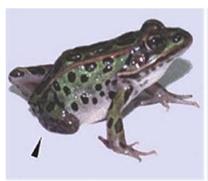
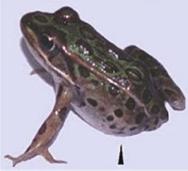
## **Malformed Frogs in Vermont**

In the summer of 1996 occurrences of abnormal frogs were reported by the public to the Vermont Department of Environmental Conservation from 12 sites. The twelve sites reported to have abnormal frogs were all located within the Lake Champlain Basin and spanned over 120 miles of Lake Champlain from the northenmost sites close to the Canadian border to the southernmost site near the Poultney River. VT DEC staff surveyed four of the reported locations in 1996 and found abnormal frogs at all four of



the sites. All of the abnormalities encountered were with young of the year northern leopard frogs (*Rana pipiens*). A total of 290 northern leopard frogs were collected at the four sites, 13.1% of the frogs had observable external abnormalities. Abnormality rates ranged from 5 - 23%.





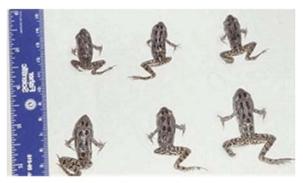
In 1997 survey work targeting the northern leopard frog was conducted by VT DEC, USEPA, USFWS, USGS-BRD and Middlebury College. This set the frame work for additional surveys conducted through 2000. During the 1997 survey over 2500 northern leopard frogs were examined from 19 sites representing 13 towns in 5

counties of Vermont. Roughly 8% of the frogs examined had some category of abnormality. Laboratory experiments conducted on normal and abnormal frog samples from the 1997 sites indicated that the abnormalities were not caused by viruses, bacteria, or parasites.

Abnormal frogs were found at 17 of the 19 sites surveyed. Categories of abnormalities were primarily missing and partial hind limbs and shortened and missing digits.

Systematic field surveys have now been conducted by VT DEC for five consecutive summers (1997- 2002) at a total of 20 sites within the Lake Champlain Basin. Over 11,000 recently-metamorphosed northern leopard frogs have been recorded. External abnormalities were detected in 628/11,186 Rana pipiens examined yielding an abnormality rate of 5.6%.

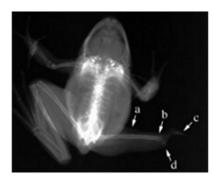




Categories of abnormalities were primarily missing and incomplete hind limbs and shortened and missing digits. Based on five years of seasonal sampling records, observed abnormality rates can vary considerably both seasonally and annually at sites. The data suggest that several sampling events at a site seasonally and annually may be needed before adequately characterizing a site for presence or absence of abnormal frogs.

The USGS-BRD

National Wildlife Health Center (NWHC) has evaluated *R pipiens* specimens collected from sites in the Lake Champlain Basin in 1997 (pdf, 957 KB), 1998, and 1999. Radiographic examinations performed by NWHC support current theories that many of the abnormalities are caused by primary errors in development (malformations) as opposed to deformations which occur later and are mechanical. "Of the Vermont frogs missing entire limbs, x-rays revealed that 73% were also missing bones in the hip, providing evidence that a predator had not removed the limbs, but that developmental errors were to blame." (USGS



News Release, "X-Ray Studies Shed Light on Frog Deformities", March 29, 2000).



### Malformed, Deformed, and Abnormal Frogs

The term "abnormality" is often used instead of "deformity" or "malformation" because it covers all aspects of frog disfigurement. It is very difficult to determine if a frog is "deformed" or "malformed" without conducting detailed examinations using x-rays or other laboratory tests. The term "malformed" refers to an "early developmental error," and the

term "deformed" refers to an abnormality that occurs later in development, such as trauma.

#### **Natural Deformity Rate**

Nationally the prominence of "deformities" has only appeared during the last 5 years. While there are historical accounts of malformed frogs in the literature the number of locations and proportions appear to be unprecedented.

A major discussion point among researchers has been whether there is a natural rate of malformations that can be expected among wild populations of frogs. Most researchers believe a 1 percent malformity rate might be considered normal.

# **Amphibians are Environmental Sponges**

Amphibians are very vulnerable to the environment around them. Their eggs and embryo's lack protective shells, are laid in water, and are sensitive to chemical, UVB and other environmental assaults during early cell division. The permeable skin of amphibians is designed to efficiently exchange gases and water; however they must remain in frequent contact with water or wet soil to prevent drying out. Thus they are vulnerable to any environmental factors



that affect the skin. Amphibians are environmental sponges, soaking up whatever chemicals and toxins are present in the soil and water.



# Citizens Reports of Abnormal Frogs in Vermont (1996-2000)

Citizens have reported abnormal frogs from over 100 towns representing all 14 counties in Vermont. Most of these reports have not been verified. Seven species have been reported with abnormalities including: Northern leopard frog, Green frog, Wood frog, Mink frog, Bull frog, Pickerel frog and American Toad. Most abnormal frog reports from citizens are from a very small sample size. The reports are valuable, but may not be accurately representing the incidence of abnormal frogs above the normal background level of 1%. Surveys that collect and examine dozens of frogs from one site

will provide more useful data. Click here for help identifying amphibians.

# Non-Profit Organizations Survey Hundreds of Sites in Vermont

Thousands of frogs have been collected and examined through the efforts of non-profit organizations. The National Wildlife Federation NWF) surveyed dozens of sites during the 1998 and 1999 season. Click <a href="here">here</a> to go to NWF's website. Vermont Public Interest Research Group (VPIRG) has organized Vermont citizens of all ages to help survey frogs in Vermont for the last several years.



#### **Research Conducted in 2001**

VT DEC conducted a US EPA funded study in the spring of 2001, "Investigations into the Causes of Amphibian Malformations in the Lake Champlain Basin of New England". The study included chemical characterization of water and sediment from sites, laboratory limb development assays using northern leopard frogs and field surveys characterizing northern leopard frogs from tadpole to metamorph. The final report can be downloaded by clicking on the links below:

- Amphibian Malformations in the Lake Champlain Basin (pdf, 11.7 MB)
- Executive Summary Amphibian Malformations in the Lake Champlain Basin (pdf, 183 KB)

## **Reporting Abnormal Frogs and other Amphibians**

To report abnormal frogs and other amphibians that you have found in Vermont contact Rick Levey. Please include your name, phone number, location, description of species and abnormalities observed.

# What are Some of the Possible Causes of Malformed Frogs?

No one knows for sure yet what's causing the frog abnormalities. It is likely that there is no "one" cause that will be responsible for all of the abnormalities observed. It is more likely that several factors may be involved. Frog deformities have many causes, some natural and some quite unnatural. There are three leading theories: ultraviolet radiation, parasites and "something in the water."



# Theory #1: Ultraviolet Radiation

One theory is that ultraviolet (UV) radiation from the sun plays a role, either through direct toxicity or by breaking down chemicals in the environment into other, unknown compounds. Ambient UV-B radiation will kill some amphibian eggs under field and lab conditions. Ozone depletion of the atmosphere leads to more UV-B radiation striking the earth. In experiments with UV, the EPA lab in Duluth has produced deformities in lab frogs that resemble some of those seen in the field. And a researcher in Oregon is accumulating evidence that UV radiation deforms Pacific tree frogs there.

#### **Additional Information:**

- USGS: "Duluth Researchers Cite Link Between Ultraviolet Radiation, Deformed Frogs"
- USGS: "Amphibian Embryo Mortality Believed Caused by UV-B Radiation Exposure"

#### **Theory #2: Parasites**

At least 41 species of trematodes (flatworm) have amphibian larvae as intermediate host (i.e. borrow into their tissues to form metacercarial cysts). A California researcher made news in 1999 with a study that established the role of parasitic flatworms (Ribeiroia) in malformations of tree frogs in California. Results from these studies reveal that 30-50% of the abnormalities observed are extra limbs. In Vermont, after examining over 7000 northern leopard frogs, only one frog had an extra limb. Researchers that have examined normal and abnormal leopard frogs from Vermont have not found a consistent association between abnormal frogs and trematode parasite cysts. Minnesota researchers have studied hundreds of frogs, and have found that both normal and abnormal frogs can be heavily, lightly or not at all infected with trematodes.

#### Additional Information:

- ScientificAmerican.com: "Parasites or Pollution?"
- USGS: "Parasites and Malformed Frogs"

## Theory #3: Something in the Water - Contaminants

Numerous laboratory studies of contaminants amply demonstrate the susceptibility of amphibians to compounds such as metals, petroleum products, herbicides and pesticides. Researchers from the National Institute of Environmental Health (NIEHS) found that water from study sites in Minnesota and Vermont causes malformed frogs in the laboratory, and that water from "normal" sites (no malformed frogs) does not. Some of the chemicals in the water have been identified and



tested for their ability to cause frog malformations. Moreover, filtering the water in these experiments through activated carbon removes the effect, indicating probable organic compounds. Also, adding thyroid hormone to the water moderates some of the effects. Thyroid hormone is essential for normal frog development, so this finding could point to chemicals in the water acting as endocrine disrupters. Several papers have been published on the results of this ongoing work.

#### **Additional Information:**

- ScienceNews.org: "Thyroid Linked to Some Frog Defects"
- <u>USGS: "Pesticide Linked to Deformed Frogs, Studies Indicate"</u>

## **Link Between Pesticides and Abnormal Frogs?**

Dr. Martin Ouellet, a researcher with McGill University in Montreal, is convinced that a major factor in frog malformities is pesticides. His findings are based on years of research involving the evaluation of over 30,000 frogs along a 150-mile stretch of the St. Lawrence River Valley. His studies show that the incidence of limb malformities averages 20% in agricultural regions subject to a variety of pesticides and other chemicals and only 1.5% in non-agricultural areas. On one farm in the St. Lawrence Valley of Quebec, every frog he found was malformed. Dr. Ouellet's autopsy reports reveled three conditions: frogs that resemble males externally but are actually females; frogs poisoned by clogged and yellowed livers; and, frogs with altered DNA.

#### Additional Information:

- USGS: "Another Pesticide Surprise"
- USGS: "Pesticides: On the Farm. In the Fields."

#### **Links of Interest**

- North American Reporting Center for Amphibian Malformations
- The Herptox Page- The Effects of Environmental Contaminants on Reptiles and Amphibians
- Minnesota Pollution Control Agency Deformed Frogs
- North American Amphibian Monitoring Program
- A Thousand Friends of Frogs

# **Suggested Reading**

- Investigations of Abnormal Northern Leopard Frogs in Vermont Abstract from 43rd Conference of the International Association for Great Lakes Research
- Fort, D.J.,T.L. Propst, E.L.Stover, J.C. Helgen, R.B. Levey, K. Gallager, J.G. Burkhart. 1999a. Effects of pond water, sediment and sediment extracts from Minnesota and Vermont, USA on early development and metamorphosis of Xenopus. Environ Toxicol Chem 18 (10):2305-2315.
- Meteyer, C.U., R.A. Cole, K.A. Converse, D.E. Docherty, M. Wolcott, J.C. Helgen, R.Levey, L. Eaton-Poole, and J.G. Burkhart. 2000. Defining anuran malformations in the context of a developmental problem. Journal Iowa Acad. Sci. 107(3)72-78.
- Meteyer C.U., K. Loeffler, J. Fallon, K.A. Converse, E. Green, J.C. Helgen, S. Kersten, R. Levey. L. Eaton-Poole, J.G. Burkhart. 2000. Hind limb malformations in free-living Northern Leopard frogs (Rana pipiens) from Maine, Minnesota, and Vermont suggest multiple etiologies. Teratology 62: 151-171.