Development of an Informatics Algorithm to Link Seasonal Infectious Diseases to Birth-Dependent Diseases Across Species: A Case Study with Osteosarcoma

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Abstract

Many diseases have been linked with birth seasonality, and these fall into four main categories: mental, cardiovascular, respiratory and women's reproductive health conditions. Informatics methods are needed to uncover seasonally varying infectious diseases that may be responsible for the increased birth month-dependent disease risk observed. We have developed a method to link seasonal infectious disease data from the USA to birth month dependent disease data from humans and canines. We also include seasonal air pollution and climate data to determine the seasonal factors most likely involved in the response. We test our method with osteosarcoma, a rare bone cancer. We found the Lyme disease incidence was the most strongly correlated significant factor in explaining the birth month-osteosarcoma disease pattern ($R=0.418$, $p=2.80X10^{-23)}$, and this was true across all populations observed: canines, pediatric, and adult populations.

1. Introduction

1.1 Importance of Birth Month on Disease Risk

Prior research has established that birth seasonality affects disease risk for diseases and conditions falling into four broad categories (1): mental, cardiovascular, respiratory and women's reproductive health conditions (2). The etiologies underlying the relationship between birth seasonality (and or month) and each disease vary depending on the type of disease. For example, exposure to air pollution during the early stages of development (first trimester in humans) has been implicated in increased risk of cardiovascular disease in humans (3) and a similar finding was found in dogs (4). Additionally, climate factors have been implicated in the birth month - disease relationship. For example, climate factors (heat and humidity) are responsible for an increase in the dust mite population have been implicated in increased risk of asthma during certain birth months validating early work in this area (1,5).

1.2 Seasonality of Infectious Diseases May Play a Role

Another seasonally varying factor that may play a role in birth month - disease relationships is infectious diseases. Infectious diseases fall into two main categories based on their underlying causes: bacterial and viral. Prior work had investigated the relationship between flu in Asia and the USA across six sites to uncover relationships between flu and birth season-disease relationships (3). However, the results of this work revealed that flus often differ across the globe, and therefore, the specific strains of a flu may be important in understanding its specific role in the etiology of birth month dependent diseases.

1.3 Literature Gap: Country-Specific Information on Infectious Diseases Needed

A major gap of our prior work and the literature is the lack of detailed exploration of the role of infectious diseases, both bacterial and viral, on birth month dependent diseases. To properly address this issue, we need to catalog all of the seasonal bacterial and viral infectious diseases and conditions that occur in the USA. This must be done regional (e.g., for USA and separately for Asia, and so forth) to properly capture the local seasonality of each infectious disease and virus in that particular region. Therefore, the first step of this informatics algorithm is to mine the publicly available data from the Centers for Disease Prevention and Control (CDC) to establish a set of seasonally varying infectious diseases and viruses. These will form our dataset of potential factors in birth month dependent diseases.

1.4 Case Study: Osteosarcoma

Osteosarcoma is a form of cancer that originates in bone. In the United States, approximately 800 new osteosarcoma cases are diagnosed in humans each year, about half of which occur in patients under twenty years old (6). Osteosarcoma incidence is largely bimodal, with 10% of new cases occurring in patients aged 60 and older (7). In older adults, osteosarcoma is often linked to an existing, underlying condition (8), many of which are explicitly bone-related. Although rare, we found that osteosarcoma varied by birth month in both dogs and humans. The birth month-disease risk curve was the same in dogs and humans indicating that the culprit exposure was likely perinatal (i.e., at the time of birth). If the exposure were prenatal, we would have expected to see a shift in the birth month-disease curves between dogs and humans due to their different gestational lengths (2 vs. 9 months) (9).

1.5 Purpose of Study

The purpose of this study is to develop an informatics method to link diseases caused by seasonal viruses and bacteria within the United States to birth month-dependent diseases. Our method utilizes publicly available data and links prior methods that utilize seasonally varying pollution and climate factors. Our method finds the most significant seasonal factor associated with a birth month dependent disease whether it be a seasonal infection, pollutant, or climate factor (e.g., rainfall).

2. Materials and Methods

This process collected data from the Children's Hospital of Philadelphia (CHOP), Penn Medicine, and Penn Vet, health systems, and public data available from US government sources including the CDC, NOAA, and EPA. The collection and analysis methods are visualized in **Figure 1**.

2.1 Obtaining Seasonal Infection Data

2.1.1 National Data

We created a list of potentially seasonal viruses and bacteria to investigate. Shiga toxin-producing *Escherichia coli*, legionellosis, listeriosis, Lyme disease, pertussis, measles, mumps, rubella, salmonellosis, tetanus, and tuberculosis (named as listed in the CDC tables) were included. We obtained annual data categorized by month over a ten-year period from web-based CDC resources.

Data from 2009-2015 was collected from the CDC's Morbidity and Mortality Weekly Report (MMWR) Summary of Notifiable Infectious Diseases and Conditions — United States (10). Data from 2016-2018 was collected from the CDC's Wide-ranging Online Data for Epidemiologic Research (WONDER) Nationally Notifiable Infectious Diseases and Conditions, United States: Annual Tables (11). These resources presented the total number of reported cases for each month in a series of columns, with the last column as the total. Between the 'December' and 'Total' columns was a space for cases that were included in the total for the year, but not categorized within a month. However, this space remained blank, presuming that all counted cases in the year were reported within a specific month. The tabular, numeric values were compiled and saved in disease-specific tables for further analysis. The CDC also has a national outbreak database tool, the National Outbreak Reporting System (NORS) Dashboard (12). This tool provides insight into foodborne illnesses outbreaks including E. coli, listeriosis, and salmonellosis; allowing the user to sort data by year, setting, and vector. These data were helpful for identifying strong categories for further investigating case counts across certain food groups, although the aggregate case counts were more definitive than the stratified.

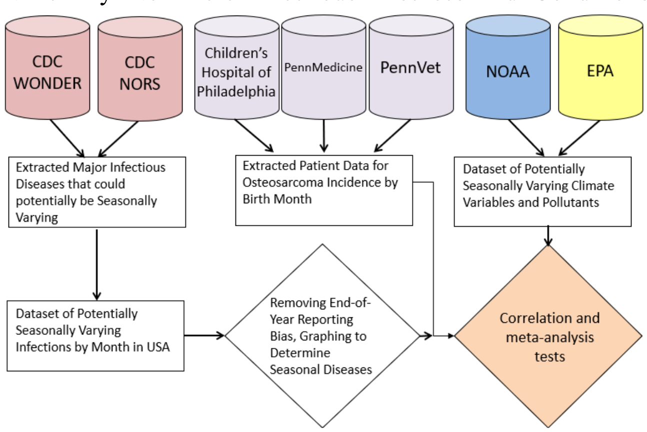

Figure 1. Overview of Our Method to Determine and Analyze Seasonal Diseases and Factors Related to Osteosarcoma Birth Season.

2.1.2 Regional Data

Lyme disease is prevalent in the northeast and northcentral parts of the United States (13). Because the osteosarcoma CHOP dataset comes from patients diagnosed in Pennsylvania, which has thousands of Lyme disease cases each year, state-level data were collected from the Pennsylvania Department of Health (14). Lyme disease seasonality data by month in Pennsylvania were only available from a 2017 report, unlike all other diseases analyzed, which had 10 years of data to average. County-level data from 2009-2018 in Pennsylvania were modified only by labelling case counts under their respective 5-digit Federal Information Processing Standard codes before analysis.

To investigate potential geographic similarities in Lyme disease and osteosarcoma distribution, osteosarcoma case counts were collected by state (15).

2.2 Obtaining Seasonal Pollution and Climate Data

Because all osteosarcoma cases were diagnosed in Philadelphia, Pennsylvania, county climate and pollutant data for the city were collected from NOAA (16). Six climate variables, measured by month, were factored into the analysis, including: mean sunshine hours, high and low temperatures, relative humidity, precipitation days, and precipitation inches. Five pollutant variables, measured daily and averaged by month, were included; SO2, CO, NO2, PM 2.5, and ozone Air Quality Index values (17). **Table 1** includes the data sources for all regional and national infection, climate and pollution data.

Table 1. Sources of All Exposure Variables

Exposure	Study Site	Collection Site	Collection Period	Source	Ref.
Infectious Disease					
E. coli	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
	Multi-state	State-based, reported to CDC	2009-2019	CDC	(18)
Legionellosis	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Listeriosis	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
	Multi-state	State-based, reported to CDC	2009-2019	CDC	(19)
Lyme	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
	Pennsylvania	Pennsylvania, USA	2017	PA DOH	(14)
Measles	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Mumps	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Pertussis	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Rubella	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Salmonellosis	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
	Multi-state	State-based, reported to CDC	2009-2019	CDC	(20)
Tetanus	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Tuberculosis	USA	State-based, reported to CDC	2009-2018	CDC Wonder	(11)
Climate					
All 6 Sunlight/Moisture Variables	Philadelphia County, Pennsylvania	Philadelphia County, Pennsylvania	1981-2010	NOAA	(16)
Pollutants					
All 5 Pollutant Variables	Philadelphia County, Pennsylvania	Philadelphia County, Pennsylvania	2000-2019	EPA	(17)

2.1.3 Assessing Infections' Potential to Affect Bone from Literature

Across the diseases studied, Lyme had the most literature related to bone conditions in various populations. A 1980 study of a man "whose disease appears to be tick‐transmitted" reported joint destruction like that seen in rheumatoid arthritis (21). A Dutch study found "subluxation of the toe joint and periostitis of the bones of the lower limb" (22), while a study in rodents found that Lyme disease "induces trabecular bone loss" (23). In children, orthopedic Lyme can present itself as swelling and pain in the large joints, like in the knees, and potential abnormalities like osteopenia and arthritis, though the latter two symptoms are not unique to pediatric Lyme (24). Lyme disease in dogs can present itself with joint swelling and lameness (25).

2.2 Clinical Data for Osteosarcoma

We obtained dog and human data broken down by biological sex due to the importance of hormonal growth patterns on bone growth. In addition, we separated pediatric human patients from adult human data due to the difference based on age for bone growth patterns that have been implicated in osteosarcoma.

2.2.1 Dog Data

We obtained data from Penn Vet on pet dogs that were diagnosed with osteosarcoma. We obtained this data from a cleaned clinical data repository specifically designed for research purposes (26). Our prior research also found that certain dog breeds were associated with greater risk of osteosarcoma with the top five dog breeds being the Anatolian Shepherd Dog, Greyhound, Irish Wolfhound, Saint Bernard and Bullmastiff (26). In addition, there is known to be an association between osteosarcoma and being of female sex (as a biological variable). Therefore, we separated our data based on biological sex, which resulted in a set of 286 male dogs with osteosarcoma and 292 female dogs with osteosarcoma. We compared the birth month distribution in the male and female dogs with osteosarcoma against the male and female dogs in a larger cohort treated between 2000 and 2017 (26). The majority of dogs both with and without osteosarcoma were spayed/neutered.

2.2.2 Human Data

We obtained data on pediatric osteosarcoma from CHOP. There were 131 children treated at CHOP with osteosarcoma that were from the general Philadelphia area. These osteosarcoma patients were compared against other patients treated at CHOP for other pediatric tumors (i.e., non-osteosarcoma tumors). We also obtained adult human data from females (recorded as biological females at PennMedicine) treated between 2010 and 2017. We compared 402 adult females with osteosarcoma against our entire female-only adult cohort of 771,954 patients.

2.3 Statistical Analysis

2.3.1 Determining Seasonality of Infections

Despite all reported cases being sorted under a specific month, there were consistent increases in cases during December, seemingly contrary to expected counts from November and January data. This may be a result of individual states receiving and reporting an influx of cases at the end of the calendar year and reporting them all to the CDC as occurring during December (11). To normalize the effects of potential over-estimation in December, a hypothetical December value was created by taking the average of January and November cases. The hypothetical December considers that December lies between January and November and assumes the number of cases would do the same. We created overlaid scatterplots of average case counts by month with lines in one chart and an average case count by month with standard deviations for each disease to conduct an initial visual assessment of seasonality, in **Figure 2**. Pennsylvania Lyme only has the one scatterplot, because only data from 2017 was available.

2.3.2 Stratification by Sex

The adult human and dog data were stratified by sex to examine the connection between hormonal regulation and bone growth. In adolescents, osteosarcoma often occurs around the time of a growth spurt, when osteoblasts are rapidly forming to make new bone. Growth spurts are caused by hormonal changes in adolescents, which concurrently spur sexual maturation (27). While there are no definitive causes of osteosarcoma, genetics may play a role (28), and incidence in adolescents, particularly males, and post-menopausal women suggest that a hormonal distinction may affect osteosarcoma occurrence across sexes.

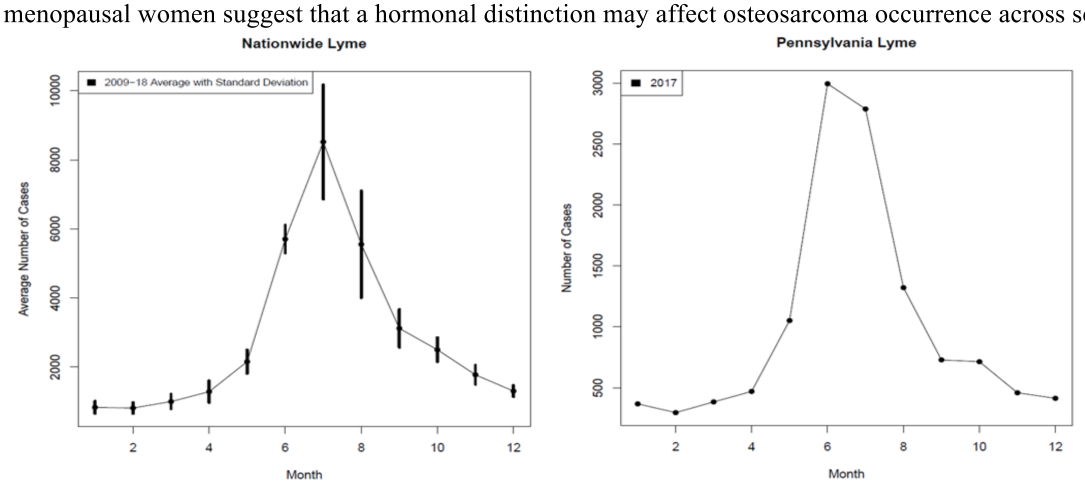

Figure 2. Curves for Nationwide and Pennsylvania Lyme Disease. Nationwide Data was Averaged from 2009-18, while Pennsylvania-specific Data was Exclusive to 2017.

2.3.3 Correlation of Osteosarcoma and Meta-Analysis of Osteosarcoma Incidence - Infections Across Multiple Populations

We then ran t-tests on all eleven disease datasets after their hypothetical December modification against the four osteosarcoma datasets. Observing the average trends of monthly disease cases, we pursued further analysis on eight diseases, including: E. coli, legionellosis, aggregate listeriosis, Lyme, Lyme in Pennsylvania, rubella, salmonellosis, and tuberculosis.

With these eight groups, we used correlation testing to make plots of the disease's average monthly cases against each osteosarcoma dataset, and exported a matrix of the dataset size, correlation value, and p-values.

We used the metacor R package to generate forest plots with the DerSimonian-Laird function. There are plots for national data of each of the eight diseases that appeared to be seasonal, along with Lyme disease monthly trends in Pennsylvania in 2017. The forest plots included meta-analysis of all eight diseases with children, adult females, and sex-stratified dog osteosarcoma datasets.

We again used the metacor R package DerSimonian-Laird function to extract the summary values for each of the eight disease and osteosarcoma correlations and generated a Manhattan plot of the -log(p) * sign(R) to demonstrate the correlation of the disease with osteosarcoma incidence. These processes were repeated with the osteosarcoma correlations with climate and pollutant variables, respectively, and then compiled into a total plot of factors and osteosarcoma correlations in **Figure 3.**

2.3.4 Determining Effects of Vaccination on Seasonally Varying Infections Across Species

Except for rubella, all other seasonal diseases investigated are bacterial infections. In the US, the only common human vaccine administered for any of the diseases was MMR, the combination vaccine preventing measles, mumps, and rubella (29). Among dogs, the only common vaccine is for canine Lyme disease. While dogs cannot get legionellosis, they are susceptible to leptospirosis, another disease frequently linked to bacteria living in outdoor bodies of water, or rodents in urban areas (30). Dogs are also not capable of contracting measles, but can get canine distemper, another illness within the measles family (31). **Table 4** includes detailed vaccine schedules for humans and dogs.

3. Results

3.1 Seasonality of Infectious Diseases in USA

Our method found that eight infectious diseases were seasonal while four were not seasonally varying. Some of these were found to be seasonal in the literature (**Table 2),** however, given the presence of vaccines for both dogs and humans, many previously seasonal diseases are no longer seasonal. Therefore, it is important to investigate the current seasonality of infectious bacterial and viral diseases and conditions. A couple of diseases that we investigated are not reported to infect dogs (**Table 2**). However, we included these diseases (e.g., Legionnaires) due to the presence of related infections that do affect dogs (e.g, Leptospirosis).

Table 2. Seasonality of Common Infectious Diseases

Infectious Disease	Seasonal in Current Data (2009-2018)?	Historically Seasonal (from literature)?	Affects Bone in Literature?	Affects Dogs	Similar Canine Virus/Bacteria	Ref.
E. coli	Yes	Yes	Yes	Yes		(32–34)
Legionellosis	Yes	Yes	Yes	No	Leptospirosis	(35–37)
Listeriosis	Yes	Yes	Yes	Yes		(38–40)
Lyme	Yes	Yes	Yes	Yes		(21,41,42)
Measles	No	Yes	Yes	No	Canine distemper	(31,43,44)
Mumps	No	Yes	No	Yes		(45,46)
Pertussis	No	No	No	No	*Bordetella bronchiseptica*	(47,48)
Rubella	Yes	Yes	Yes	No		(49,50)
Salmonellosis	Yes	Yes	Yes	Yes		(51–53)
Tetanus	No	Yes	Yes	Yes		(54–56)
Tuberculosis	Yes	Yes	Yes	Yes		(57–59)

3.2 Seasonal Infection, Climate and Pollutant Data

Importantly, many infectious diseases could be correlated to birth month dependent diseases not because the infection is related to the birth month dependent disease, but rather because of shared climate factors that may be underlying the relationship. To help address this issue, we also investigate the effects of climate and pollutant factors on the osteosarcoma - birth month relationship, in a manner similar to prior work (3). We next gathered government data on six climate variables and five pollutants in Philadelphia county in **Table 1**. These additional factors are important to consider because of the established disease seasonality. The variation in months and seasons is a result of various climate and pollutant interactions which affect breeding conditions for bacteria or virus carriers like humidity.

Table 3. Results for Correlation with Osteosarcoma Birth Seasonality

Factor	R	Nominal P-value	Significant After Bonferroni Correction?
Seasonal Infection			
E. coli	0.189	0.004	No
Legionellosis	0.156	0.054	No
Listeriosis	0.069	0.191	No
Lyme	0.209	4.31 X 10^{-4}	Yes
PA Lyme	0.418	2.80 X 10^{-23}	Yes
Rubella	0.005	0.481	No
Salmonellosis	0.109	0.103	No
Tuberculosis	0.291	0.011	No
Seasonal Climate Factor			
Mean Sunshine	0.324	0.001	Yes
High Temp	0.241	0.002	Yes
Low Temp	0.233	8.96 X 10^{-4}	Yes
Relative Humidity	0.108	0.277	No
Precipitation Days	-0.005	0.473	No
Precipitation Inches	-0.008	0.441	No
Seasonal Pollutant Factor			
SO2	-0.121	0.128	No
CO	-0.226	0.041	No
NO2	-0.247	0.002	Yes
PM 2.5	0.107	0.108	No
Ozone	0.281	0.008	No

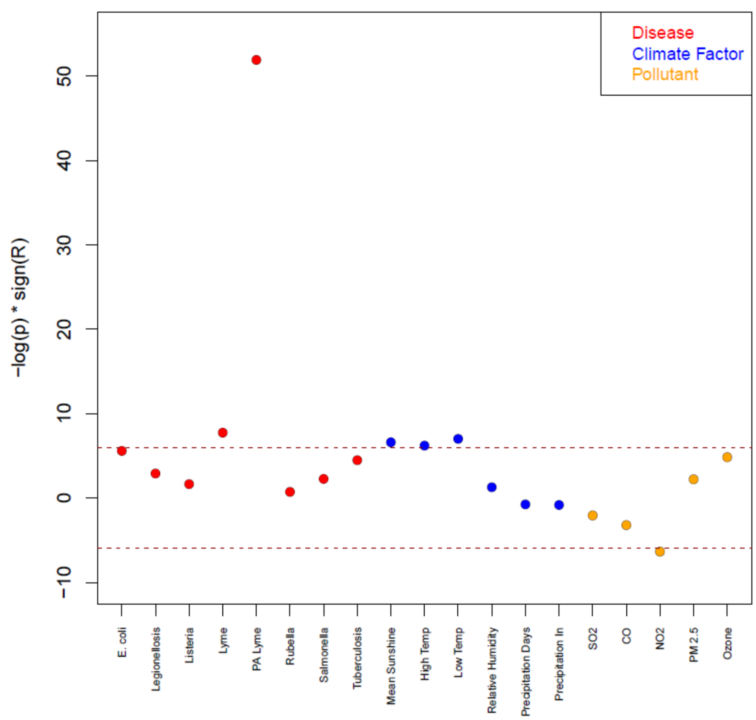

Figure 3. Overall Modified Manhattan Plot Showing All Seasonal Factors Relationship with Osteosarcoma Birth Season. The y-axis shows the log(p-value) * sign of the correlation. The positive correlations are on the top part and the negative correlations on the bottom of the figure. Red dashed line indicates the Bonferroni cutoff.

3.3 Meta-Analysis of Seasonal Factors and Osteosarcoma Risk Across Populations

Nineteen variables spanning seasonal disease, climate factors, and pollutants were assessed for correlation with osteosarcoma (results in **Table 3** and **Figure 3**). We found six of the nineteen variables were significantly associated with Osteosarcoma after adjusting for multiple testing using Bonferroni correction (**Table 3**). These six significant associations are strong candidates for understanding the relationship between Osteosarcoma and birth seasonality. These include national Lyme disease, Pennsylvania Lyme disease, Mean Sunshine hours, high and low temperature and NO2 exposure (shaded in grey in **Table 3**). Pennsylvania Lyme disease had the strongest positive correlation and a breakdown of the association in each of the four datasets is shown in **Figure 4**.

NO2 exposure was the only exposure that was anti-correlated with Osteosarcoma and was also the only pollutant that was significant.

3.4 Assessing the Effect of Vaccination on Infection - Osteosarcoma Risk Signal

Of the seasonal diseases, only human rubella and canine Lyme disease have widespread vaccine usage in the United States. An individual rubella vaccine was developed by 1969, and was soon combined into the Measles, Mumps, and Rubella (MMR) vaccine in 1971 (60). As of the 2017-2018 school year, school-aged children in Pennsylvania are required to have two doses of the MMR vaccine, barring medical or religious exemptions (61). In general, people aged 18 or older and born after 1956 needs one dose the MMR vaccine if they have not already had rubella (62). However, the bimodal age distribution of osteosarcoma suggests that an at-risk population aged 60 and older, whose age group comprises 10% of osteosarcoma cases, may not have had the MMR vaccine in any dosage. The dataset of children with osteosarcoma are far more likely to have been vaccinated using two doses against rubella for school within their lifetimes, whereas the adult females may have been born before the vaccine was available and/or enforced. However, it is also important to note that within the last decade, there were fewer than ten reported rubella cases in the United States each year.

4. Discussion

4.1 Links Between NO2, Estradiol, and Cancer

Nitrogen dioxide (NO2) was the only finding that was significantly anti-correlated with Osteosarcoma (**Figure 3).** Importantly, nitric oxide synthase induced by 2-methoxyestradiol has led to cytotoxicity and apoptosis followed the generation of intracellular NO2 (63). NO2 is known to be toxic, and capable of causing cell and DNA damage, triggering cell death (63). Nitro-oxidative stress encourages NO2 to impact DNA (63). While NO2 has been linked to causing cell and DNA damage, it has also been found to kill metastatic osteosarcoma cells (63). As a known toxin, NO2 has the potential to kill healthy cells, encourage their mutation into potentially cancerous cells, and even eventually kill the cancerous cells themselves (63). Estradiol is a form of estrogen, whose levels peak and plateau within a females' reproductive years (64). A study found that 2-methoxyestradiol caused apoptosis in osteosarcoma cells (65). The increase in estradiol around puberty and decrease around menopause might indicate changes in NO2 production, and various cellular consequences, like DNA mutations. When estradiol levels are maintained throughout the reproductive years, osteosarcoma incidence also plateaued.

4.2 Links Between Lyme Disease, Bone, and Cancer

Lyme disease can have lasting effects on bone, including arthritis and reduced density. While only 50% of Lyme patients develop the characteristic rash, orthopedic symptoms may lead to a more conclusive diagnosis (24). Like osteosarcoma, Lyme disease often causes joint swelling and pain and knees are commonly affected for both conditions. Patients infected with Lyme disease (especially chronic infection) increases susceptibility to cellular changes due to a chronically weakened immune system, which leaves them vulnerable to developing illnesses (66). Importantly, our literature review found that two diseases (mumps and pertussis) did not affect bone in the literature (**Table 2**) and therefore, we would not expect these diseases to be involved in explaining the osteosarcoma birth seasonal relationship. Our method revealed that one of the two was not seasonal (pertussis) and therefore not included in our meta-analysis and the other (mumps) was not correlated with osteosarcoma (**Figure 3, Table 3).**

4.3 Our Method Optimized for Hypothesis Generation

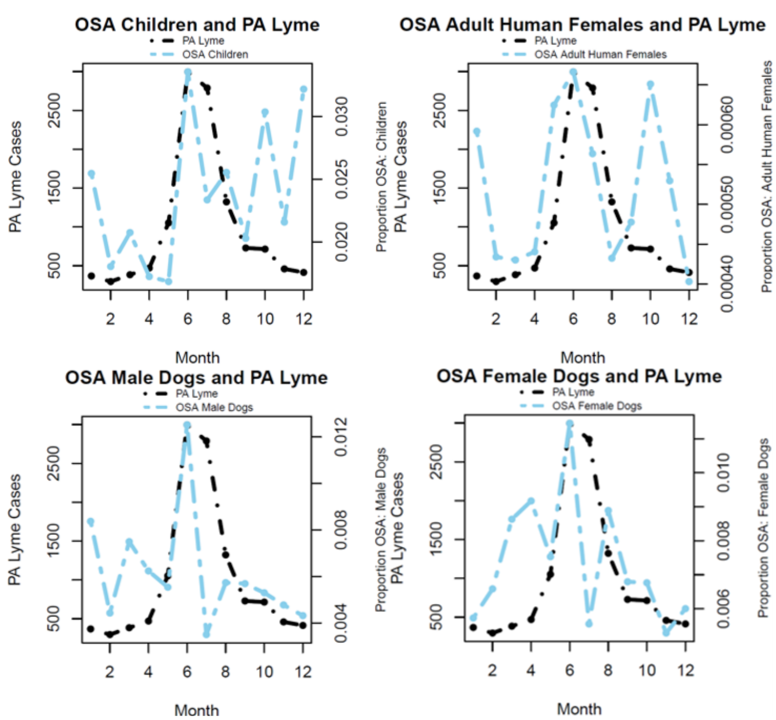

Figure 4. Pennsylvania Lyme and Osteosarcoma (OSA) Dataset Correlations.

The methods described here are useful in hypothesis generation, but not for investigating causality. Investigating relationships between osteosarcoma and various seasonal factors is a necessary step in identifying strong candidates for further analysis, including confounding variables, and interactions of multiple factors.

4.4 Limitations

Limitations of this study include that we do not know who among our population was vaccinated (either dogs or humans). We are also assuming that the national data on seasonal infections collected between 2009-2018 is constant throughout the entire time period. This is most correct for the pediatric patients, but for older women, they may have been exposed to a different seasonal infection pattern.

Generalizability of our method to other regions beyond the contiguous 48 states in the United States of America may be limited by prevalence of infectious diseases at those locations and availability of vaccination at a given location. For example, yellow fever vaccination is more common in states such as Hawaii, but is not broadly administered in the contiguous USA states (67).

Table 4. Vaccine Availability

Disease	**Cause**	**Population / Year Vaccine Became Available**	**Schedule**	**Source**	**Ref.**
E. coli	Bacteria	n/a	n/a	IntechOpen	(68)
Legionellosis	Bacteria	n/a	n/a	CDC	(69)
Listeriosis	Bacteria	n/a	n/a	Frontiers in Cellular and Infection Microbiology	(70)
Lyme	Bacteria	Dog Only / 2009	First dose: 8 weeks, Booster after 1 year, Boosters every 3 years	Merck Animal Health	(71)
PA Lyme	Bacteria	Dog Only / 2009	First dose: 8 weeks, Booster after 1 year, Boosters every 3 years	Merck Animal Health	(71)
Rubella	Virus	Human Only / 1971 (MMR)	First dose: 12-15 months, Second dose: 4-6 years	CDC, CHOP	(29) (60)
Salmonellosis	Bacteria	n/a*	n/a	Human Vaccines & Immunotherapeutics	(72)
Tuberculosis	Bacteria	n/a*	n/a	CDC	(73)

* Both salmonellosis and tuberculosis have vaccines in the US, although neither are commonly administered. The salmonella serotype typhi causes typhoid fever, and this is one serotype covered by a vaccine (72). Foodborne salmonella infection is often manageable, and not vaccinated against. BCG is a tuberculosis vaccine rarely administered in the US because of low infection counts (74). However, it is often given to children in other countries where tuberculosis risk is higher.

5. Conclusion

We developed an algorithm to harness publicly available data on infectious diseases to assess the seasonality of infectious diseases and to incorporate these seasonal patterns into a larger analysis framework that includes pollutant and climate factors that seasonally vary. This enables analysis of multiple factors to assess their role in birth seasonal relationships, one example being osteosarcoma. We found significant correlations with monthly national and Pennsylvania-based Lyme disease counts, Philadelphia mean sunshine hours, and a significant anti-correlation with Philadelphia NO2 air quality values. We adjusted all of our findings' significance using Bonferroni to address multiple testing. These results warrant further investigation to elucidate the underlying biological mechanisms. In addition, our method could be applied to other birth month dependent diseases that could have a seasonal infectious disease as a potential mechanism to explain the findings.

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References

1. Boland MR, Shahn Z, Madigan D, Hripcsak G, Tatonetti NP. Birth month affects lifetime disease risk: a phenome-wide method. J Am Med Informatics Assoc. 2015;22(5):1042–1053.
2. Boland MR, Fieder M, John LH, Rijnbeek PR, Huber S. Female Reproductive Performance and Maternal Birth Month: A Comprehensive Meta-Analysis Exploring Multiple Seasonal Mechanisms. Sci Rep. 2020;10:Article 555.
3. Boland MR, Parhi P, Li L, Miotto R, Carroll R, Iqbal U, et al. Uncovering exposures responsible for birth season – disease effects: a global study. ournal Am Med Informatics Assoc. 2017;25(3):275–288.
4. Boland MR, Kraus MS, Dziuk E, Gelzer AR. Cardiovascular Disease Risk Varies by Birth Month in Canines. Sci Rep. 2018;8:Article 7130.
5. KORSGAARD J, DAHL R. Sensitivity to house dust mite and grass pollen in adults: Influence of the month of birth. Clin Exp Allergy. 1983;
6. Johns Hopkins Medicine. Osteosarcoma [Internet]. Health. 2020. Available from: https://www.hopkinsmedicine.org/health/conditions-and-diseases/sarcoma/osteosarcoma
7. American Cancer Society. Key Statistics for Osteosarcoma [Internet]. 2020. Available from: https://www.cancer.org/cancer/osteosarcoma/about/key-statistics.html
8. American Cancer Society. Osteosarcoma Risk Factors [Internet]. 2018. Available from: https://www.cancer.org/cancer/osteosarcoma/causes-risks-prevention/risk-factors.html

9. Burke A. How Long Are Dogs Pregnant? [Internet]. 2016. Available from: https://www.akc.org/expert-advice/dog-breeding/how-long-are-dogs-pregnant/#:~:text=The normal gestation period in,is often hard to determine.
10. CDC. Summary of Notifiable Infectious Diseases and Conditions — United States. MMWR. 2015.
11. CDC. Nationally Notifiable Infectious Diseases and Conditions, United States: Annual Tables. WONDER. 2018.
12. CDC. NORS Dashboard [Internet]. National Outbreak Reporting System (NORS). 2018. Available from: https://wwwn.cdc.gov/norsdashboard/
13. CDC. Reported Cases of Lyme Disease — United States, 2018 [Internet]. Lyme Disease. 2018 [cited 2020 Jul 7]. Available from: https://www.cdc.gov/lyme/datasurveillance/maps-recent.html
14. Division of Infectious Disease Epidemiology. 2017 Lyme and Other Tickborne Diseases Surveillance Report. 2019.
15. CDC. United States and Puerto Rico Cancer Statistics, 1999-2016 Incidence Request [Internet]. WONDER. 2016 [cited 2020 Jul 6]. Available from: http://wonder.cdc.gov/cancer-v2016.html
16. NOAA. Climate data for Philadelphia [Internet]. 2020 [cited 2020 Jul 22]. Available from: https://en.wikipedia.org/wiki/Philadelphia#Climate
17. EPA. Air Data - Multiyear Tile Plot [Internet]. Outdoor Air Quality Data. 2020. Available from: https://www.epa.gov/outdoor-air-quality-data/air-data-multiyear-tile-plot
18. CDC. Reports of Selected E. coli Outbreak Investigations [Internet]. E. coli Outbreaks. 2020 [cited 2020 Jun 17]. Available from: https://www.cdc.gov/ecoli/outbreaks.html
19. CDC. Listeria Outbreaks [Internet]. Listeria Outbreaks. 2020 [cited 2020 Jun 17]. Available from: https://www.cdc.gov/listeria/outbreaks/index.html
20. CDC. Reports of Selected Salmonella Outbreak Investigations [Internet]. Salmonella Previous Outbreaks. 2020. Available from: https://www.cdc.gov/salmonella/outbreaks.html
21. Steere AC, Brinckerhoff CE, Miller DJ, Drinker H, Harris Jr. ED, Malawista SE. Elevated levels of collagenase and prostaglandin e2 from synovium associated with chronic lyme arthritis. Arthritis Rheumatol. 1980;23(5):591–9.
22. Houtman PM, Tazelaar DJ. Joint and bone involvement in Dutch patients with lyme borreliosis presenting with acrodermatitis chronica atrophicans. Neth J Med. 1999;54(1):5–9.
23. Tang TT, Zhang L, Bansal A, Grynpas M, Moriarty TJ. The Lyme Disease Pathogen Borrelia burgdorferi Infects Murine Bone and Induces Trabecular Bone Loss. Infect Immun. 2017;85(2).
24. Davidson RS. Orthopaedic complications of Lyme disease in children. Biomed Pharmacother. 1989;43(6):405–8.
25. American Veterinary Medical Association. Lyme disease: A pet owner's guide [Internet]. 2020. Available from: https://www.avma.org/resources/pet-owners/petcare/lyme-disease-pet-owners-guide
26. Boland MR, Casal ML, Kraus MS, Gelzer AR. Applied Veterinary Informatics: Development of a Semantic and Domain-Specific Method to Construct a Canine Data Repository. Sci Rep. 2019;Article 18641.
27. Center for Academic Research and Training in Anthropogeny. ADOLESCENT GROWTH SPURT [Internet]. 2020. Available from: https://carta.anthropogeny.org/moca/topics/adolescent-growth-spurt#:~:text=In humans%2C the hormones responsible,the adolescent life history stage.
28. Children's Hospital of Philadelphia. Osteosarcoma (Bone Cancer in Children) [Internet]. 2020. Available from: https://www.chop.edu/conditions-diseases/osteosarcoma
29. CDC. MMR Vaccination [Internet]. 2020. Available from: https://www.cdc.gov/vaccines/vpd/mmr/public/index.html#:~:text=CDC recommends all children get,days after the first dose.
30. American Veterinary Medical Association. Leptospirosis [Internet]. 2020. Available from: https://www.avma.org/resources/pet-owners/petcare/leptospirosis
31. Racine E. American Kennel Club [Internet]. Can Dogs Get Measles? 2019. Available from: https://www.akc.org/expert-advice/health/can-dogs-get-measles/#:~:text=Fortunately%2C the short answer is,the same family as measles.
32. Crémet L, Broquet A, Brulin B, Jacqueline C, Dauvergne S, Brion R, et al. Pathogenic potential of Escherichia coli clinical strains from orthopedic implant infections towards human osteoblastic cells. Pathog Dis. 2015;73(8).
33. Freeman JT, Anderson DJ, Sexton DJ. Seasonal peaks in Escherichia coli infections: possible explanations and implications. Clin Microbiol Infect. 2009;15(10).
34. Clark M. E. Coli Infection In Dogs: Symptoms, Causes, And Treatments [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://dogtime.com/dog-health/53345-e-coli-infection-dogs-symptoms-causes-treatments
35. McClelland MR, Vaszar LT, Kagawa FT. Pneumonia and Osteomyelitis Due to Legionella longbeachae in a Woman with Systemic Lupus Erythematosus. Clin Infect Dis. 2004;38(10).
36. FALCONI TMA, CRUZ MS, NAUMOVA EN. The Shift in Seasonality of Legionellosis in the U.S. Epidemiol Infect. 2018;146(14):1824–33.
37. Wag Walking. Can Dogs Get Legionnaires' Disease? [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://wagwalking.com/wellness/can-dogs-get-legionnaires-disease
38. Charlier C, Leclercq A, Cazenave B, Desplaces N, Travier L, Cantinelli T, et al. Listeria monocytogenes-associated joint and bone infections: a study of 43 consecutive cases. Clin Infect Dis. 2012;54(2):240–8.
39. CDC. Get the Facts about Listeria [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://www.fda.gov/animal-veterinary/animal-health-literacy/get-facts-about-listeria
40. MacGowan AP, Bowker K, McLauchlin J, Bennett PM, Reeves DS. The occurrence and seasonal changes in the isolation of Listeria spp. in shop bought food stuffs, human faeces, sewage and soil from urban sources. Int J Food Microbiol. 1994;21(4):325–34.
41. Moore SM, Eisen RJ, Monaghan A, Mead P. Meteorological Influences on the Seasonality of Lyme Disease in the United States. Am J Trop Med Hyg. 2014;90(3):486–496.
42. Meyers H. Lyme Disease in Dogs: Symptoms, Tests, Treatment, and Prevention [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://www.akc.org/expert-advice/health/lyme-disease-in-dogs/
43. ScienceDaily. Measles virus plays role in Paget's disease of bone, researchers say [Internet]. Science News from research organizations. 2011. Available from: https://www.sciencedaily.com/releases/2011/01/110114155338.htm#:~:text=Measles virus plays role in Paget's disease of bone%2C researchers say,-Date%3A January 16&text=Summary%3A,bone%2C

according to new research.
44. Micaela Elvira Martinez. The calendar of epidemics: Seasonal cycles of infectious diseases. PLOS Pathog. 2018;14(11).
45. Heuer V. Mumps in Dogs [Internet]. 2009 [cited 2020 Dec 8]. Available from: https://www.petmd.com/dog/conditions/infectious-parasitic/c_dg_mumps
46. Shah AP, Smolensky MH, Burau KD, Cech IM, Lai D. Seasonality of primarily childhood and young adult infectious diseases in the United States. Chronobiol Int. 2006;23(5):1065–82.
47. CDC. Pertussis [Internet]. Immunology and Vaccine-Preventable Diseases – Pink Book. 2015 [cited 2020 Dec 8]. p. Pertussis. Available from: https://www.cdc.gov/vaccines/pubs/pinkbook/downloads/pert.pdf
48. Wag Walking. Can Dogs Get Pertussis? [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://wagwalking.com/wellness/can-dogs-get-pertussis
49. Reed GB. Rubella bone lesions. J Pediatr. 1969;74(2):208–13.
50. World Health Organization. Rubella [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://www.who.int/biologicals/areas/vaccines/rubella/en/
51. S. McAnearney, McCall D. Salmonella Osteomyelitis. Ulster Med J. 2015;84(3):171–2.
52. US FDA. Get the Facts about Salmonella [Internet]. 2020 [cited 2020 Dec 8]. Available from: https://www.fda.gov/animal-veterinary/animal-health-literacy/get-facts-about-salmonella#:~:text=Salmonellosis is uncommon in dogs,other pets in the household.
53. Magossi G, Bai J, Cernicchiaro N, Jones C, Porter E, Trinetta V. Seasonal Presence of Salmonella spp., Salmonella Typhimurium and Its Monophasic Variant Serotype I 4,[5],12:i:-, in Selected United States Swine Feed Mills. Foodborne Pathog Dis. 2019;16(4).
54. Cedars-Sinai. Tetanus [Internet]. 2020. Available from: https://www.cedars-sinai.org/health-library/diseases-and-conditions/t/tetanus.html
55. Barnette C. Tetanus in Dogs [Internet]. VCA Hospitals. 2020 [cited 2020 Dec 8]. Available from: https://vcahospitals.com/know-your-pet/tetanus-in-dogs#:~:text=Although tetanus can be seen,toxin than humans and horses.
56. Clark W. Health J, Zusman J, Sherman IL. Tetanus in the United States, 1950-1960. Am J Public Health. 1964;
57. Carlos Pigrau-Serrallach, Rodríguez-Pardo D. Bone and joint tuberculosis. Eur Spine J. 2013;22(4):556–66.
58. Ampel NM. Tuberculosis Is Seasonal in the U.S. New Engl J Med J Watch. 2012;
59. Thoen CO. Tuberculosis in Dogs [Internet]. 2018 [cited 2020 Dec 8]. Available from: https://www.merckvetmanual.com/dog-owners/disorders-affecting-multiple-body-systems-of-dogs/tuberculosis-in-dogs
60. Children's Hospital of Philadelphia. Vaccine History: Developments by Year [Internet]. Vaccine Education Center. 2019 [cited 2020 Jul 13]. Available from: https://www.chop.edu/centers-programs/vaccine-education-center/vaccine-history/developments-by-year#:~:text=In 1963 the measles vaccine,the MMR vaccine in 1971.
61. PA DOH. School Vaccination Requirement Fact Sheet [Internet]. School Immunizations. 2017 [cited 2020 May 8]. Available from: https://www.health.pa.gov/topics/Documents/School Health/SIR8.pdf
62. U.S. Department of Health & Human Services. Rubella (German Measles) [Internet]. vaccine.gov. 2020 [cited 2020 May 8]. Available from: https://www.vaccines.gov/diseases/rubella#:~:text=for your child.-,Adults,doses of the rubella vaccine.
63. Gorska-Ponikowska M, Ploska A, Jacewicz D, Szkatula M, Barone G, Bosco G Lo, et al. Modification of DNA structure by reactive nitrogen species as a result of 2-methoxyestradiol–induced neuronal nitric oxide synthase uncoupling in metastatic osteosarcoma cells. Redox Biol. 2020;32:101522.
64. Dimitrakakis C, Bondy C. Androgens and the breast. Breast cancer Res. 2009;11(5).
65. Maran A, Zhang M, Kennedy A., Sibonga J., Rickard D., Spelsberg T., et al. 2-methoxyestradiol induces interferon gene expression and apoptosis in osteosarcoma cells. Bone. 2002;30(2):393–8.
66. Envita Medical Center. How Lyme Disease and Chronic Infections Can Lead to Cancer [Internet]. 2020. Available from: https://www.envita.com/lyme-disease/lyme-disease-and-chronic-infections-can-lead-to-cancer
67. Hawai'i Pacific Health Straub Medical Center. Travel Medicine [Internet]. 2020 [cited 2020 Dec 18]. Available from: https://www.hawaiipacifichealth.org/straub/services/travel-medicine/
68. Larzábal M, Cataldi AA, Vilte DA. Human and Veterinary Vaccines against Pathogenic Escherichia coli. In: The Universe of Escherichia coli. IntechOpen; 2019. p. 1–20.
69. Cooley LA. Legionellosis (Legionnaires' Disease & Pontiac Fever) [Internet]. Travelers' Health. 2019 [cited 2020 May 8]. Available from: https://wwwnc.cdc.gov/travel/yellowbook/2020/travel-related-infectious-diseases/legionellosis-legionnaires-disease-and-pontiac-fever
70. Calderón-González R, Frande-Cabanes E, Bronchalo-Vicente L, Lecea-Cuello MJ, Pareja E, Bosch-Martínez A, et al. Cellular vaccines in listeriosis: role of the Listeria antigen GAPDH. Front Cell Infect Microbiol. 2014;4.
71. Merck Animal Health. Nobivac Lyme Vaccine [Internet]. Nobivac. 2020. Available from: https://www.merck-animal-health-usa.com/nobivac/nobivac-canine-vaccine-lyme
72. MacLennan CA, Martin LB, Micoli F. Vaccines against invasive Salmonella disease: Current status and future directions. Hum Vaccin Immunother. 2014;10(6):1478–1493.
73. CDC. Tuberculosis (TB) Vaccination [Internet]. Vaccines and Preventable Diseases. 2009. Available from: https://www.cdc.gov/vaccines/vpd/tb/index.html
74. CDC. Questions and Answers About TB [Internet]. Tuberculosis (TB). 2012. Available from: https://www.cdc.gov/tb/publications/faqs/qa_latenttbinf.htm#Latent4