	10.5
Georgia	0.31	24.80	2.47	18231.50	8.0
Idaho	3.04	12.30	1.35	16188.47	9.1
Illinois	1.47	17.77	2.56	15591.87	8.2
Indiana	0.55	17.36	2.93	15603.23	7.6
Iowa	0.69	15.06	2.82	15311.84	6.2
Kansas	0.98	19.16	2.57	16770.71	7.4
Kentucky	0.76	20.20	2.89	16220.59	8.3
Louisiana	0.631	25.37	3.71	17654.09	7.9
Maine	0.845	9.41	3.25	14242.49	8.4
Maryland	0.267	17.55	2.48	16034.71	7.5
Massachusetts	0.903	31.12	3.06	15315.42	7.8
Michigan	1.26	12.65	2.09	14985.34	9.4
Minnesota	1.79	11.77	1.83	14622.10	6.4
Mississippi	0.465	24.88	2.87	17554.24	8.7
Missouri	0.20	19.21	2.92	15957.14	7.0
Montana	1.27	12.22	1.30	15080.89	11.0
Nevada	0.53	16.08	0.54	18346.94	11.1
New Jersey	0.703	16.28	3.25	15758.56	7.6
New York	0.593	11.92	3.06	14393.31	6.3

Table A2: Mean total microcystins and mean meteorological factors from 2007 and age-adjusted chronic liver disease and cirrhosis death rates from 2003 to 2007 in the United States.