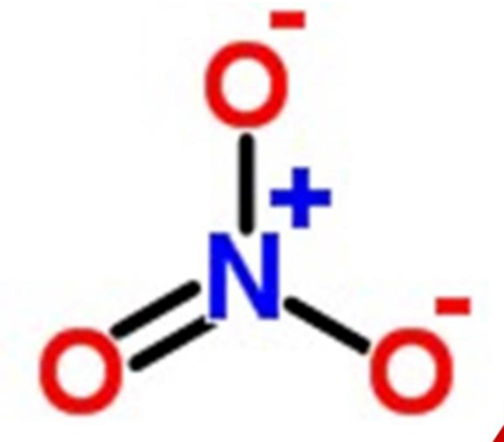
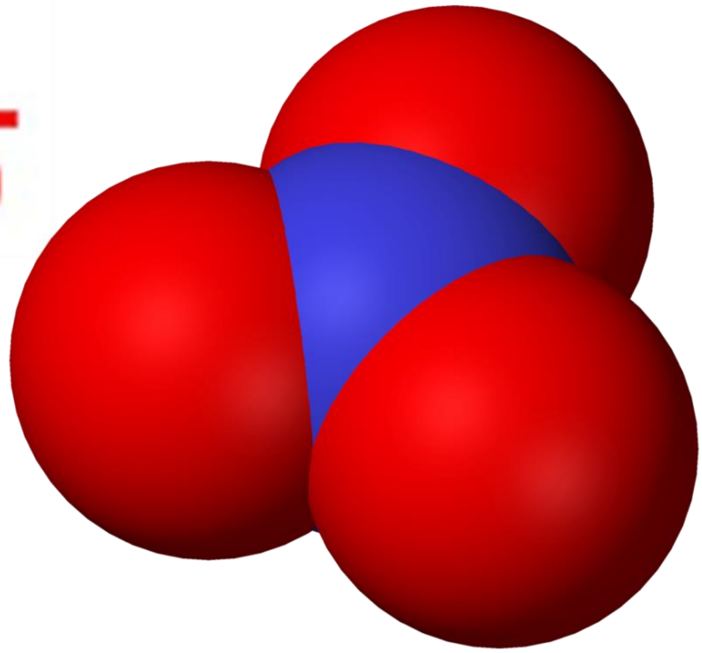
Think of this as crime scene investigation: CSI. The scene of the crime is the various streams that make up the Ventura watershed. Unlike most criminal cases, however, we already know "who did it": Nitrogen. What we don't know is how it got to the scene of the crime, and from whence it came. Oh yes, the recent TMDL lined up the usual suspects and tried to point the finger, but these were mostly guesses, guesses based on who deposited what amounts of nitrogen on the land. There was no examination of whether or not this nitrogen ended up in the river. What I'm about is a closer look at the existing evidence, and the result of some recent efforts to find the proverbial "smoking gun."





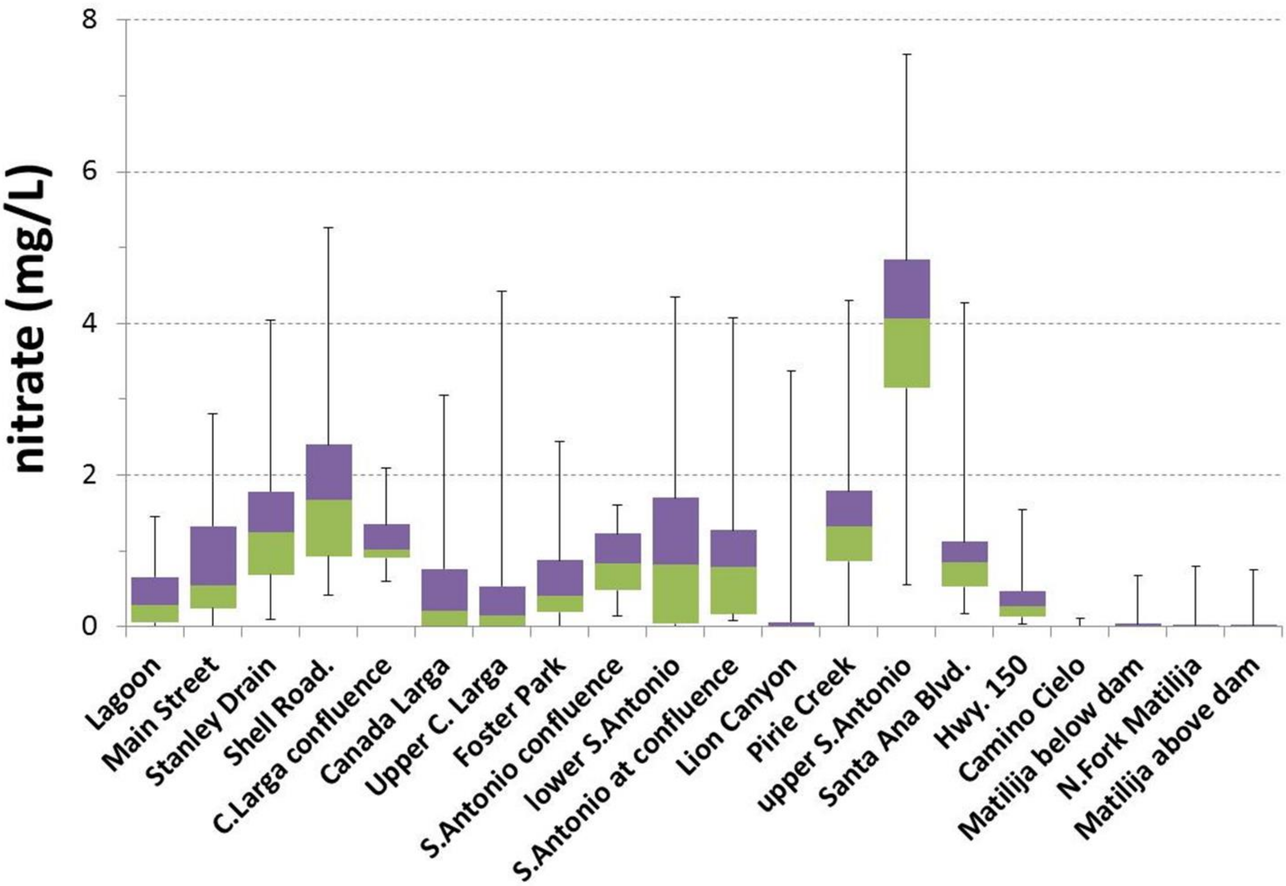
Nitrogen exists in many forms and has a complicated natural cycle, but in terms of water pollution the only form we need concern ourselves with is *nitrate*. If we are talking pollution, we're talking nitrate. In waters with excessive nitrogen, more than 90% of that excess will be found in the form of nitrate. So here's our criminal: nitrate. A molecule with one nitrogen and 3 oxygen atoms. It carries an electrical charge of minus one. Which causes it to be highly soluble and easily transported by water – which is precisely why it can become a problem.

"Poison is in everything, and no thing is without poison. The dosage makes it either a poison or a remedy."

Paracelsus

There is nothing fundamentally dangerous about nitrogen or nitrate. Nor about any of the other possible contaminants I'll be referring to. In fact, all are necessary for life – nitrogen is essential for building proteins, phosphorus for energy utilization and storage, and chloride for fluid transport in biological cells. We'd be dead without 'em. As Paracelsus said, it's excess that creates the danger. With nutrients (nitrogen and phosphorus), excessive amounts in aquatic systems lead to exuberant over-growth of algae and plants, and overgrowth leads to the oxygen depletion that can turn streams septic, creating a public nuisance unfit for any kind of oxygen-breathing life.

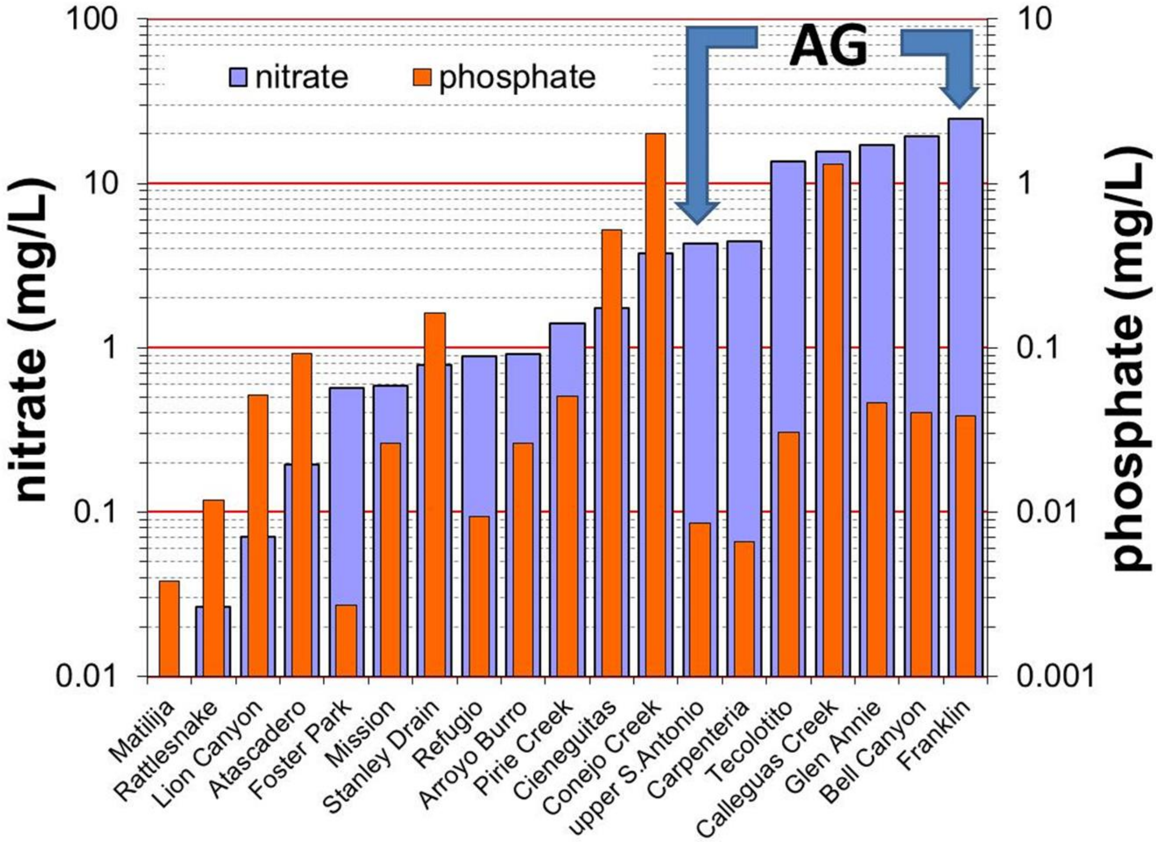
One of the secrets of humanity's success is that we've tripled the supply of fixed nitrogen – the only nitrogen that can be utilized for plant growth – available on the planet. Without which, we would never be able to feed the seven billions of us now alive. But nitrate pollution in natural systems is the reverse side of this success.



This is a box plot of nitrate in the Ventura watershed. Each box represents the range of the middle half of monthly measurements made by Santa Barbara Channelkeeper (SBCK) since 2001; the line in the middle of the box shows the median – half the measurements were above this value, half below. The end of each bar extending above and below the box shows the highest and lowest monthly concentrations measured.

The location with the highest nitrate concentration, i.e. the most significant nitrogen pollution, is – and always has been – upper San Antonio Creek. In second place is Shell Road, not far below the Ojai waste water treatment plant (WWTP). But we know exactly where the Shell Road nitrogen is coming from, so for the remainder of my talk I'll be concentrating on the source of the problem on San Antonio Creek. More specifically, I'll be looking mostly at the differences between upper San Antonio and Pirie creeks.

As a first step, knowing the magnitude of the nitrate concentration can, by itself, provide a clue as to the source. Different land uses typically generate nitrate concentrations in runoff and streamflow characteristic of that land use. (The background photo was taken upstream of the Santa Ana Bridge in August, 2005.)

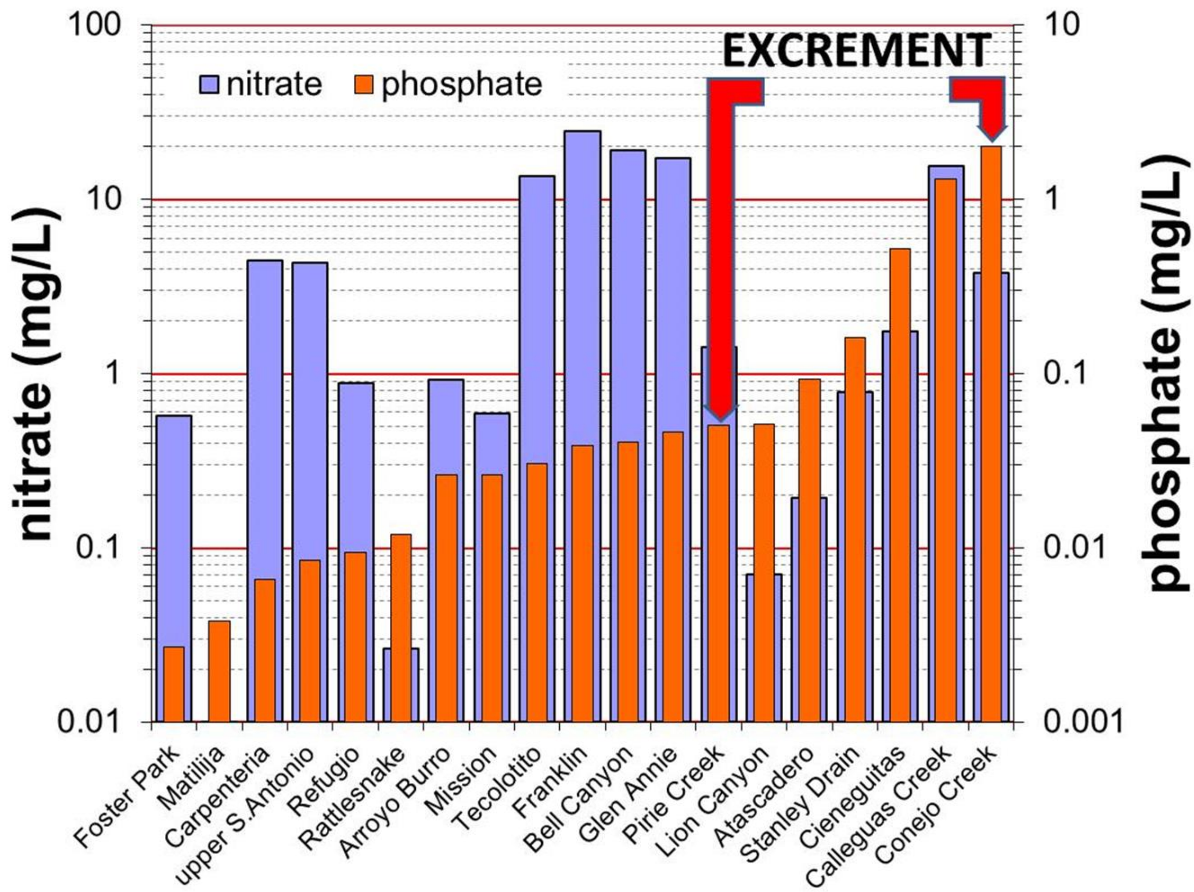


Mean nitrate and phosphate concentrations measured in various coastal streams in the area between Santa Barbara and Ventura are shown in the graph; they are arranged by lowest to highest nitrate values. The scale is logarithmic, so that widely varying results can be shown on a single graph. A logarithmic scale, however, makes large differences look small; the sampling location with the highest nitrate concentrations (Franklin Creek in Carpenteria) has a mean concentration *3,000-times* greater than the location with the lowest (Matilija Creek). Streams with the lowest nitrate (<0.1 mg-N/L) are relatively pristine, those with <1.0 mg-N/L tend to flow from urban watersheds, while those with concentrations above 3-4 mg/L are predominately agricultural: the more intensive the agriculture, the greater the nitrate. Naturally, there is some overlap. Streams monitored directly downstream of WWTPs (e.g. Conejo) or with mixed land uses (Cieneguitas, urban and horses) or with severe septic tank/leach field failure problems can fall into the urban-ag gap. Three-fold higher nitrate concentrations on upper S. Antonio compared with Pirie argue for different origins of their nitrate problems.

Note: The TMDL calls for an eventual maximum nitrate concentration of 1 mg/L (compared with a present dry-season mean of >4 at upper S. Antonio). This is by no means a stringent requirement: the CA coastal stream standard recommended TN <0.5 mg/L for a good quality water; the similar EPA recommendation was TN <0.52, but with nitrate <0.16 (or <0.38, depending on the exact zone). The TMDLs TN limit is 1.15 mg-N/L. The Public Health drinking water limit remains 10 mg-N/L.

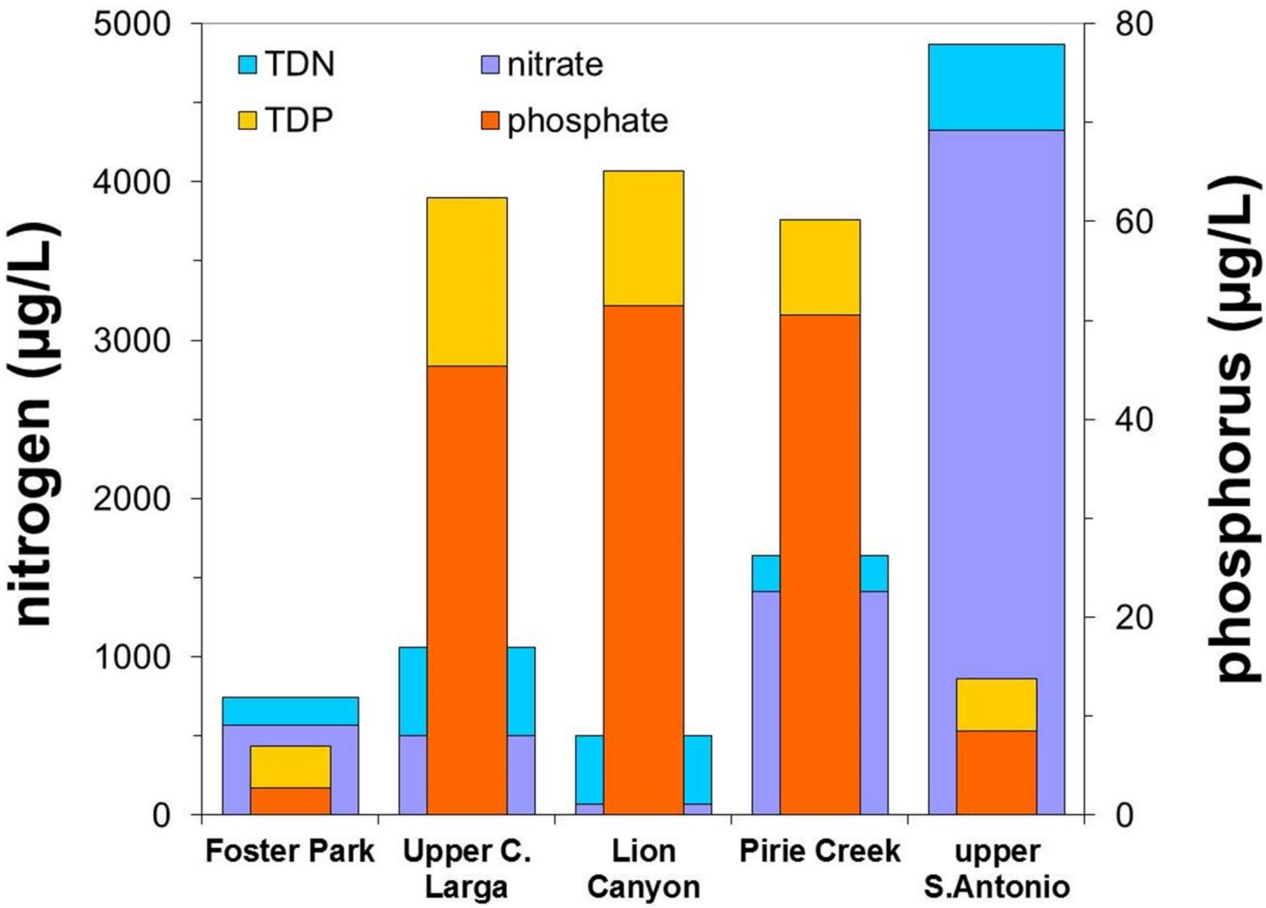
We can examine phosphate concentrations in the same way we looked at nitrate: different land uses produce characteristic phosphate concentrations in streamflow. In our area the agricultural fertilizers used tend to be high in nitrogen and low in phosphorus. In contrast, fertilizers utilized in an urban or suburban context for gardening and landscaping (and this often includes golf courses) are generally of the “let’s make sure all bases are covered” kind, much higher in phosphorus. When fertilizer is a minor incidental expense cost is rarely a concern; cost always is for agriculturists who tend not to buy what they don’t really need.

Manure – from animals and, yes, humans (hopefully, mostly in the form of treated sewage from WWTP effluent, leaking sewers and on-site waste disposal systems, e.g. septic tanks/leach fields) generally causes the highest phosphorus concentrations in streamflow. Manure is about 3-times higher in phosphorus then it needs to be for most plant growth, and the disproportion grows even higher as manure ages and highly volatile ammonia escapes to the atmosphere.



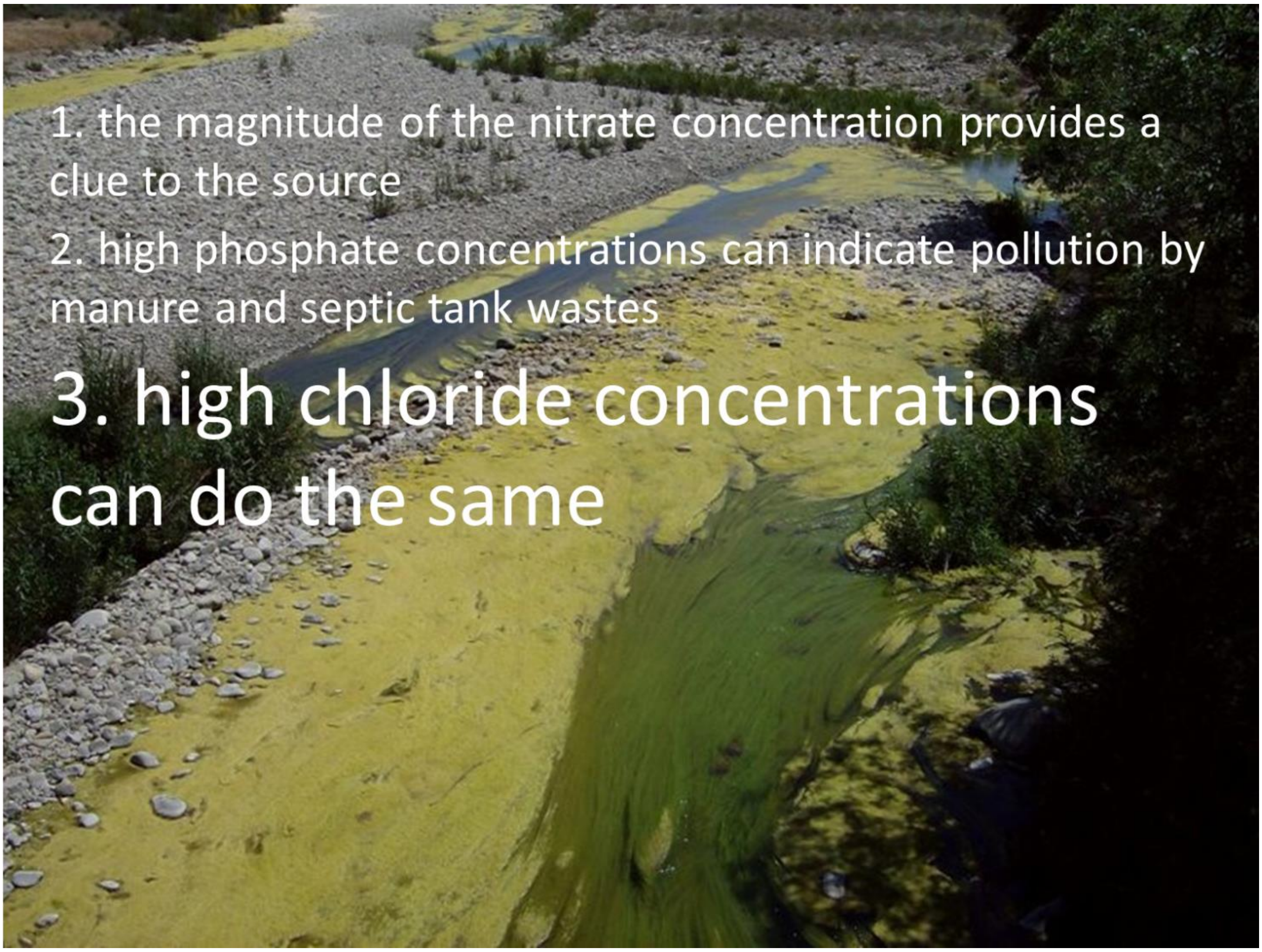
This the same graph shown earlier, except that sampling locations are now arranged, from lowest to highest, by mean phosphate concentration. The low and middle ranges are quite mixed: near-pristine, urban and agricultural land uses are all jumbled together (the background nitrate values indicate which are probably which). But the high end almost invariably represents contamination by manure or treated sewage effluent: Conejo Creek and Stanley Drain are locations downstream of WWTPs, Lion, Atascadero and Cieneguitas all have appreciable horse or cattle use).

The relative proportion of nitrate to phosphate can be an even better guide. The vertical scales are arranged in a 10 to 1, nitrate to phosphate, ratio (by weight). Only Calleguas Creek exhibits a ratio near this value; there is a great unevenness among all the others. The predominately agricultural streams have ratios averaging around *500 to 1*; those with heavy animal usage, or an upstream source of WWTP effluent, a ratio around *3*. Upper S. Antonio Creek clearly fits in the agricultural catchment class with a ratio >500. The Pirie nitrate to phosphate ratio (by weight) is 28 (similar to urban and mixed use catchments).

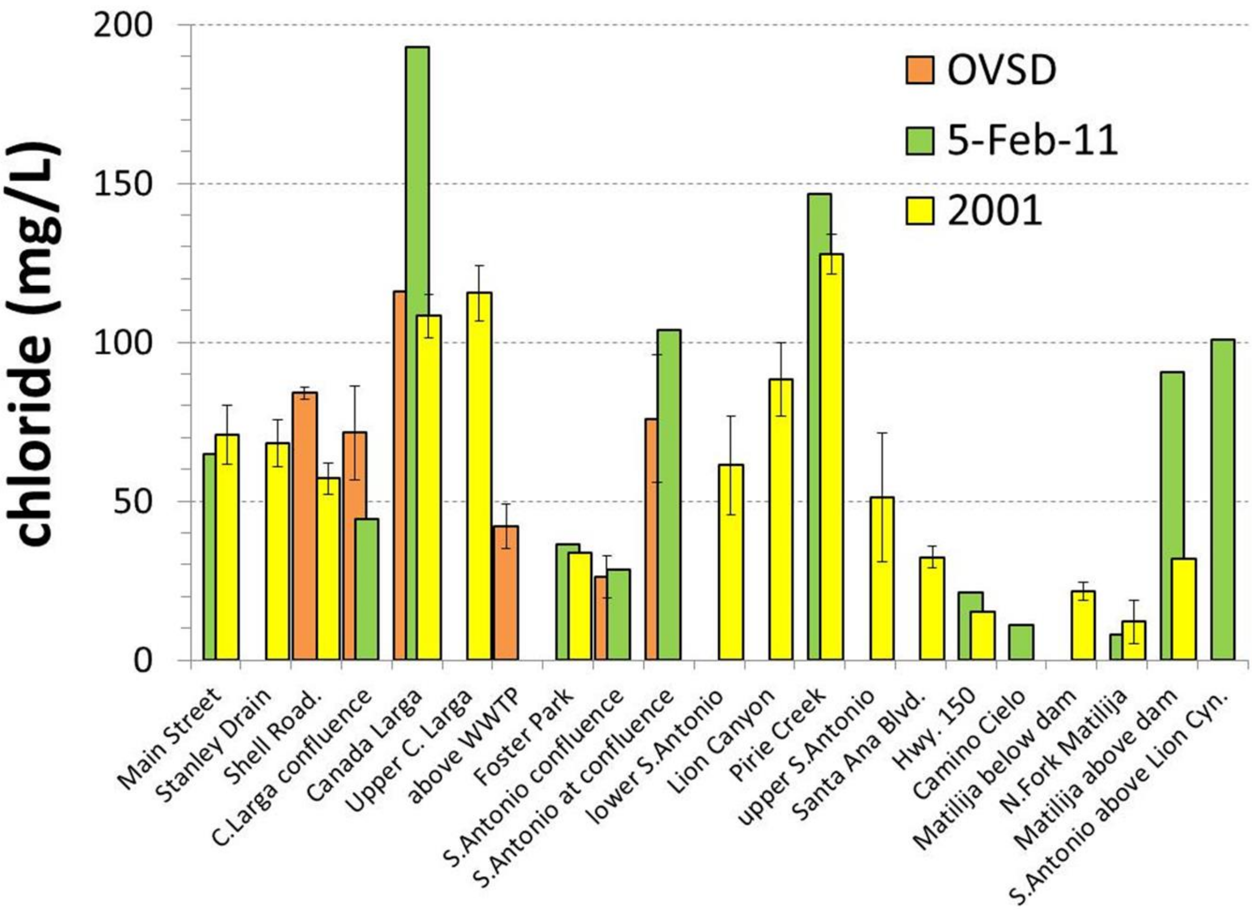


This chart takes a closer look at some Ventura watershed sampling locations: total dissolved nitrogen (TDP) and total dissolved phosphorus (TDP) are shown along with nitrate and phosphorus (using a linear scale with concentrations in µg/L). [Mean seasonal SBCK nitrate and TDN, 2001-08, mean phosphate and TDP, 2005-08]

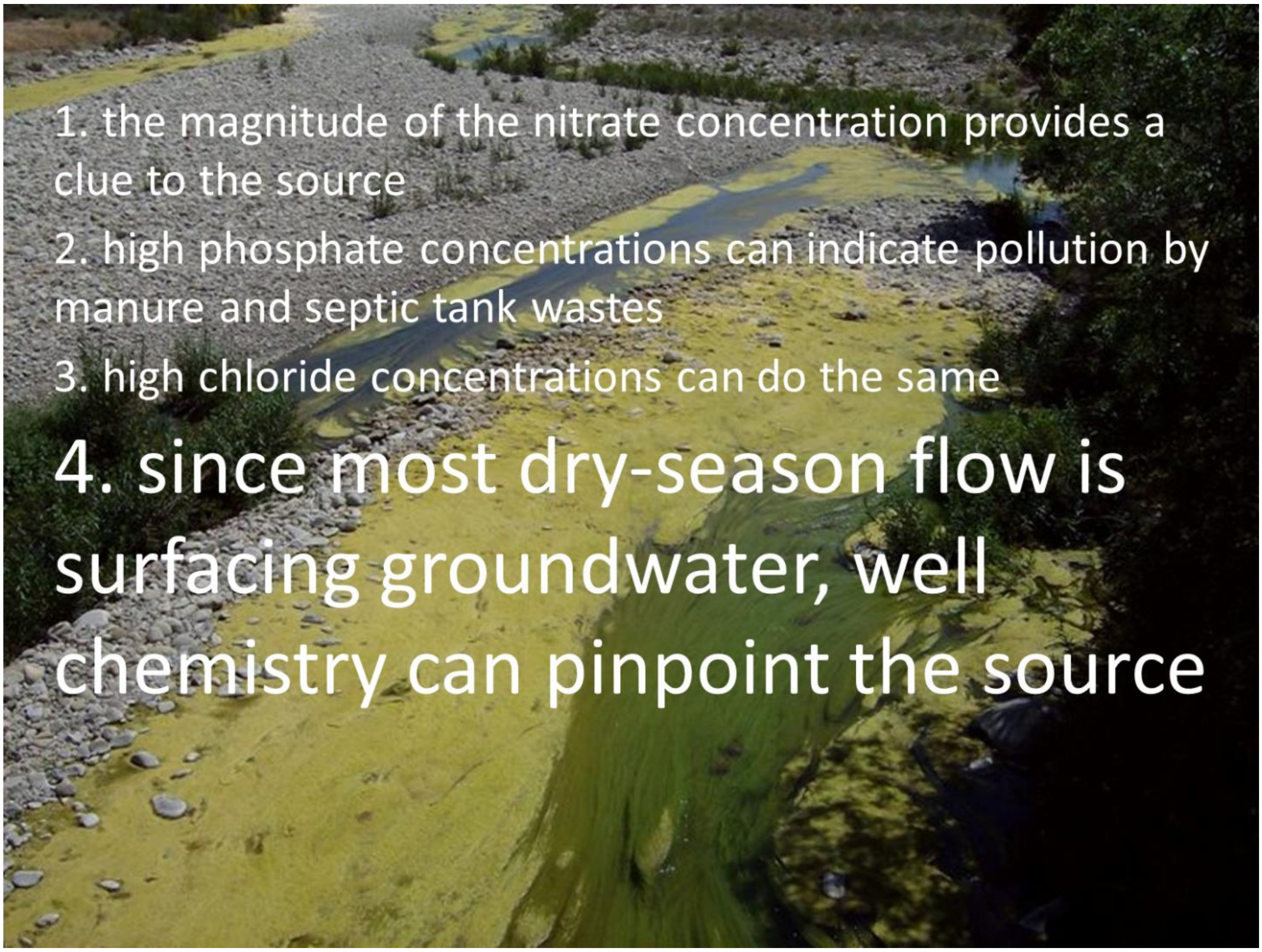
The contrast between upper S. Antonio (very high nitrogen/low phosphorus) and Pirie Creek (moderate nitrogen/high phosphorus) is clear. That phosphorus concentrations at Pirie are similar to concentrations at upper Canada Larga and in Lion Canyon – catchments devoted primarily to animal grazing – implies a similar animal or human excrement source. That total nitrogen at Pirie is much higher than in the two grazing watersheds implies some kind of additional pollution. The N to P ratios at these monitoring locations support these inferences: >400 at upper S. Antonio, 5 at Lion, and 28 at Pirie. Typical plant growth requires an N to P ratio (by weight) of about 15 (effluent from the Ojai WWTP has a median N to P ratio of 3.5). (That flow at Foster Park has an average ratio of 136 to 1 points to agriculture as a probable major source at that location also.)



High chloride concentrations can also be an indicator of contamination by manure or failing septic systems. Aside from natural sources (geologic salt deposits, etc.), chloride can come from septic systems, wastewater treatment plant effluent, animal waste (we, and other animals, excrete chloride in our sweat, urine and excrement) and potash fertilizer (potassium chloride; potassium is a necessary plant nutrient). Disposal of water softener back-wash brine to a septic tank or to the ground can also appreciably increase chloride concentrations in catchment streams.

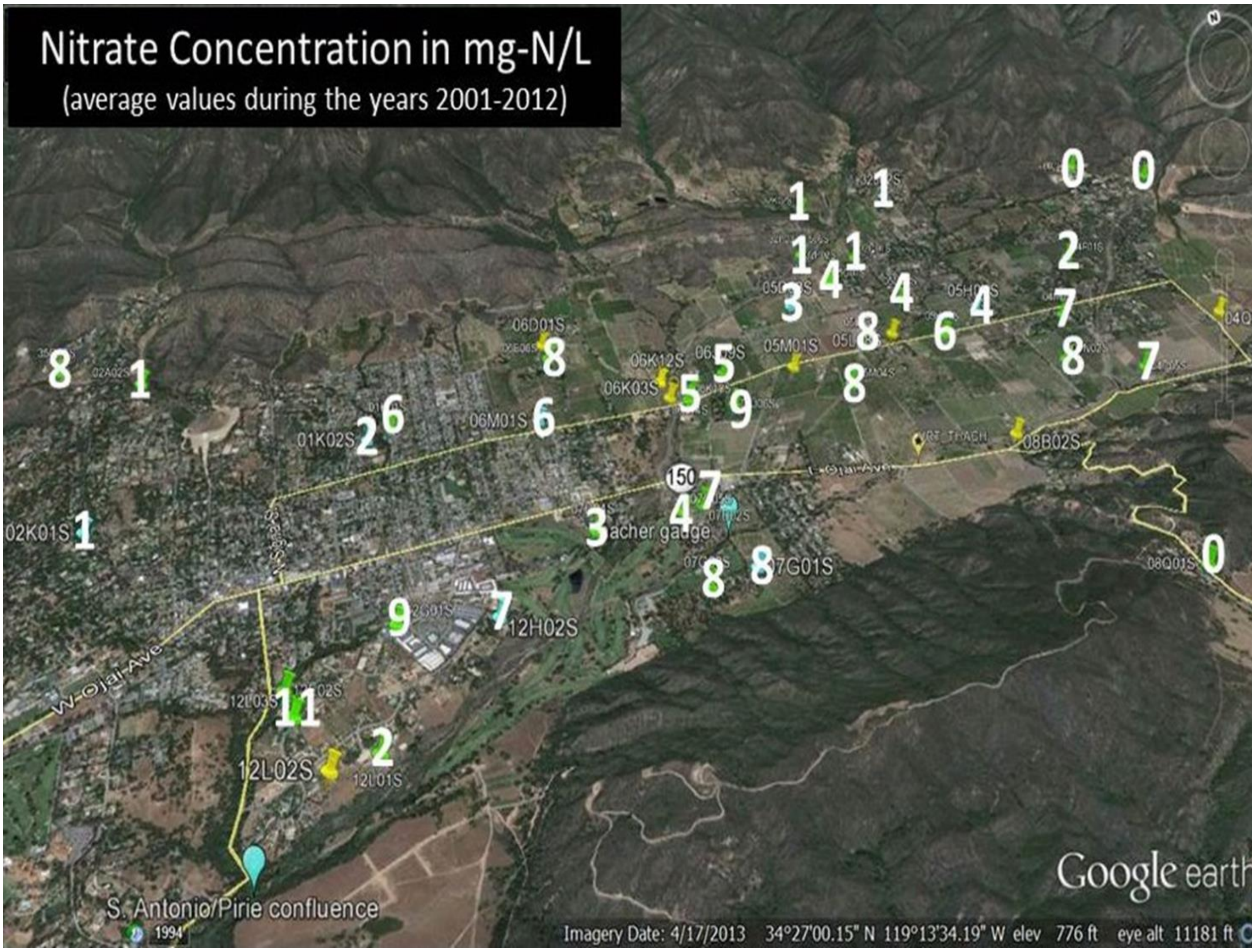


Chloride is not regularly sampled by SBCK in the Ventura River and its tributaries (which is a probably a shame since it's a relatively easy and inexpensive test), but it was sampled extensively during the 2001 dry-season and again in February 2011. These results are shown in the graph along with the results of some limited sampling by OVSD (the error bars indicate 2*SE-mean). Pirie and Lion creeks, and Canada Larga have the highest chloride concentrations. Sites downstream of Pirie also show elevated chloride levels (as would be expected). Note that the Pirie concentration is much higher than at upper S. Antonio. There are also elevated concentrations below the WWTP, but these are not as high as we might expect because of dilution by low-chloride water coming from upstream (compare with Foster Park; WWTP effluent itself has an average chloride level of 117±4 mg/L, comparable with concentrations on Canada Larga and Pirie). Again, the Pirie chemistry points to some form of excrement pollution, whereas that of upper S. Antonio looks quite different (more similar, but higher, to what is seen on the main stem of the Ventura River).

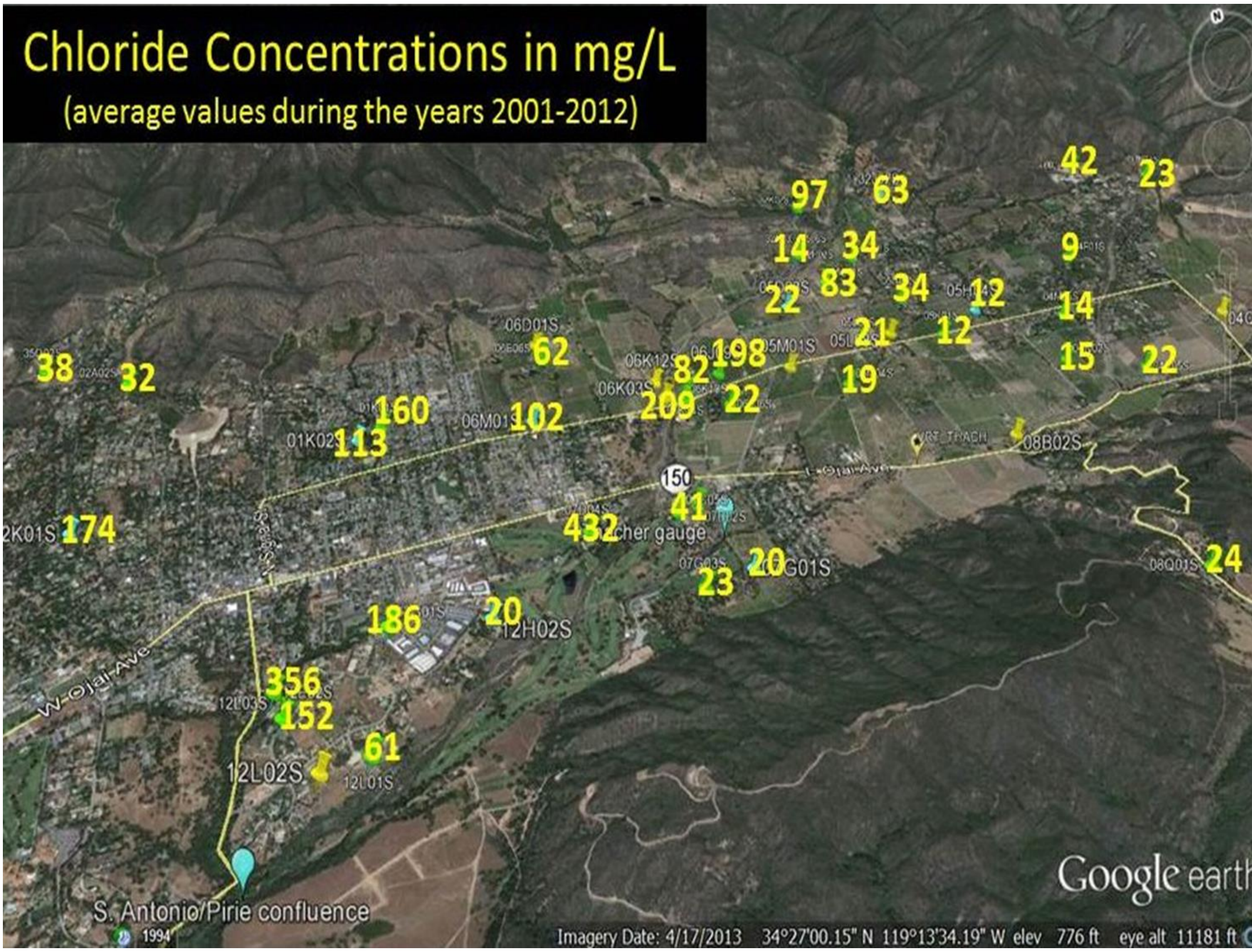


Up to this point we've only examined stream chemistry. But since almost all dry-season flow in the Ventura River and its tributaries is surfacing groundwater, well data and the chemistry of well water can also tell us a lot about the source of contaminants. Luckily, Ventura County has excellent well data.

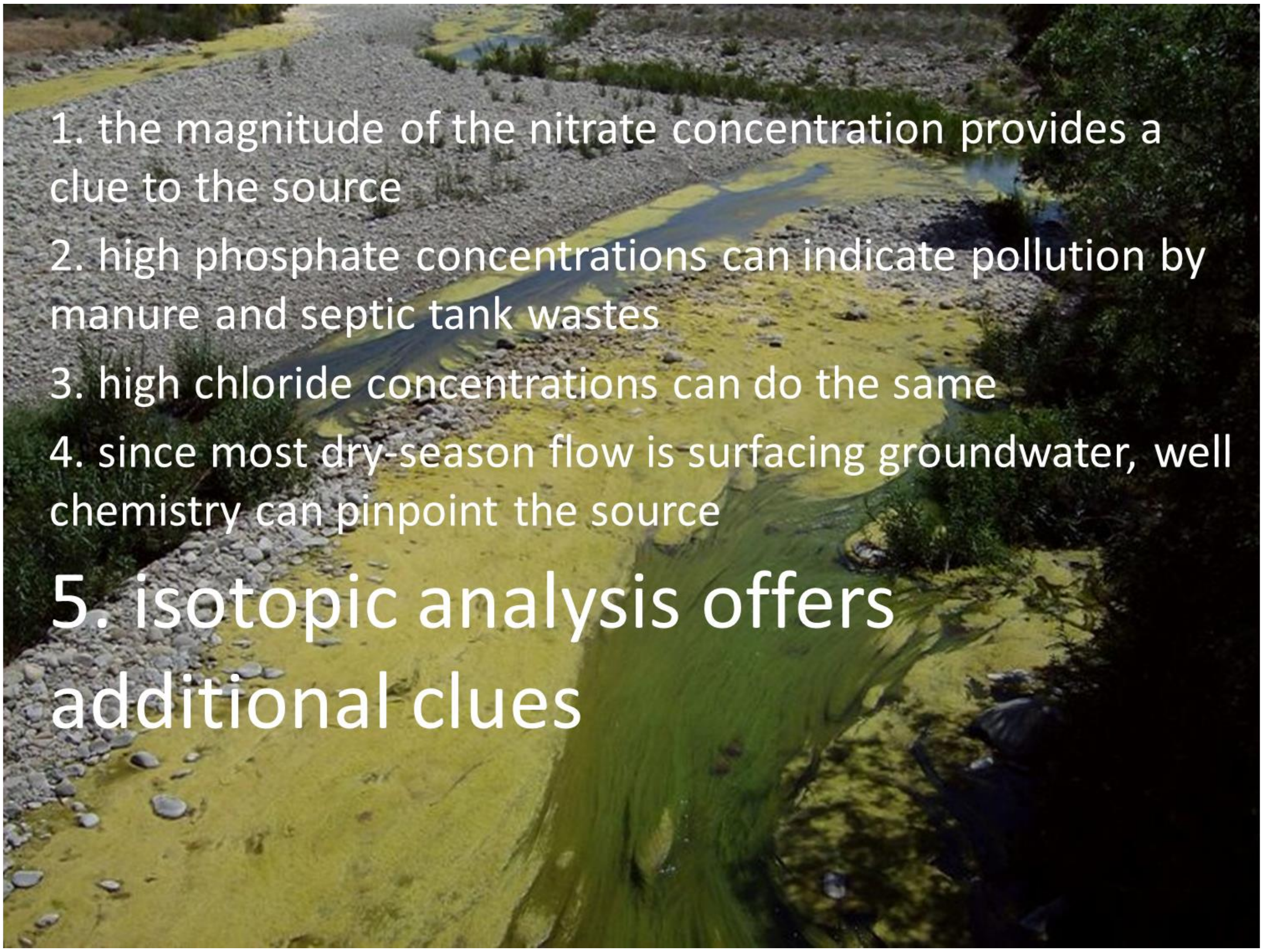
[A good rule of thumb in a semi-arid environment: if there is water in a stream during the summer dry-season it's either groundwater or water someone has put there – irrigation or over-watering runoff (either directly or via an elevated local water-table), urban nuisance waters (car washing and such) or wastewater dumping; in the absence of rain there are no other options.]



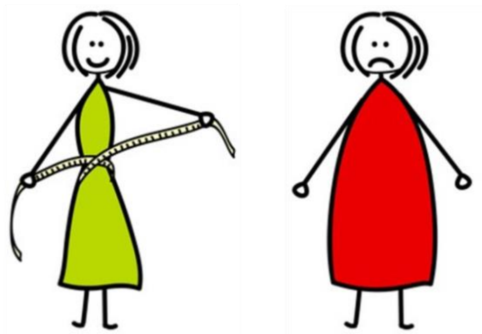
This is an aerial view (my thanks to Google Earth) of the Ojai valley upstream of the S. Antonio/Pirie confluence (the blue "bulb" in the lower left). The white numbers indicate well-water nitrate concentrations at specific well locations. These represent the average of values recorded over the years 2001-2012 (very few wells are tested every year and some were tested only once or twice during this period). [Nitrate concentrations are given in milligrams of *nitrogen* per liter; the County records nitrate in in units of milligrams of *nitrate* per liter. There's a big difference: the Public Health drinking water limit is 10 mg-N/L in the first case, 45 mg-NO_3/L in the second.] The difference between urban and ag is easily seen in the photo, and the highest nitrate concentrations generally underlie the "green" ag areas. Concentrations also generally increase with the downslope flow of groundwater (from upper-right to lower-left). There is a clear, easily noticeable, land use difference between the upper S. Antonio (agriculture) and Pirie (urban/suburban) drainages.

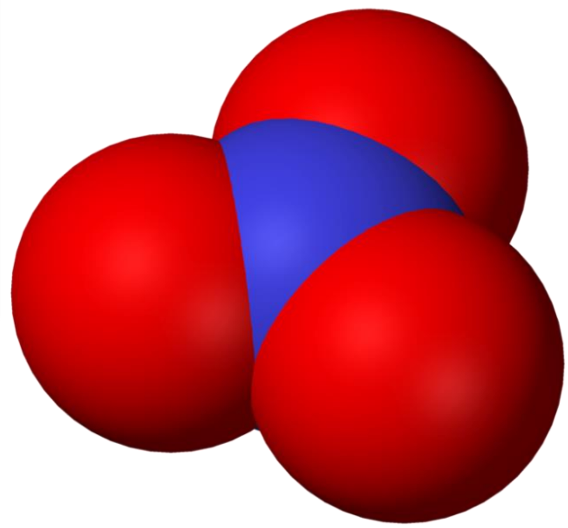


The same aerial view, but now showing average 2001-2012 water-table chloride concentrations in yellow (in mg/L). Again, there is a clear difference between concentrations in the urban vs. the agricultural areas, but the difference is now reversed: the ag areas showing much lower chloride values than the urban (in some cases more than 10-times lower). The groundwater data reinforce the conclusions made from stream chemistry: that high nitrate in upper San Antonio Creek is primarily due to agricultural fertilizer use while high phosphate and high chloride in Pirie are being caused by excrement of one form or another (possibly animal, possibly improperly placed or inadequate septic systems). The higher than usual nitrate in Pirie may be coming from some combination of these same sources and other sources typical of the urban/suburban environment (fertilizer used in landscaping would be a major suspect).



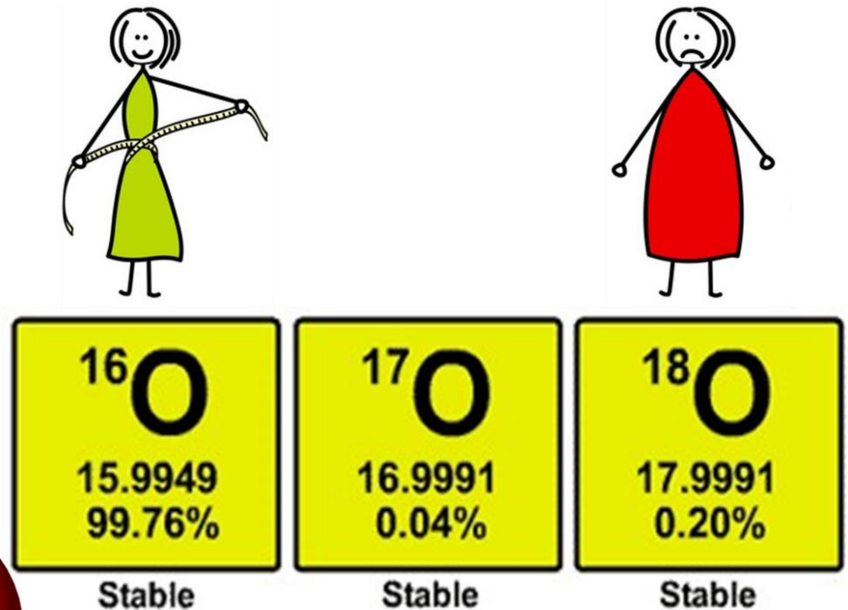
So we have lots of evidence, but it's all of the circumstantial kind. No smoking gun. No incontrovertible DNA. Not even a lousy undeniable fingerprint. So this Spring SBCK collected samples from their usual monitoring sites and sent them off to be analyzed by a friend of mine at University of California, Riverside. The price was right: no charge. The purpose of the analysis was to look at nitrate molecules in each of the samples for isotopic evidence. As to what is isotopic evidence . . . And why it might be of interest . . . We need to make a slight digression.

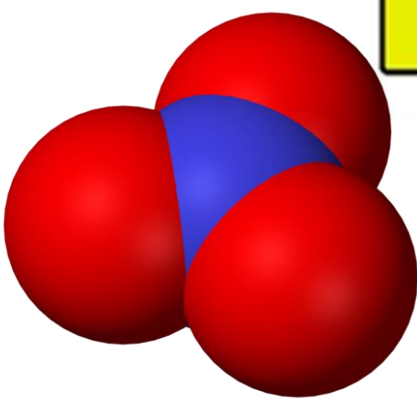




Let’s go back to the nitrate molecule – made up of one nitrogen and three oxygen atoms. Nitrogen comes in two flavors: ^{14}N (called N-fourteen), the lighter isotope, and ^{15}N, the heavier. Think of ‘em as fat and skinny; fat atoms act exactly like skinny atoms but contain an extra neutron or two in their nucleus. During biological transformations (assimilation, nitrification, denitrification) the lighter isotope is preferentially used – it’s easier for an organism to use the more energetic version. In other words, the lighter isotope (^{14}N) becomes more concentrated in the transformed product, while greater amounts of the heavier isotope (^{15}N) are left behind. As an example, when nitrate is denitrified by bacteria into nitrogen gas the gas ends up having less of the heavier ^{15}N isotope (becomes isotopically lighter) while the remaining nitrate becomes isotopically heavier (containing more of the left behind ^{15}N).

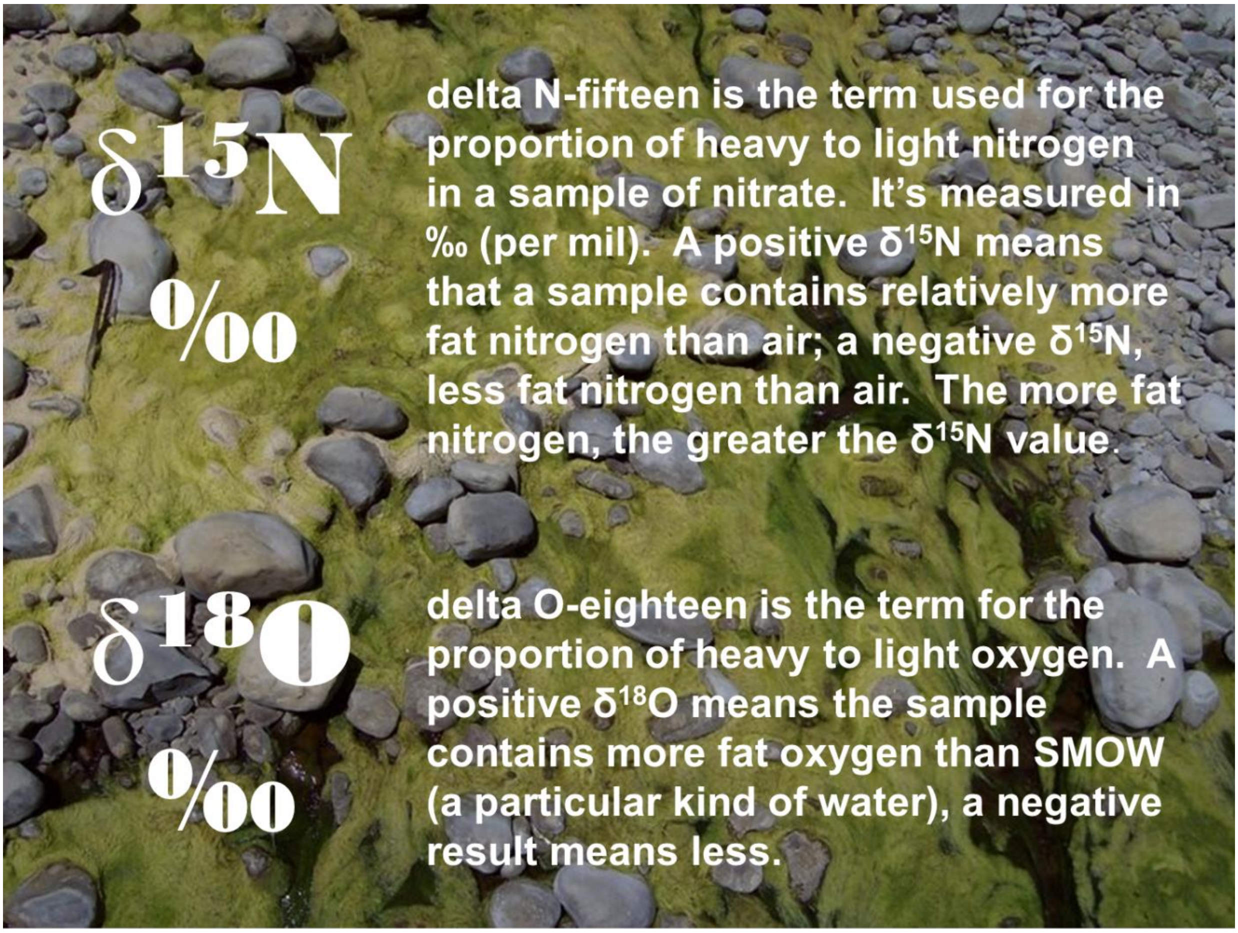
The whole process of discriminating between isotopes is called “fractionation.” The fractions in fractionation are very small. There are relative few fat nitrogen atoms: 99.6% of all nitrogen atoms are of the lighter (or skinner) ^{14}N flavor; only 0.4% are fat. Isotopic analysis measures the relative proportion of heavy to light atoms in a sample; it yields very small numbers and to make things easier results are expressed in comparison with a known standards: air in the case of nitrogen.





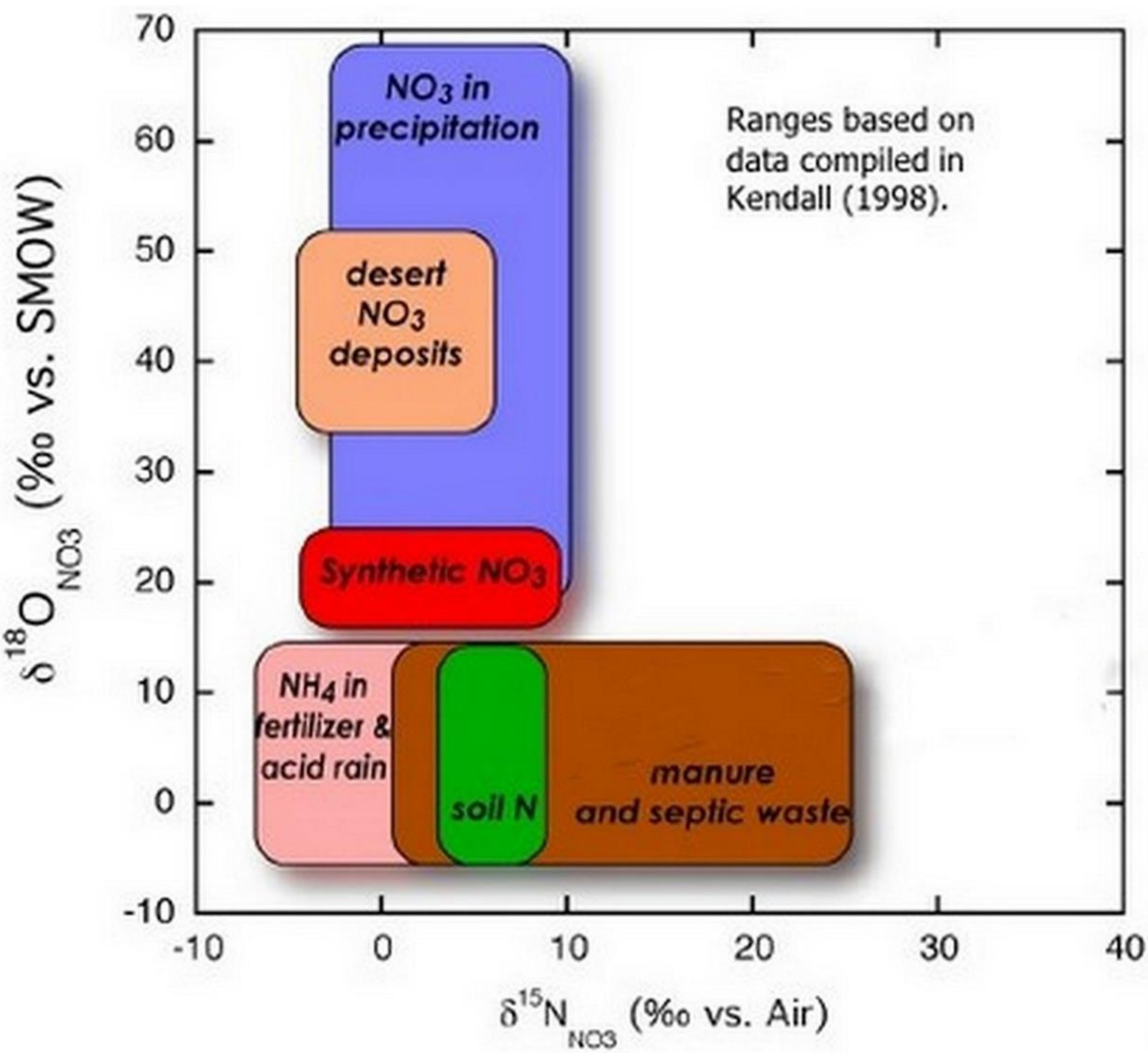
Just as the nitrogen in a nitrate molecule can be either fat or skinny, any of the oxygen atoms can also be fat or skinny. The skinny, or light, oxygen atom is called ^{16}O (oxygen-sixteen), the fat, or heavy, version ^{18}O (oxygen-18). As with nitrogen, the vast majority of oxygen atoms are skinny (99.8% are skinny, 0.2% are fat). And as with nitrogen, biological processes fractionate between different oxygen isotopes. As can physical processes. For example, evaporation, which requires energy to transform a water molecule from liquid to vapor, leaves more of the heavier ^{18}O behind, while rainfall, which represents the loss of energy as vapor becomes liquid, contains a higher percentage of the less energetic fat guys.

As there are fat nitrogen or oxygen atoms, there are also fat nitrate molecules (usually containing either a fat nitrogen or a fat oxygen, very, very rarely more than one fat atom – if there is only a 0.2% chance of running into a fat oxygen in a nitrate molecule, the chance of running into two fat oxygen in the same molecule is 0.0004%, a chance of only four in a million).

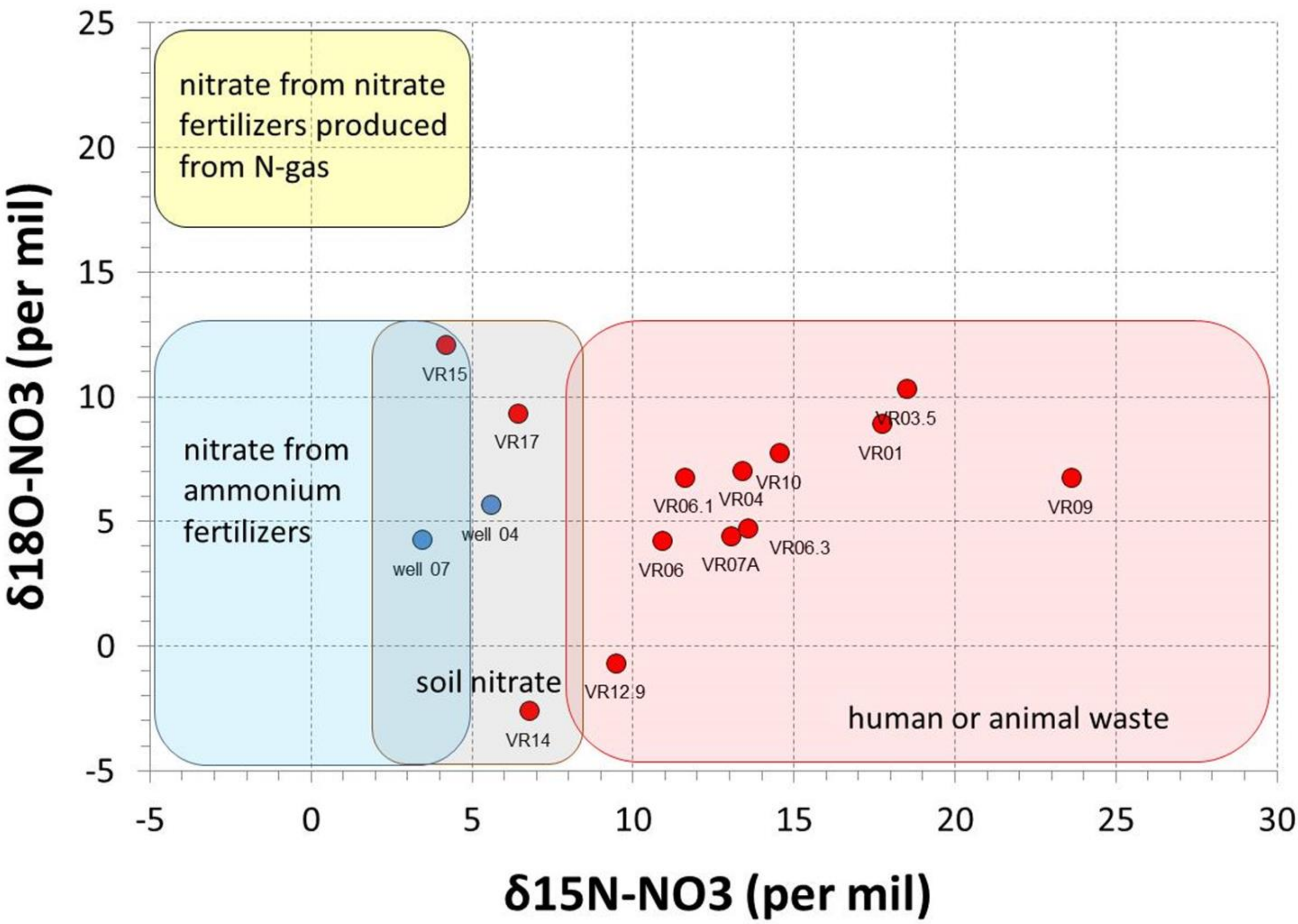


Fractionation produces very small differences in the ratios between fat and skinny isotopes (usually in the third, fourth or even fifth number after the decimal point) and a special notation is needed to show these small differences in a meaningful way: "delta" notation (as defined above) does that. A more formal definition of delta notation would be the isotopic ratio of the sample minus the isotopic ratio of the standard all divided by the isotopic ratio of the standard, and then multiplying this result by 1000 to end up with a recognizable number.

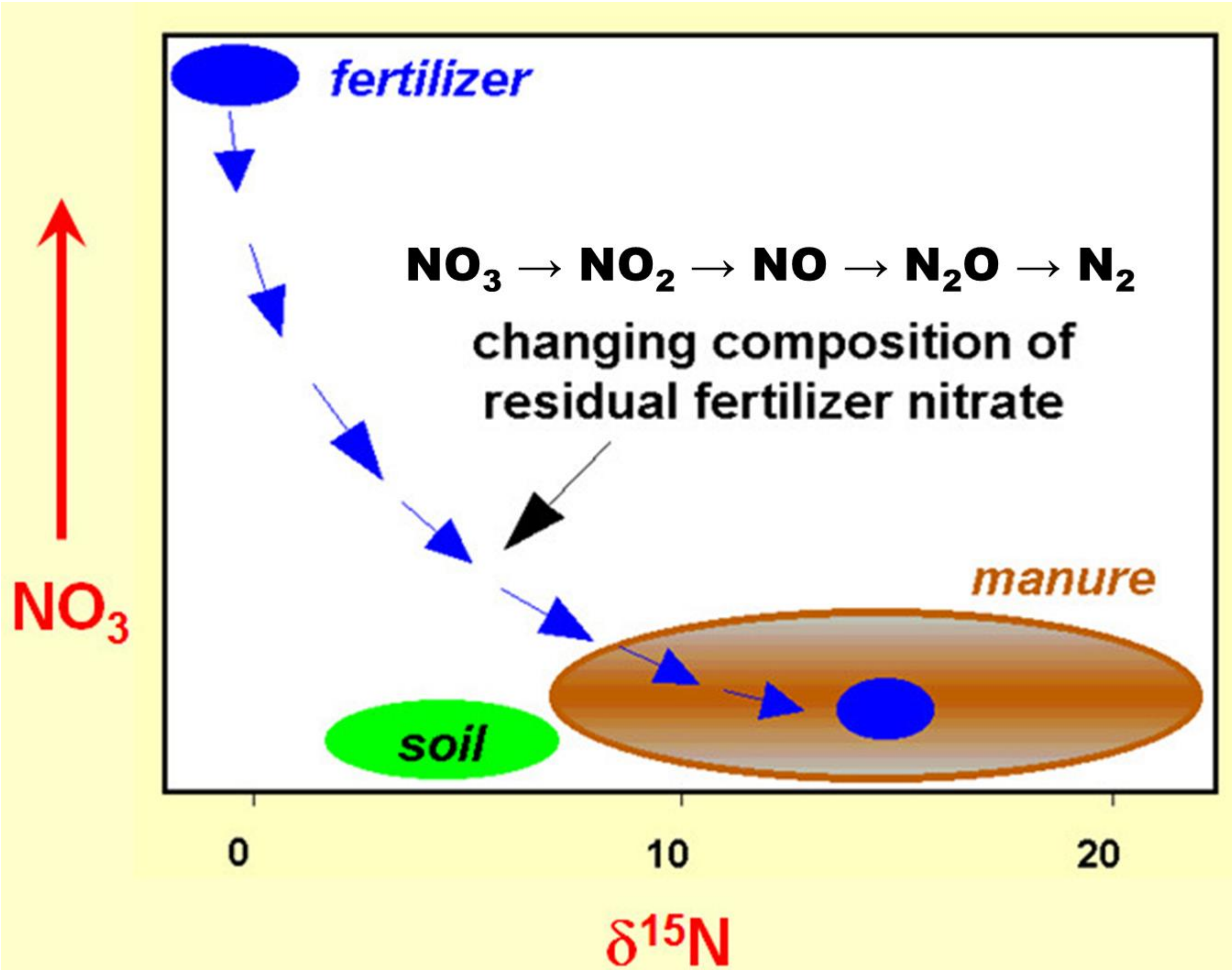
The result is expressed as per mil ((‰) i.e. parts per thousand (just as percent (%) means parts-per-hundred, per mil (‰) is defined as parts-per-thousand). But all one really needs to know is this: with isotopic results a positive number means more fat atoms than the standard, a negative number means less; and the larger the number the further away the sample is from the standard. (The background photo is an algal close-up taken from the Hwy. 150 Bridge in August 2005.)



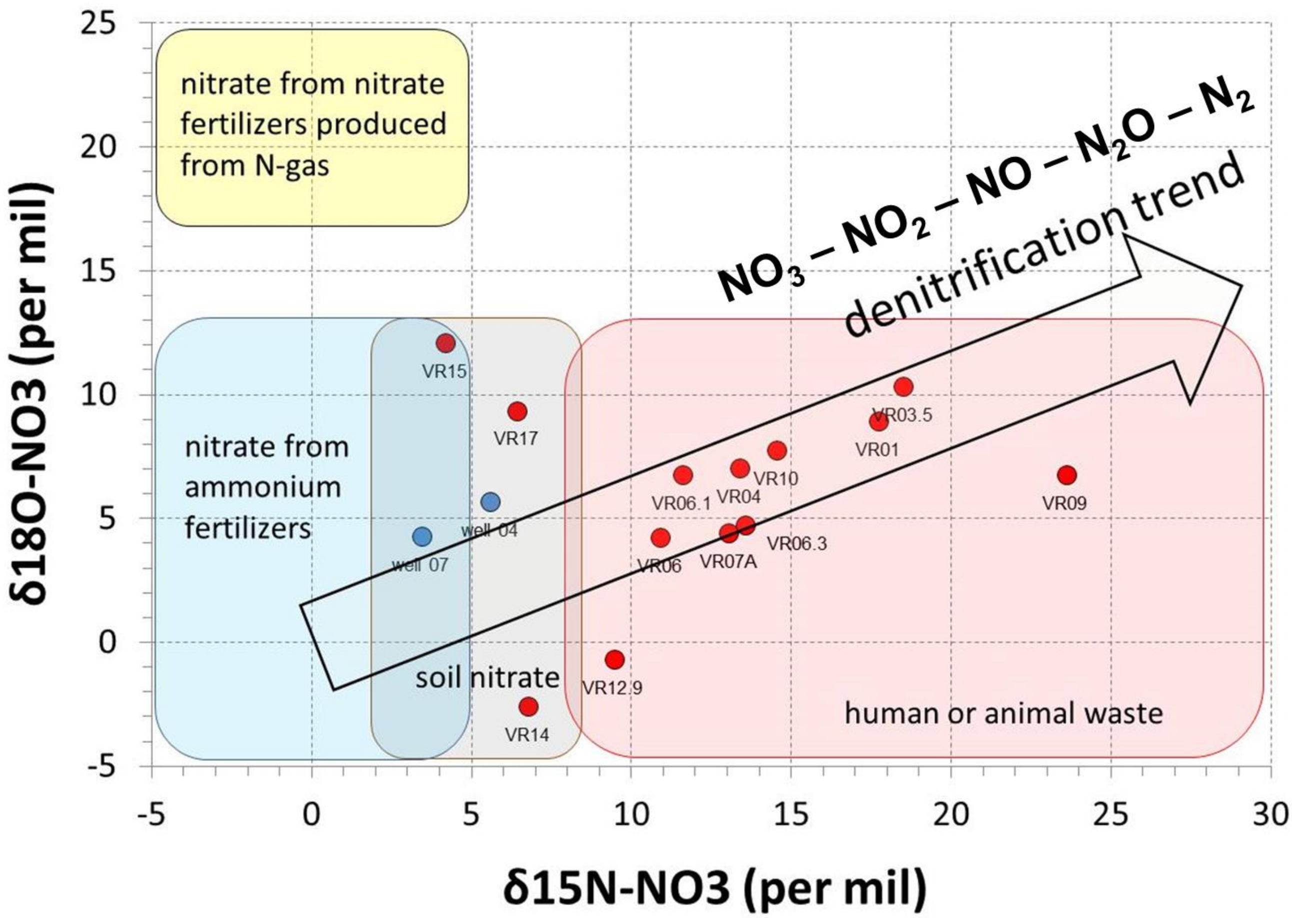
Nitrate molecules in a water sample can be analyzed for the relative proportions of fat to skinny nitrogen *and* fat to skinny oxygen. Two isotopic signatures are better than one if the objective is to determine the nitrate source (just as it is better to know *both* the distance *and* the direction to some destination); the chart shows the approximate regions of δ^{15}N and δ^{18}O for different sources of nitrate. The heaviest δ^{15}N values are found in manure and septic tank wastes (fat N concentrates as it works its way up the food chain and in waste products, and manure from mammals is at the end of a long line of previous processes); the heaviest δ^{18}O is found in “wet deposition,” the nitrate that accompanies rain (formed from various oxides of nitrogen and the high proportion of fat oxygen molecules that accompany rain).



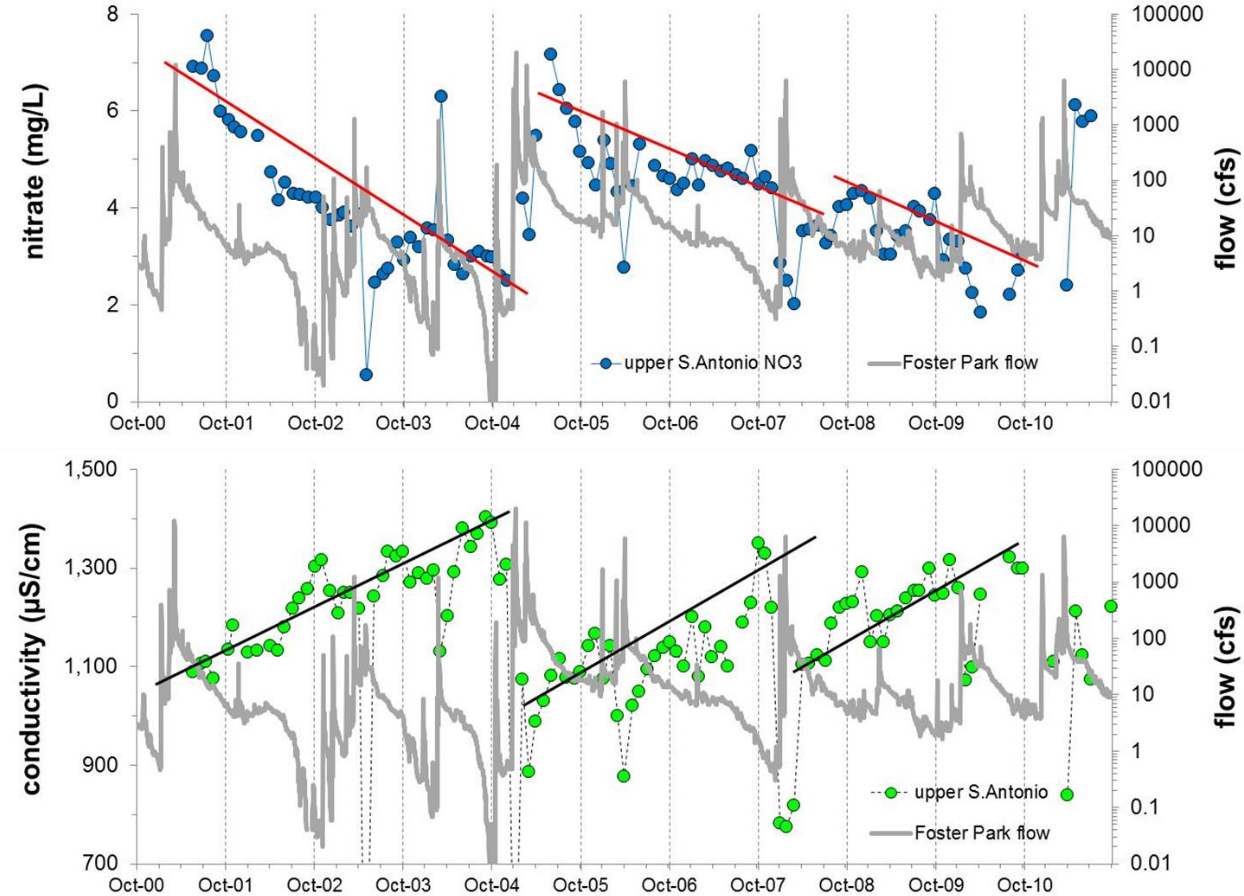
But while knowing two things is better than knowing only one, it still might not be enough to adequately determine the source. The SBCK results are plotted here on a slightly simpler version of the previous graph. Matilija Creek (VR15, above the dam) and N. Fork Matilija Creek (VR14) plot nicely in the soil nitrate box – just what we might expect of these rather pristine waters. But so does middle S. Antonio Creek (nr. Lion Canyon), a location with lots of cattle and horses. All the other watershed locations fall within the human and animal manure box. Except in the case of Pirie Creek (VR09), which undoubtedly ended up exactly where expected, the other results can not, as yet, be considered definitive. The major problem is that the isotopic signature of nitrate can change along the path from source to stream, and in the stream itself – especially if travel along that path takes considerable time. [Wells 04 (in the upper S. Antonio drainage above the Pirie/S. Antonio confluence) and 07 (near the Ventura River in the vicinity of Miners Oaks) represent groundwater isotopic results collected by USGS in Apr.-June 2007; the only other Ventura isotopic values I could locate.]



The particular problem is that denitrification (de-ni-tra-fi-ca-tion) of fertilizer nitrogen in a low-oxygen, water-table environment increases both the $\delta^{15}N$ and $\delta^{18}O$ signatures of nitrate – making it look a lot like manure or septic waste. The cartoon shows the path of increasing $\delta^{15}N$ as nitrate (NO_3) is converted to nitrous oxide (N_2O, probably more familiar as "laughing gas") and nitrogen (N_2). Both N_2O and N_2 exit the water-table as gasses, leaving the remaining nitrate behind; and the remaining nitrate is now both lower in concentration and isotopically heavier (recall that biological processes preferentially use the lighter, or skinner, molecules, excluding more of the fat guys). And we know from the sampling results on upper San Antonio Creek that denitrification within the water-table is taking place.

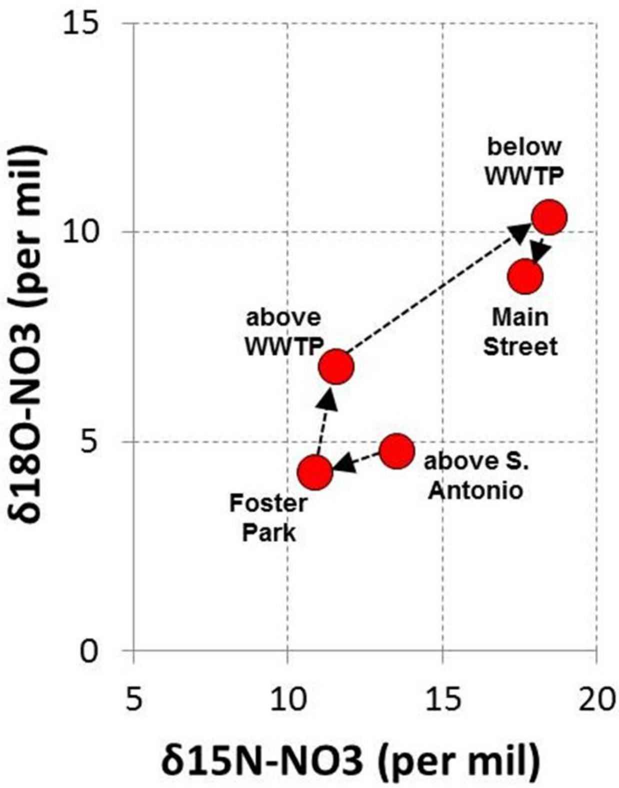


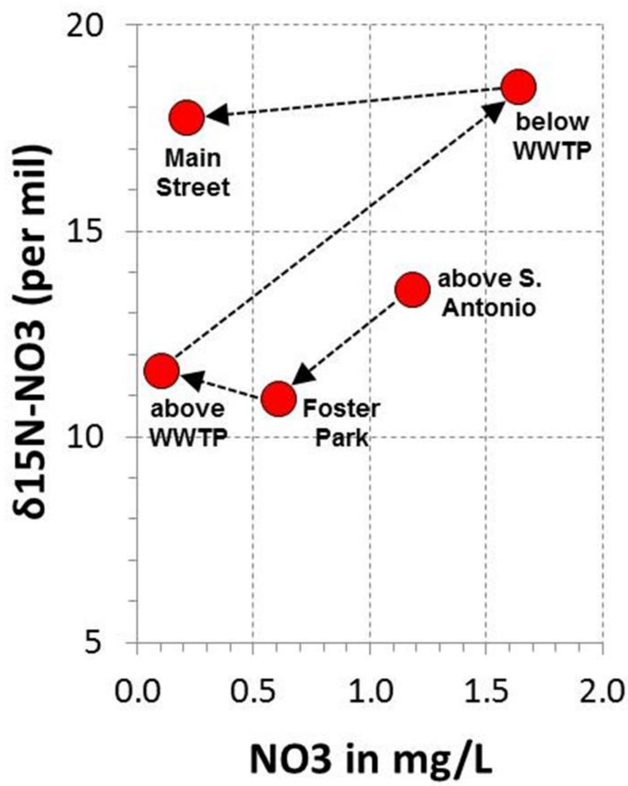
Denitrification affects both the $\delta^{15}N$ and $\delta^{18}O$ values and follows a particular path, the path indicated by the broad arrow on the graph; denitrification changes $\delta^{15}N$ more than it does $\delta^{18}O$ (the ratio of change is roughly 2 to 1) and produces this characteristic slope. Given the possibility of denitrification, the nitrate in any of the samples around the vicinity of the arrow could have initially started out as ammonium fertilizer (or soil nitrate in the case of Camino Cielo on the upper Ventura River). The only exception, as mentioned earlier, being Pirie Creek where the isotopic result (human or animal waste) can be considered relatively conclusive. It should be kept in mind that other processes, such as nitrification of ammonium or biological uptake, or something as simple as mixing nitrate from two or more sources, can also modify the isotopic signature and complicate analysis.



So, is denitrification occurring in the groundwater underlying Ojai and other areas of the Ventura watershed? The simple answer is yes. But the evidence is again, at this point, circumstantial. There is, of course, the USGS well data from 2007 that I've plotted on earlier graphs. It shows groundwater nitrate, in samples taken earlier in the season and at a time of more recent and substantial recharge (2005 & 2006 were both good rainfall years) having significantly lower isotopic values – values that lie nicely at the lower end of the denitrification trend shown by this year's stream samples.

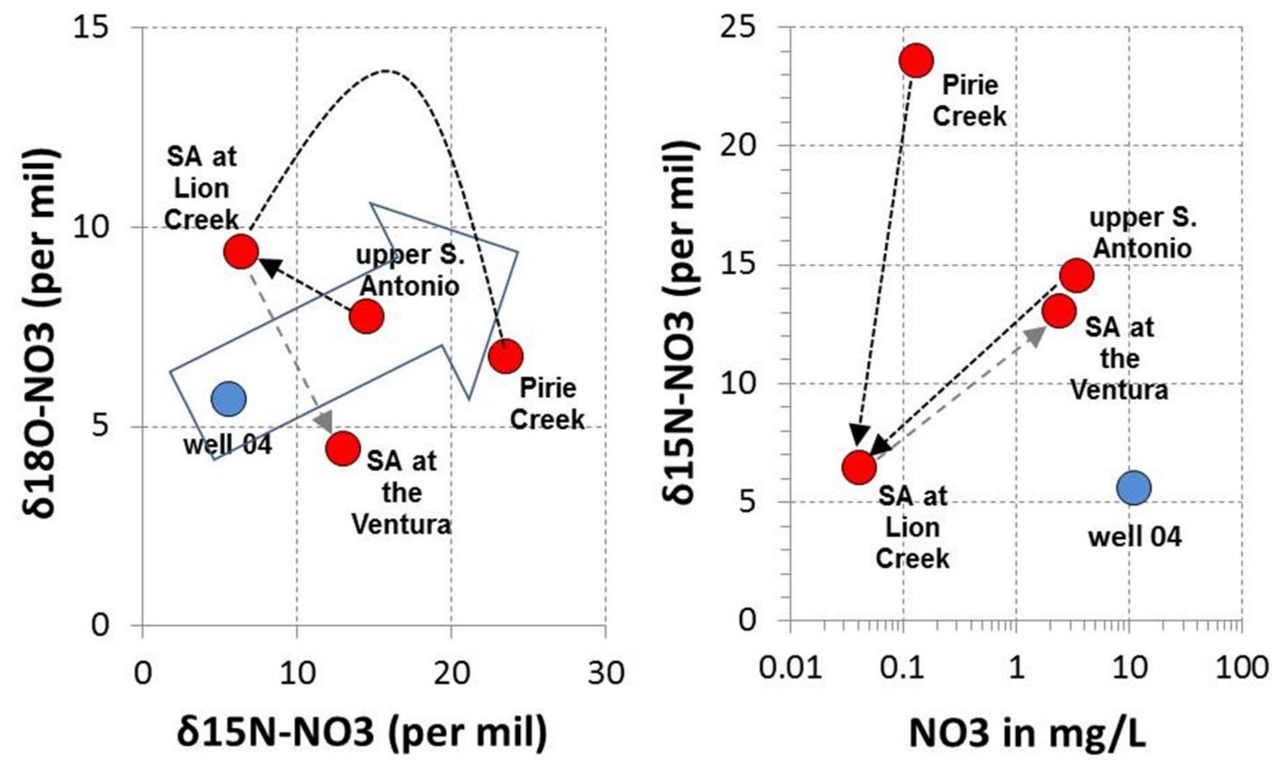
And there is other evidence: The figure shows monthly nitrate (the upper graph) and conductivity (the lower) measured on upper S. Antonio Creek since 2001. Notice that nitrate concentrations reach a maximum just after the rainy season in big rainfall years (2001, 2005, 2008 & 2011), while at the same time conductivity drops to a minimum. In-between, during low rainfall years of little or no recharge, nitrate concentrations decline as conductivity increases. The conductivity increase is due to groundwater aging between episodes of significant recharge (caused by longer times of contact with water-bearing sedimentary strata); the nitrate decrease from denitrification (the loss of nitrate to bacteriological production of N_2O and N_2 gases).





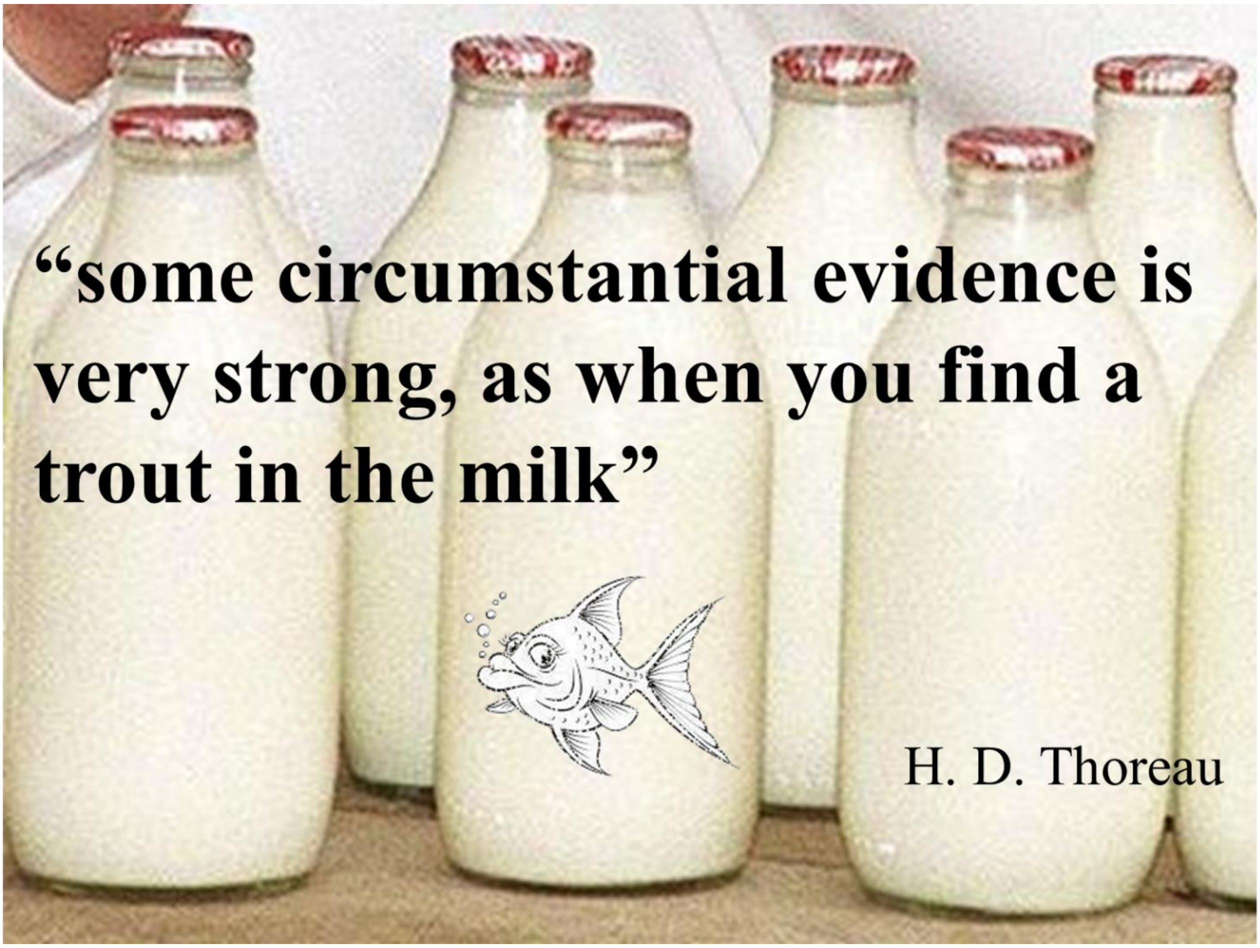
main stem of the Ventura River

I'll use the lower Ventura River results to illustrate how both isotopic data and nitrate concentrations along a path of flow can be analyzed. The graph on the left plots $\delta^{18}O$ vs. $\delta^{15}N$; on the right, $\delta^{15}N$ is plotted against nitrate. Starting just above the S. Antonio confluence, partially denitrified groundwater surfaces in the river. While flowing towards Foster Park biological uptake reduces the nitrate concentration, but since uptake should leave isotopically enriched nitrate behind, the reduction shown is probably related to additional groundwater inflows of lower isotopic content. Flow from Foster Park to just above the WWTP exhibits the nitrate decrease and isotopic enrichment expected of continued uptake and assimilation. As flow passes the WWTP the addition of treated sewage effluent increases both the isotopic signature and the nitrate concentration. From below the WWTP (just above the Canada Larga confluence) to Main Street, nitrate concentrations decrease due to uptake, but the isotopic reduction is again unexpected. It could be caused by either the addition of Canada Larga water (which is isotopically lighter, although the proportional reduction does not fit a simple mixing model) or the entry of ag runoff just above Main St. or both.



San Antonio Creek

Looking at the San Antonio results: Flow on the middle S. Antonio at Lion Canyon originates from the upstream confluence of upper S. Antonio (primarily agricultural) and Pirie (developed and suburban Ojai) creeks; upper S. Antonio is the dominant contributor. That middle SA has a very different isotopic signature than either of these upstream tributaries indicates that flow was probably not continuous between these points and we are simply looking at some other nearby source of nitrate. The depleted $\delta^{15}N$ signature would seem to eliminate the most probable alternative, Lion Canyon: low in nitrate due to appreciable algal uptake, but noted for contamination by cattle and horses. Similarly, flow was not continuous between middle and lower S. Antonio; the water in lower S. Antonio at the Ventura confluence comes from the same groundwater source supplying the adjacent Ventura River (conductivity measurements support this conclusion, as do similar isotopic values shown in earlier graphs). Well 04 (USGS data collected Apr.-June 2007), located upstream of VR10, suggests that denitrified, ag-contaminated, groundwater is the probable source of the isotopic signature in upper S. Antonio; other evidence substantiates appreciable water-table denitrification in low rainfall years.



So at the end it's still all circumstantial. But men have been hanged with less evidence. As Henry David Thoreau said, "some circumstantial evidence is very strong, as when you find a trout in the milk."