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Swedish monitoring programme in terrestrial biota

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SAKRAPPORT

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Metaller i älg

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TIME TRENDS OF METALS IN LIVER, KIDNEY AND MUSCLE OF MOOSE (*Alces alces*) FROM SWEDEN, 1980-2004

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Introduction

The long-term monitoring of persistent and bio-accumulating chemicals in the Swedish environment is part of the Swedish National Environmental Monitoring Programme. It is based on chemical analyses of tissues and organs from species collected in selected reference areas of the Swedish mainland, lakes and coastal areas (Odsjö & Olsson 1979a,b, Bernes 1985). As part of the terrestrial contaminant monitoring programme, specimens of muscle, liver and kidney of moose (*Alces alces*) have been collected since 1980 from Grimsö, a reference area in the monitoring programme and a coherent hunting district in the Örebro county (T) in south-central Sweden. In 1996, the monitoring was extended by collection and chemical analysis of organs of moose from six further counties and districts in Sweden. These districts are situated in the Norrbotten county (BD), Jämtland county (Z), Västmanland county (U), Älvsborg county (P), Jönköping county (F) and Kronoberg county (G) (Figure 1).

Moose, with a diet dominated by twigs and leaves of trees and shrubs (Cederlund *et al.* 1980), was chosen in the monitoring programme as a representative of biota in the Swedish forest areas. Since the moose is distributed almost all over the country, it was considered as an ideal material also for studies of spatial distribution of environmental pollution and bioaccumulation, which is the reason for the extended collection of samples in 1996 onwards.

This report presents levels and time trends of Ca, Cd, Co, Cr, Cu, Fe, Mg, Mn, Mo, Ni, Pb, V and Zn in liver and kidney and Se and Hg in liver and muscle from the period 1996-2004. For Grimsö data from an extended period, 1980-2004 is presented.

Material and methods

Grimsö area

From the start of the collection in the Grimsö area (Figure 1), samples of liver, kidney and muscle have been collected from approximately 45-50 individuals annually during the hunting season in the autumn and, with special permit also in the winter and spring. Samples were taken from all individuals shot in the area during hunting despite age and sex. This was done to make it possible to select the most appropriate and homogeneous material for contaminant monitoring according to influence of biological variables (*e.g.* age, sex, etc.) on the concentrations. The samples were extracted at the slaughter, prepared in laboratory and stored in a temperature of -30°C until analysis. Individual age was determined by tooth sectioning after slaughter. Calves and, certain seasons also males were initially well represented in the material. However, the age structure of the material has changed considerably during the period, which may have consequence in future for the choice of material from a smaller and spatial concentrated population like that in the Grimsö area. According to the extended hunting period and date of collection, selection of individual calves for analyses was restricted to the period October 1 - April 30 each hunting season. The selection of specimens started with the earliest shot animals each season. No significant variation in levels of Cd according to date of collection during the hunting season was revealed (Odsjö 2001).

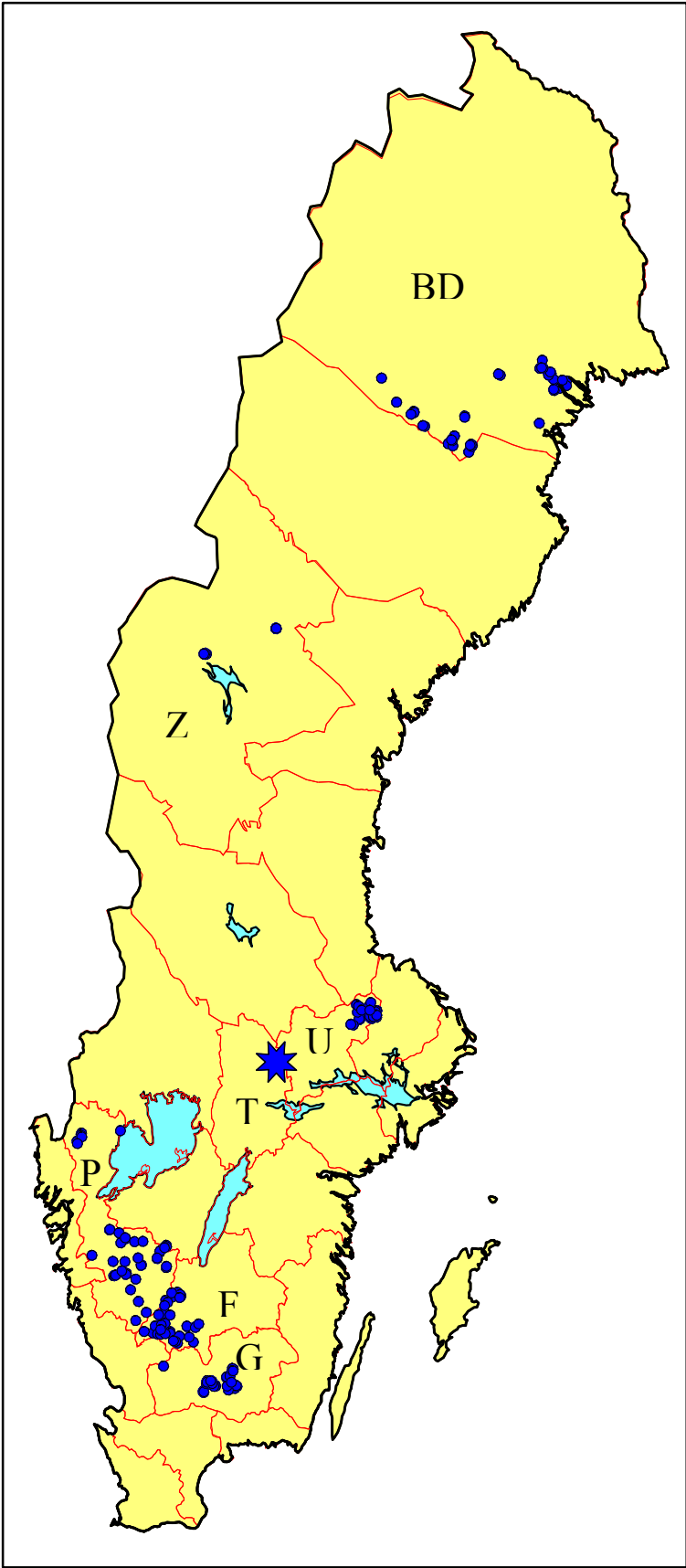
From the Grimsö area, tissue samples of male calves were selected for analyses with some few exceptions that were from female calves. Completion of male samples with samples of females was acceptable after studies of the relation level/sex for cadmium in kidney, which showed no difference according to sex. Further tests and discussion of selection criteria of matrices of moose are reported elsewhere (Odsjö 2001). The time series from Grimsö includes 25 years of analyses and 258 individual samples from calves. For chromium, nickel, lead, vanadium and mercury some samples were either missing or below limit of detection (see below).

Other districts

From the other six districts (Figure 1), samples of blood serum, muscle, liver, kidney, spleen, hair and half the lower jaw (mandible) were collected from approximately 40 calves and 50 adult moose per year. The samples were collected from animals shot during the ordinary hunt in the autumn; mid-October to the end of December in southern Sweden, and September to the end of October in northern Sweden except for three weeks interruption during mating season. These samples were also extracted at the slaughter, prepared in laboratory and stored in a temperature of -25°C until analysis. Specimens of varying ages and sex were selected for analysis. The ages vary from calves (approximately six months) up to animals 2.5 and 3.5 years old. From the six counties, moose at an age of 1 to 3.5 years were selected for statistical trend evaluations of concentrations. This was done to reduce the annual variation to an acceptable level.

In order to achieve information on the within-year variation in concentration of the studied populations, 10 or more individuals (of different ages) were analysed per year and district. However, in some years it has not been able to achieve the required number of requested individuals. From the individual analyses a geometric mean value was calculated and used as a value of the year in the time trend study.

Only two individuals in 2003 and one in 2004 from Älvsborg county (P) were analysed, why results from that years are uncertain.



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Figure 1 Sampling sites in the various counties indicated with dots. The star indicates the Grimsö field station where the moose for the 24-year time series are collected.

Analytical methods

Pre-treatment of samples

The outer layer of the tissue specimens was cut off and samples were taken from the inner part to avoid surface contamination.

Combustion of organs (5 g liver and kidney, respectively, for multi-metal determination; about 5 g liver and muscle, respectively, for analysis of Hg and Se) was performed by automatic wet digestion according to a standard program (Frank 1976, Frank, and Petersson 1983, Frank 1988, Frank *et al.* 1992). Selenium concentrations are not reported in this report. An electrically heated block of aluminium was used (Foss Tecator Digestion System, Model 40, Foss Tecator AB, Höganäs, Sweden). For digestion conditions see Table 1.

Table 1. Conditions for wet digestion of organ tissues.

| Element | Acid mixture (collection year 1996) | Acid mixture (collection years 1997-2004 and Grimsö 1980-2004) | Type of digestion tube | Final temperature of digestion (°C) |
|---|---|--|------------------------|-------------------------------------|
| Ca, Cd, Co, Cr, Cu, Fe, Mg, Mn, Mo, Ni, Pb, V, Zn | 65% HNO ₃ , 70% HClO ₄ | 65% HNO ₃ , 70% HClO ₄ , 95 % H ₂ SO ₄ | quartz glass | 240 |
| Hg | 65% HNO ₃ , 70% HClO ₄ | 65% HNO ₃ , 70% HClO ₄ | boro-silicate glass | 180 |
| Se | 65% HNO ₃ , 70% HClO ₄ | 65% HNO ₃ , 70% HClO ₄ | boro-silicate glass | 225 |

Analysis

Analysis of 13 elements (Ca, Cd, Co, Cr, Cu, Fe, Mg, Mn, Mo, Ni, Pb, V and Zn) in material from 1996 was performed using direct-current plasma atomic emission spectrometry, DCP-AES (SpectraSpan IIIA, Applied Research Laboratories Inc., Valencia, CA, USA) and inductively coupled plasma atomic emission spectrometry, ICP-AES (JY 50P, Jobin Yvon-Horiba SA, 91165 Longjumeau, France) (Frank and Petersson 1983). For material from 1997-2004 the analysis was performed using only inductively coupled plasma atomic emission spectrometry, ICP-AES, (JY 238, Jobin Yvon-Horiba SA, 91165 Longjumeau, France). (The method is accredited since June 1998).

The limits of detection (LOD) for the elements are presented in Table 2.

For the determination of Hg in material from 1996-98, flow injection cold vapour atomic absorption spectrometry, FI-CV-AAS, was used. For the determination of Se in material from 1996-98, flow injection hydride generation atomic absorption spectrometry, FI-HG-AAS, was used (Galgan and Frank 1988, Galgan and Frank 1993). (The methods are accredited since June 1998). The determination of Hg and Se in material from 1999 - 2004 was performed by using cold vapour (CV)- ICP-AES and hydride generation (HG)- ICP-AES, respectively. (The methods are accredited since Oct. 2000).

The limits of detection for Hg and Se for the different techniques are shown in Table 3. Quality control was performed using appropriate reference materials like NIST (National Institute of Standards and Technology) SRM 1577b bovine liver.

The analysis of the thirteen elements in moose tissues from Grimsö, 1980-2004, was performed using ICP-AES and the analysis of Hg and Se by use of CV-ICP-AES and HG-ICP-AES, respectively. (Limits of detection see Table 2 and 3). The number of individual samples from

Grimsö below the limit of detection of the elements is given in Table 4. They have been excluded from calculations of mean values.

The chemical analyses were carried out by the Department of Chemistry, National Veterinary Institute, Uppsala.

Table 2.

Limits of detection (LOD) for the various analytical techniques and years estimated from samples of 5 g, µg/g, fresh weight.

| | 1996 | 1997-2004 other districts | 1998-2004 Grimsö 1980-2004 |
|---------------------|----------|------------------------------|-------------------------------|
| Technique →? | DCP-AES | ICP-AES | |
| Element ↓? | LOD (3s) | LOD (3s) | |
| Ca | 0.146 | 0.44* | |
| Cd | 0.020 | 0.002 | |
| Co | 0.002 | 0.002 | |
| Cr | 0.002 | 0.005 | |
| Cu | 0.018 | 0.15* | |
| Fe | 0.056 | 0.44* | |
| Mg | 0.024 | 0.46* | |
| Mn | 0.010 | 0.014* | |
| Mo | 0.020 | 0.002 | |
| Ni | 0.006 | 0.022* | |
| Pb | 0.020 | 0.008 | |
| V | 0.002 | 0.002 | |
| Zn | 0.112 | 0.62* | |

*=blank+3s

Table 3.

Limits of detection for the various analytical techniques and years, (blank + 3s) estimated from samples of 5 g, ng/g, fresh weight.

| | 1996 | 1997 | 1998 | 1999-2004 |
|--------------------------------|-----------|-----------|-----------|------------|
| Element ?↓ Technique →? | FI-HG-AAS | FI-HG-AAS | FI-HG-AAS | HG-ICP-AES |
| Se | 1 | 1 | 1 | 2.2 |
| Element ?↓ Technique →? | FI-CV-AAS | FI-CV-AAS | FI-CV-AAS | CV-ICP-AES |
| Hg | 0.26 | 0.29 | 0.30 | 1.1 |

Table 4.

Number (n) of individual samples of moose from Grimsö out of a total of 258, found with concentrations below the limits of detection (LOD). (l)=liver, (k)=kidney, (m)=muscle

| Element | n below LOD | % below LOD | Total |
|---------|-------------|-------------|-------|
| Cr (l) | 23 | 8.9 | 259 |
| Cr (k) | 23 | 8.9 | 259 |
| Ni (l) | 67 | 26.0 | 259 |
| Ni (k) | 45 | 17.4 | 259 |
| Pb (l) | 28 | 10.9 | 258 |
| Pb (k) | 3 | 1.2 | 258 |
| V (l) | 70 | 27.1 | 259 |
| V (k) | 57 | 22.1 | 259 |
| Hg (l) | 5 | 8.5 | 59 |
| Hg (m) | 30 | 50.1 | 59 |

Statistical treatment and graphical presentation

Trend detection

One of the main purposes of the monitoring programme is to detect trends.

The slope of the line describes the annual change. A slope of 5% implies that the concentration is halved in 14 years whereas 10% corresponds to a similar reduction in 7 years and 2% in 35 years. See table 5 below.

Table 5. The approximate number of years required to double or half the initial concentration assuming a continuous annual change of 1, 2, 3, 4, 5, 7, 10, 12, 15 or 20% a year.

| | 1% | 2% | 3% | 4% | 5% | 7% | 10% | 12% | 15% | 20% |
|----------|----|----|----|----|----|----|-----|-----|-----|-----|
| Increase | 70 | 35 | 24 | 18 | 14 | 10 | 7 | 6 | 5 | 4 |
| Decrease | 69 | 35 | 23 | 17 | 14 | 10 | 7 | 6 | 4 | 3 |

Legend to the plots

The analytical results from each of the investigated elements are displayed in figures. Each site/tissue is represented by a separate plot except for time series shorter than 4 years.

The plot displays the geometric mean concentration of each year (dots) together with the individual analyses (small dots) and the 95% confidence intervals of the geometric means.

The overall geometric mean value for the time series is depicted as a horizontal, thin, dashed line.

Values reported below the limits of detection (LOD) is substituted using the 'robust' method suggested by Helsel & Hirsch (1995) p 362, assuming a lognormal distribution within a year. N.B. a minimum of three values above the LOD is required for this substitution; years with fewer results above the LOD are not represented in the figures. The LOD is marked by a shaded area

The trend is presented by one or two regression lines (plotted if $p < 0.05$, two-sided regression analysis); one for the whole time period and one for the last ten years (if the time series is longer than ten years). Ten years is often too short a period to statistically detect a trend unless it is of considerable magnitude. Nevertheless the ten-year regression line will indicate a possible change in the direction of a trend. Furthermore, the residual variance around the line compared to the residual variance for the entire period will indicate if the sensitivity have increased as a result of e.g. an improved sampling technique or that problems in the chemical analysis have disappeared. In this version of report, only one regression line for the entire period is presented.

The log-linear regression lines fitted through the geometric mean concentrations follow smooth exponential functions.

Each plot has a header with a letter for the investigated county, BD = Norrbotten, Z = Jämtland, U = Västmanland, P = Älvsborg, F = Jönköping and G = Kronoberg county. Below the header of each plot the results from several statistical calculations are reported:

n(tot)= The first line reports the total number of analyses included together with the number of years (**n(yrs)**=). Note that values below the limit of detection are included in this number.

m = The overall geometric mean value together with its 95% confidence interval is reported on the second line of the plot (N.B. d.f.= n of years - 1).

slope = reports the slope, expressed as the annual change together with its 95% confidence interval.

SD(lr) = reports the square root of the residual variance around the regression line, as a measure of between-year variation, together with the *lowest detectable change* in the current time series with a power of 80%, one-sided test, $\alpha=0.05$. The last figure on this line is the estimated *number of years* required to detect an annual change of 5% with a power of 80%, one-sided test, $\alpha=0.05$.

power = reports the power to detect a log-linear trend in the time series (Nicholson & Fryer, 1991). The first figure represents the power to detect an annual change of 5% with the number of years in the current time series. The second figure is the power estimated as if the slope were 5% a year and the number of years were ten. The third figure is the *lowest detectable change* for a ten-year period with the current between year variation at a power of 80%.

y(04) = reports the concentration estimated from the regression line for the last year together with a 95% confidence interval, e.g. $y(04)=2.51(1.92,3.27)$ is the estimated concentration of year 2004 where the residual variance around the regression line is used to calculate the confidence interval. Provided that the regression line is relevant to describe the trend, the residual variance might be more appropriate than the within-year variance in this respect.

r² = reports the coefficient of determination (r^2) together with a p-value for a two-sided test (H_0 : slope = 0) i.e. a significant value is interpreted as a true change, provided that the assumptions of the regression analysis is fulfilled.

Note. In most cases the y-axis representing the concentrations are different for liver and kidney tissue.

Summary

Significantly increasing trends of magnesium (0.41% a year, liver), manganese (0.74% a year, liver), molybdenum (both liver; 2.6% a year and kidney; 3.8% a year) and zinc (0.47% a year, liver) were shown for the time series from Grimsö. 25 years of analyses are now available. Significantly decreasing trends of iron (-0.81% a year in 1980-2004, kidney; -3.8% a year in liver and -2.5% in kidney in 1995-2004), lead (both liver; -5.8% a year and kidney; -6.2% a year) and vanadium (-3.2% a year, kidney), were shown for the time series from Grimsö, although many single concentrations of vanadium fell below the limit of detection.

Since only 7-9 years of analysis are yet available for the six other areas, it is not likely to find any significant trends in the time series unless relatively large systematic changes have occurred but some significant trends have actually been detected. The same thing is also true for geographical differences where only relatively large regional differences can be detected with the material yet available.

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Calcium Ca

Temporal variation

Significant negative trends in liver tissue from Jämtland (Z), Jönköping (F) and Kronoberg county (G) were detected (annual decrease 5.9%, $p < 0.032$, 3.3%, $p < 0.010$ and 6.4 %, $p < 0.001$, respectively). Significant negative trends were also detected in kidney tissue from Norrbotten (BD) and Kronoberg county (G) (annual decrease 2.7%, $p < 0.069$ and 3.5%, $p < 0.009$, respectively).

No significant change in calcium concentrations was found in liver and kidney tissue, neither for the period 1980-2004 nor for the period 1995-2004 in the Grimsö area. The number of years required to detect an annual change of 5% was 10 years for liver tissue and 7 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 4 and 3% in liver and kidney respectively, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time-series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 7 and 12 years for liver tissue and between 7 and 9 years for kidney tissues. In general, time series of ten years are likely to detect an annual change between 2 and 6%.

The overall geometric mean value of calcium in liver and kidney of moose from Grimsö was 54.0 and 73.1 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

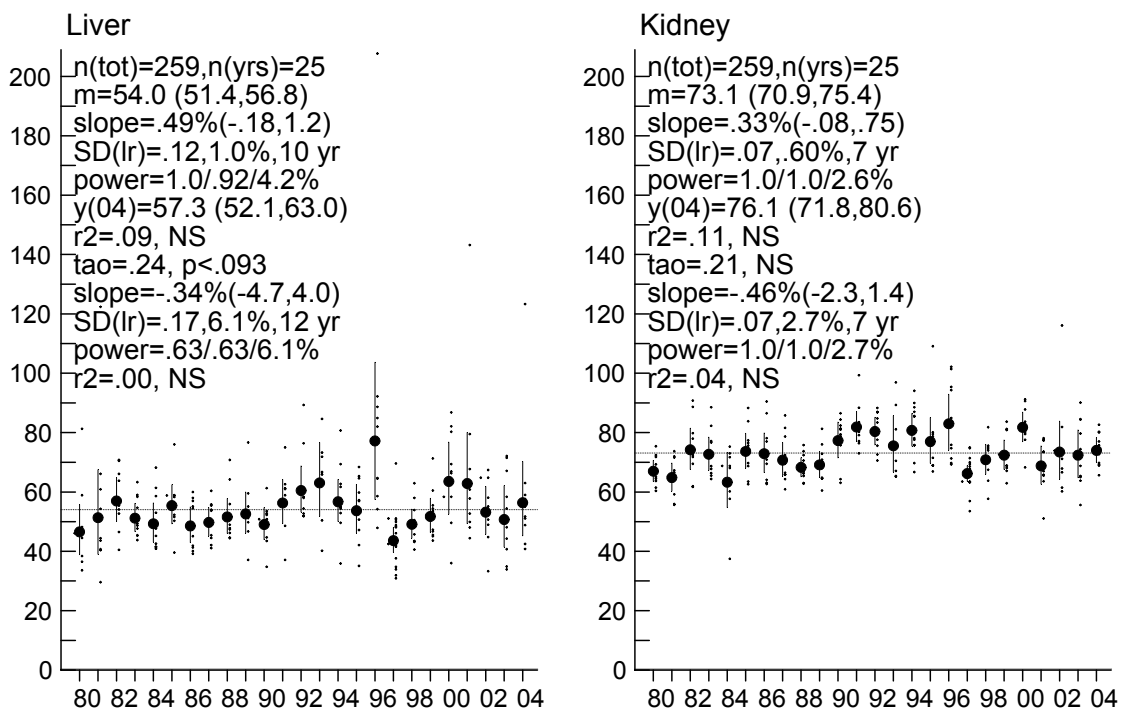
Spatial variation

No significant differences in concentrations between the various counties were detected.

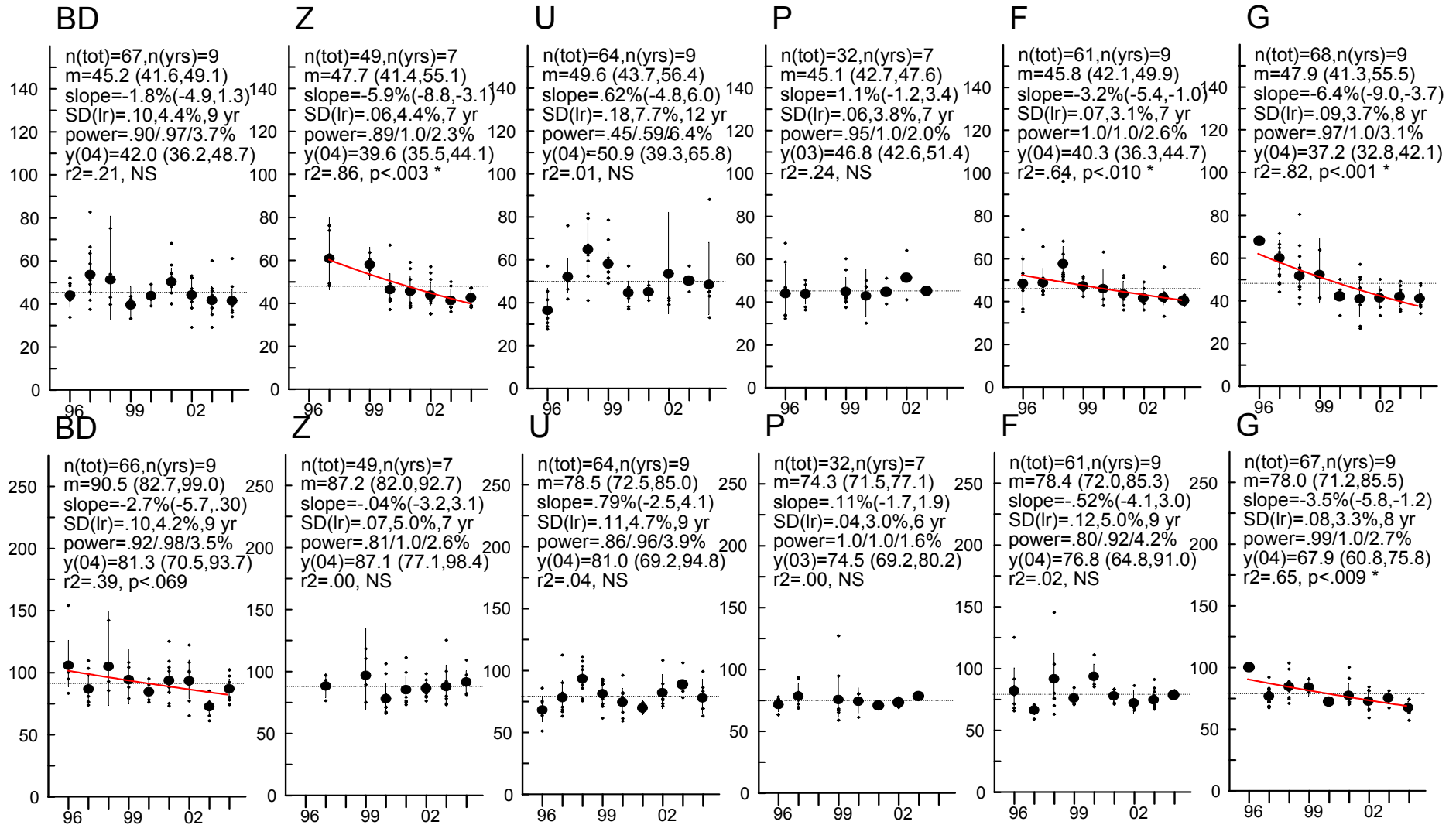
Differences between analysed tissues

The calcium concentrations in kidney tissue were somewhat higher, about 1.4 times, compared to the concentrations found in liver for the 25 years time series from Grimsö. For the shorter time series the number of years are sometimes not sufficient for a statistical significant difference but there is a general pattern of higher calcium concentrations in kidney tissue.

Calcium, $\mu\text{g/g}$ fresh w., moose from Grimsö



Calcium, ug/g fresh w., moose liver (above) / kidney (below)



Cadmium Cd

Temporal variation

A significant increase of cadmium concentrations in liver and kidney tissue from Västmanland county (U) was detected (annual increase 5.5%, $p < 0.032$, and 4.6%, $p < 0.059$, respectively). A significant negative trend was detected for cadmium concentrations in kidney tissue from Jämtland county (Z) (annual decrease 7.2%, $p < 0.059$).

No significant change in cadmium concentrations was found in liver and kidney tissue, neither for the period 1980-2004 nor for the period 1995-2004 in the Grimsö area. The number of years required to detect an annual change of 5% was 14 years for liver tissue and 16 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 9 and 11% in liver and kidney respectively, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 11 and 23 years for liver tissue and between 11 and 23 years for kidney tissue. Time series of ten years are likely to detect an annual change between 6 and 20%.

The overall geometric mean value of cadmium in liver and kidney of moose from Grimsö was 0.283 and 0.899 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

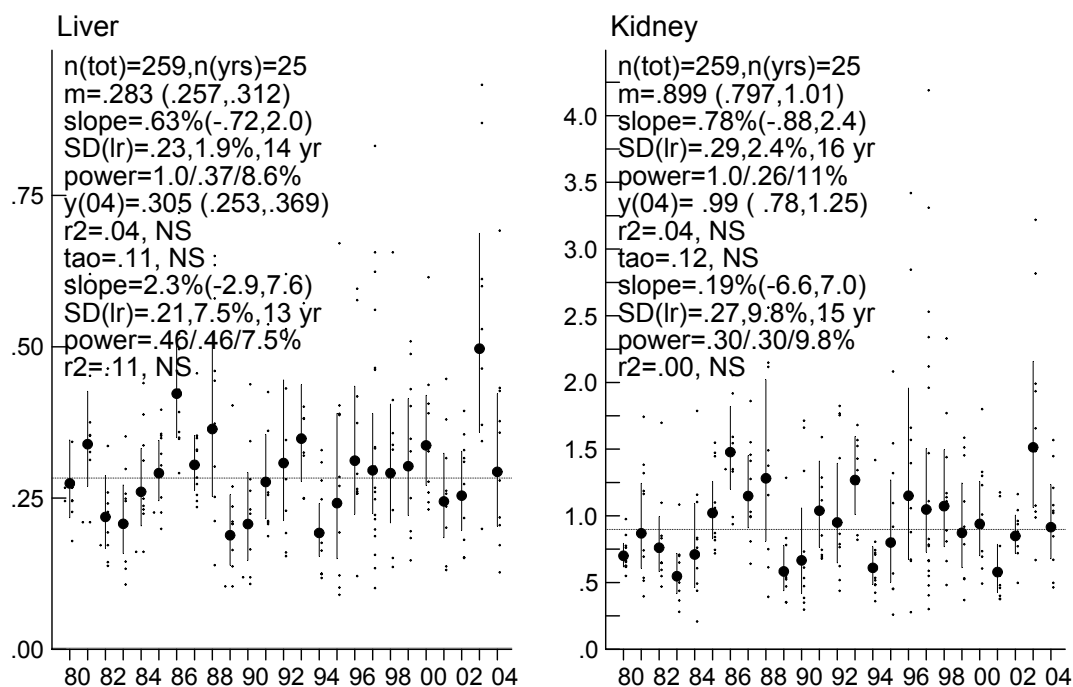
Spatial variation

No significant differences in concentrations between the various counties were detected.

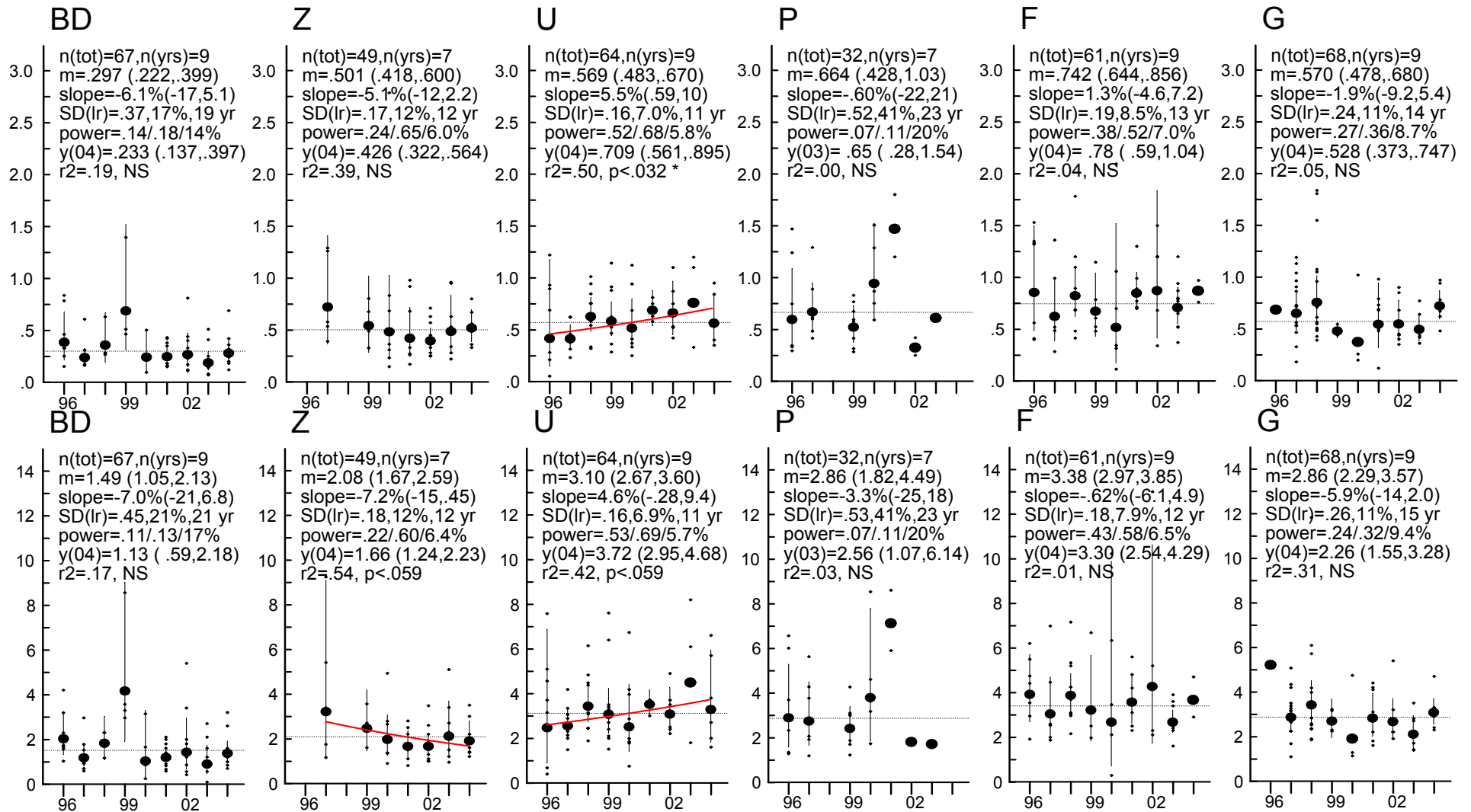
Differences between analysed tissues

The cadmium concentrations in kidney tissue were significantly higher, on average 3 to 5 times, compared to the concentrations found in liver.

Cadmium, $\mu\text{g/g}$ fresh w., moose from Grimsö



Cadmium, ug/g fresh w., moose liver (above) / kidney (below)



Cobalt Co

Temporal variation

A significant negative trend of cobalt concentrations was detected in kidney tissue from Kronoberg county (G) (annual decrease 5.5%, $p < 0.087$).

No significant change in cobalt concentrations was found in liver and kidney tissue for the period 1980-2004, but for the period 1995-2004 (-4.2% per year, $p < 0.008$) in liver tissue from the Grimsö area. The number of years required to detect an annual change of 5% was 11 years for liver tissue and 9 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 5 and 4% in liver and kidney respectively, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 10 and 16 years for liver tissue and between 11 and 19 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 5 and 14%.

The overall geometric mean value of cobalt in liver and kidney of moose from Grimsö was 0.075 and 0.035 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

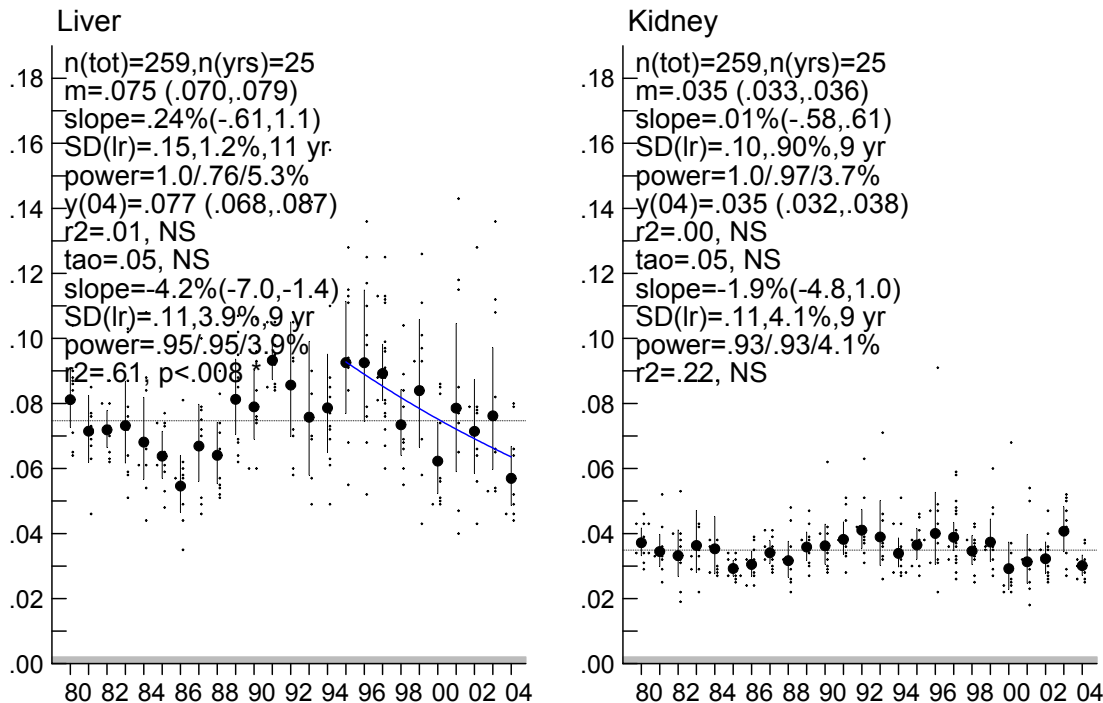
Spatial variation

No significant differences in concentrations between the various counties were detected.

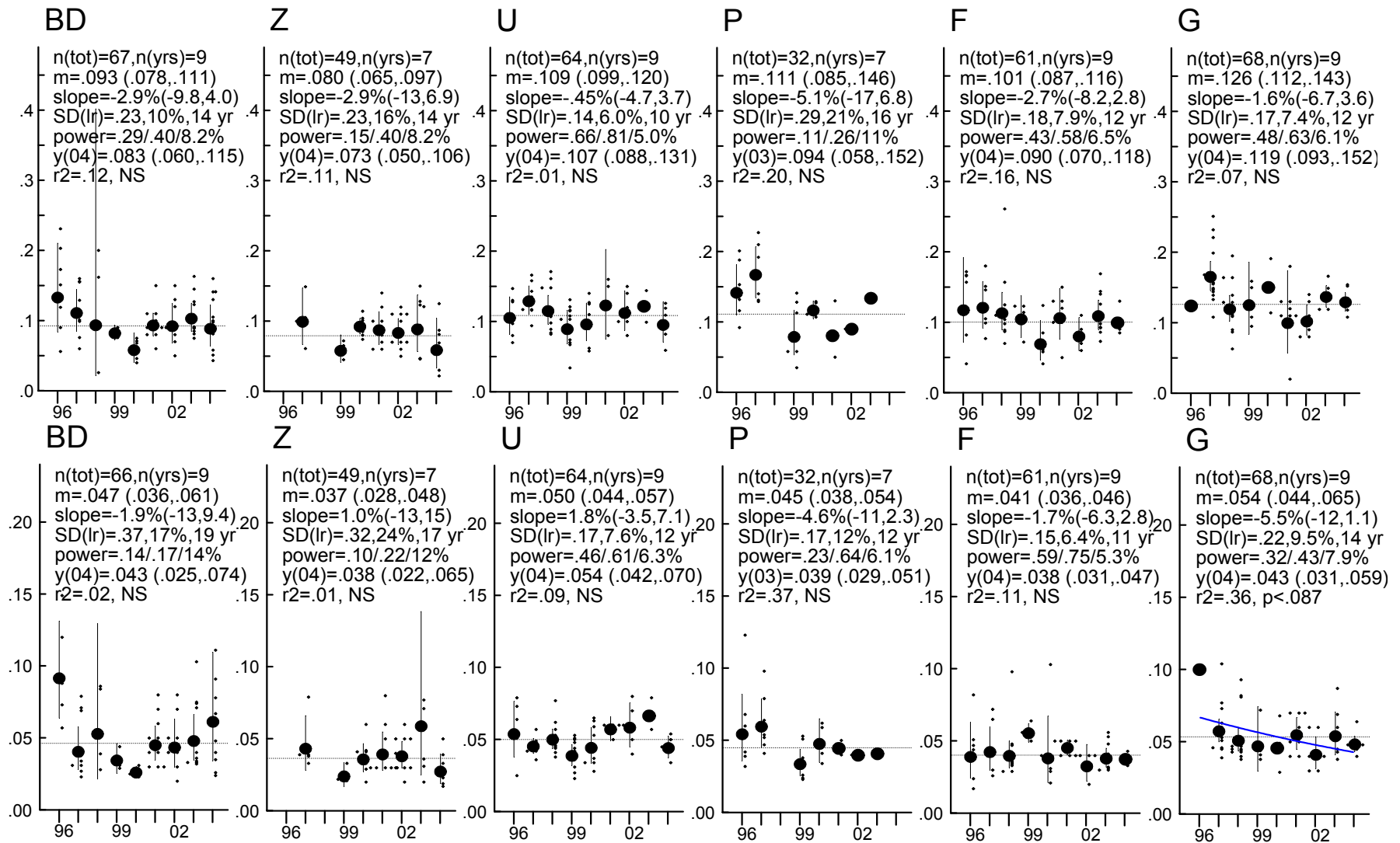
Differences between analysed tissues

The cobalt concentrations in liver tissue were generally significantly higher, on average 2 to 2.5 times, compared to the concentrations found in kidney tissue.

Cobalt, $\mu\text{g/g}$ fresh w., moose from Grimsö



Cobalt, ug/g fresh w., moose liver (above) / kidney (below)



Chromium Cr

Chromium is an element extremely sensitive for contamination that may interfere with natural levels in the samples.

Temporal variation

In the time series from Grimsö, the concentration in about 9% of the samples of both liver and kidney fall below the limit of detection. This affects the power of the analysis, but information is still sufficient to justify continued analysis.

No significant change in chromium concentrations was found in liver and kidney tissue, neither for the period 1980-2004 nor for the period 1995-2004 in the Grimsö area. However, a decreasing trend was detected for chromium in kidney of moose from Jämtland county (Z) (annual decrease 31%, $p < 0.050$).

The number of years required to detect an annual change of 5% was 23 and 24 years for liver and kidney tissue, respectively from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of 20 and 22% for liver and kidney tissues, respectively, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 18 and 26 years for liver tissue and between 16 and 24 years for kidney tissue. Time series of ten years are likely to detect an annual change between 11 and 25 %.

The overall geometric mean value of chromium in liver and kidney of moose from Grimsö was 0.013 and 0.011 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

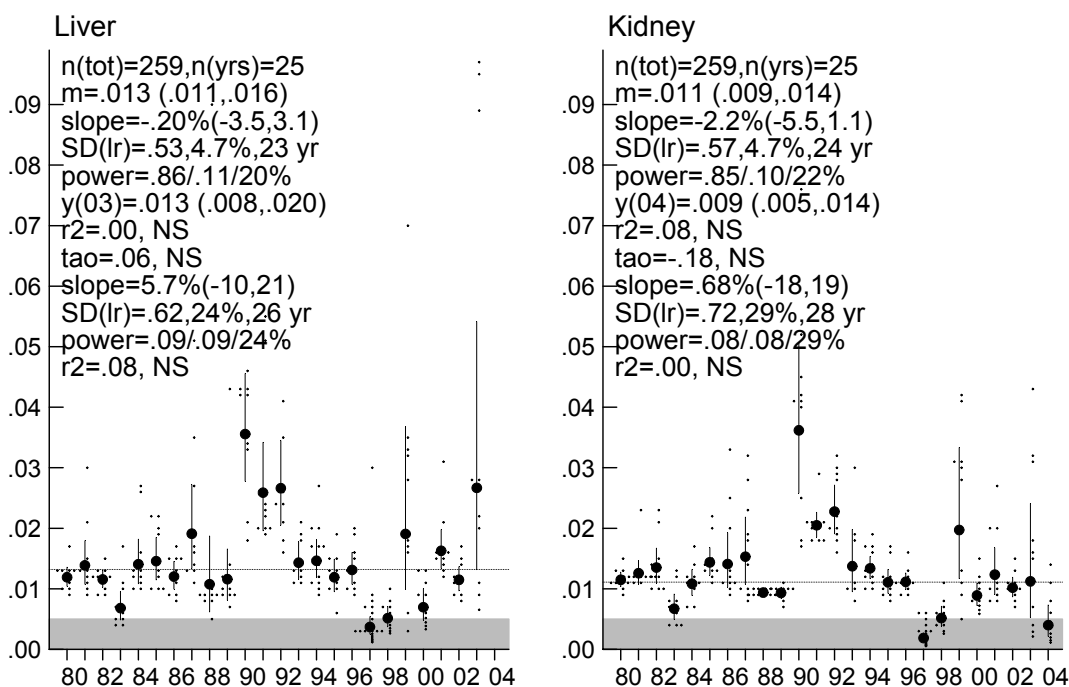
Spatial variation

No significant differences in concentrations between the various counties were detected.

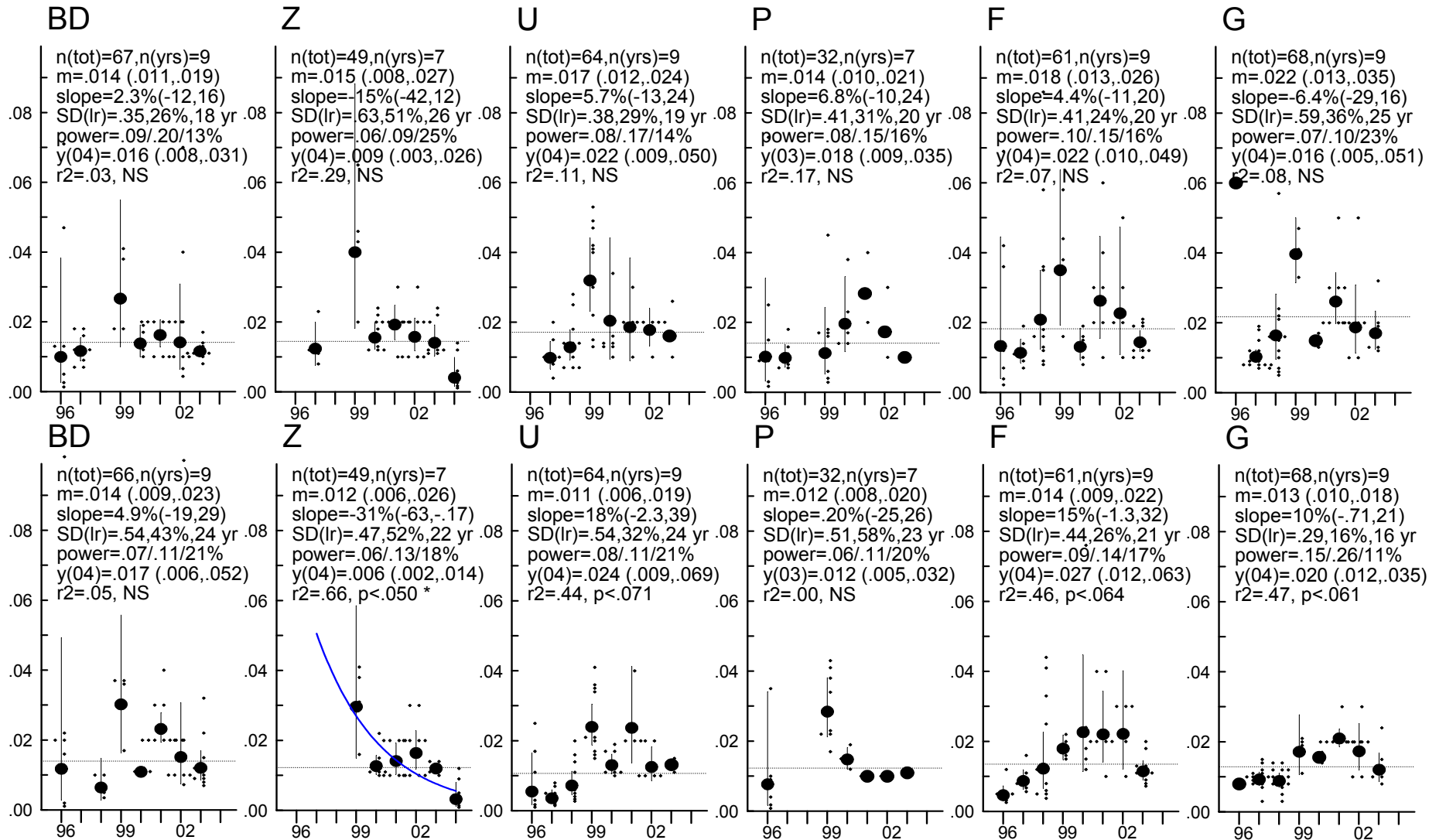
Differences between analysed tissues

No significant differences in concentrations between liver and kidney were shown.

Chromium, ug/g fresh w., moose from Grimso



Chromium, ug/g fresh w., moose liver (above) / kidney (below)



Copper Cu

Temporal variation

A significant log-linear increase was detected for copper concentrations in liver of moose from Jönköping county (F) (annual increase 5.3%, $p < 0.066$) as well as a decrease in kidney of moose from Norrbotten county (BD) and Kronoberg county (G) (annual decrease 1.1%, $p < 0.054$ and 3.2%, $p < 0.091$, respectively).

No significant change in copper concentrations was found in liver and kidney tissue, neither for the period 1980-2004 nor for the period 1995-2004 in the Grimsö area. The number of years required to detect an annual change of 5% was 14 years for liver tissue and 9 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 9 and 4 %, respectively for liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 12 and 31 years for liver tissue and between 5 and 10 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 7 and 33 % in liver and 1 and 5% in kidney.

The overall geometric mean value of copper in liver and kidney of moose from Grimsö was 26.8 and 3.02 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

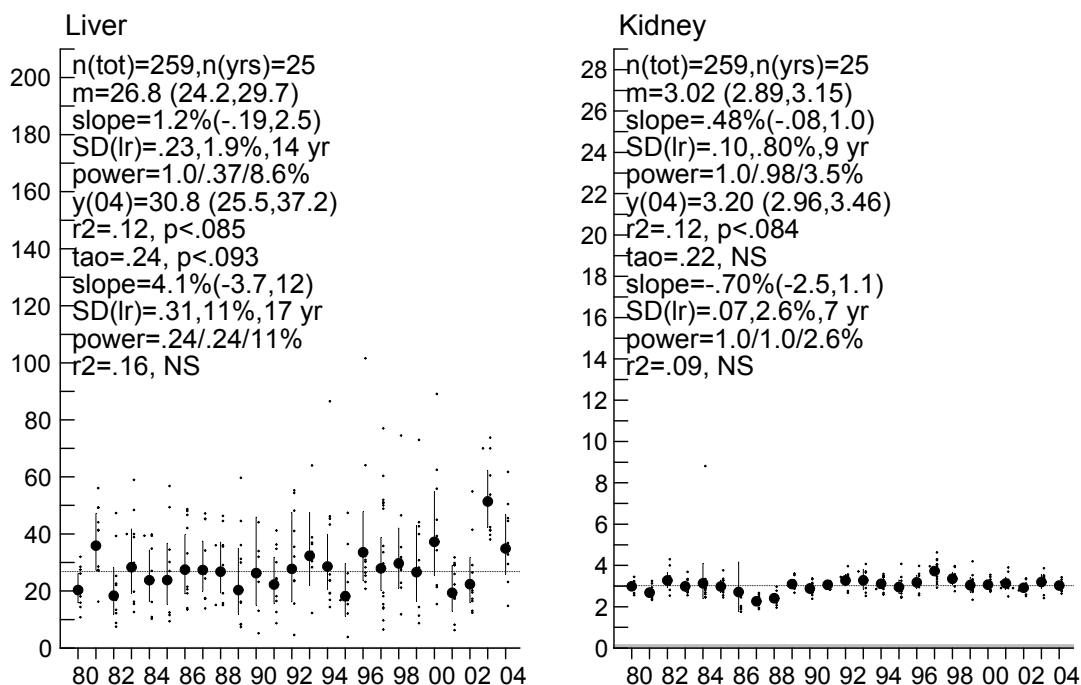
Spatial variation

No significant differences in concentrations between the various counties were detected.

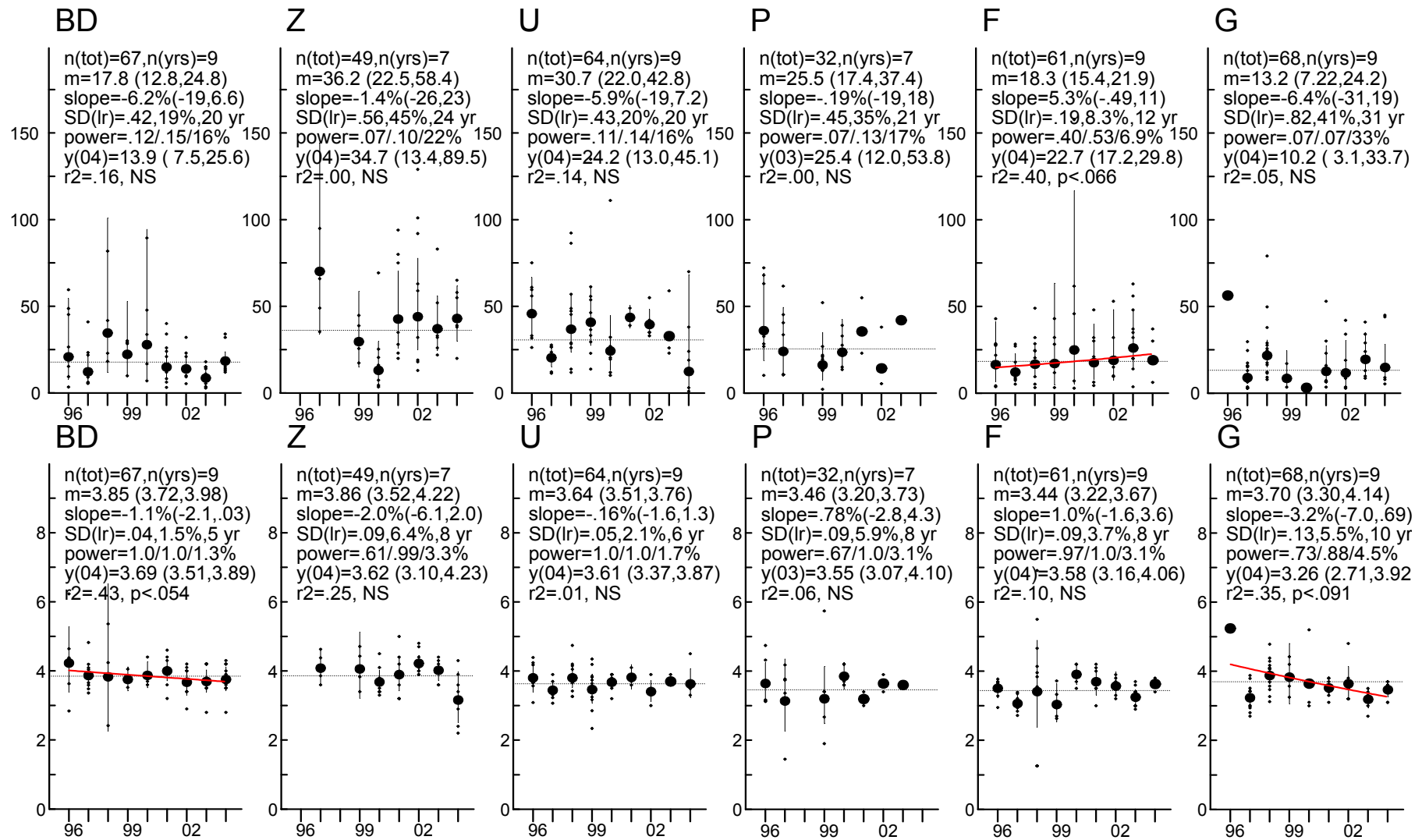
Differences between analysed tissues

The copper concentration in liver is significantly higher compared to kidney, between 3 – 9 times.

Copper, $\mu\text{g/g}$ fresh w., moose from Grimsö



Copper, ug/g fresh w., moose liver (above) / kidney (below)



Iron Fe

Temporal variation

A significant log-linear decreasing trend was detected for iron in kidney from Grimsö both for the period 1980-2004 (-0.81% a year, $p < 0.012$) and the period 1995-2004 (-2.5% a year, $p < 0.038$). A significant decrease was also shown for iron concentrations in liver tissue during the period 1995-2004 (-3.8% a year, $p < 0.006$). Significant log-linear decreases were detected for iron concentrations in liver of moose from Västmanland county (U), Jönköping county (F) and Kronoberg county (G) (4.1% a year, $p < 0.069$, 6.4%, $p < 0.046$ and 3.9%, $p < 0.012$, respectively).

The number of years required to detect an annual change of 5% was 12 years for liver tissue and 9 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 7 and 4% respectively for liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 8 and 13 years for liver tissue and between 8 and 17 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 3 and 8% in liver and 3 and 12% in kidney.

The overall geometric mean value of iron in liver and kidney of moose from Grimsö was 87.3 and 42.3 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

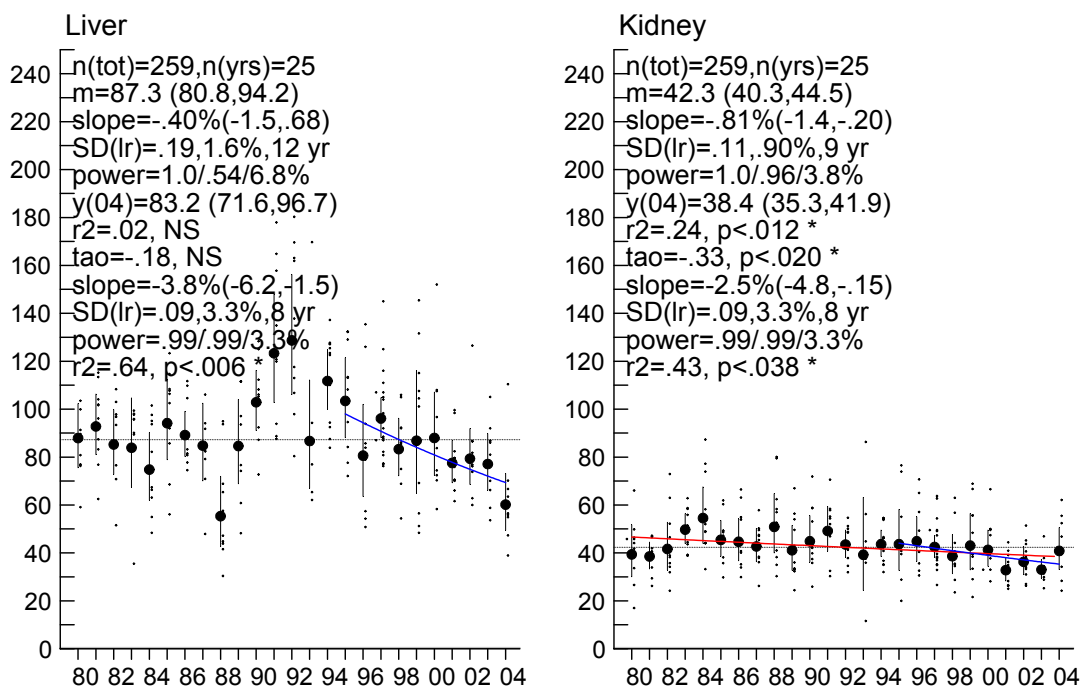
Spatial variation

No significant differences in concentrations between the various counties were detected.

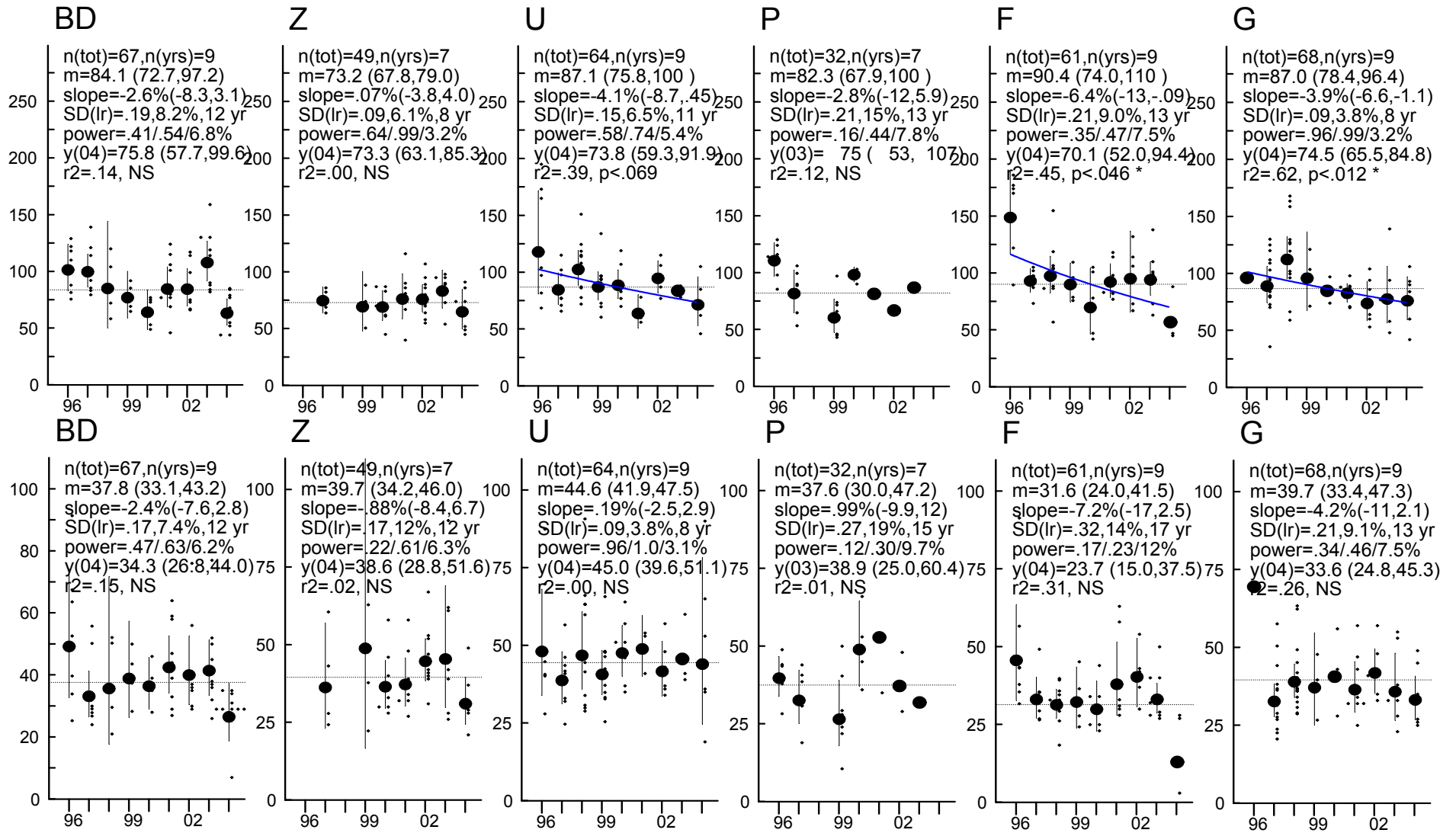
Differences between analysed tissues

The iron concentration in liver is significantly higher compared to kidney, between 2 and 3 times.

Iron, $\mu\text{g/g}$ fresh w., moose from Grimsö



Iron, ug/g fresh w., moose liver (above) / kidney (below)



Magnesium Mg

Temporal variation

A significant positive time trend was detected for magnesium concentrations in liver from Grimsö (annual increase 0.41 %, $p < 0.019$) for the whole period 1980-2004. Significant negative trends were detected for magnesium concentrations in liver tissue from Västmanland county (U) (annual decrease 2.1%, $p < 0.003$) as well as in kidney from Kronoberg county (G) (annual decrease 2.3 %, $p < 0.026$).

The number of years required to detect an annual change of 5% was 7 years for both liver and kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 2% in both liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 5 and 9 years for liver tissue and between 6 and 7 years for kidney tissue. Time series of ten years are likely to detect an annual change approximately between 1 and 4 % in liver and about 2 % in kidney.

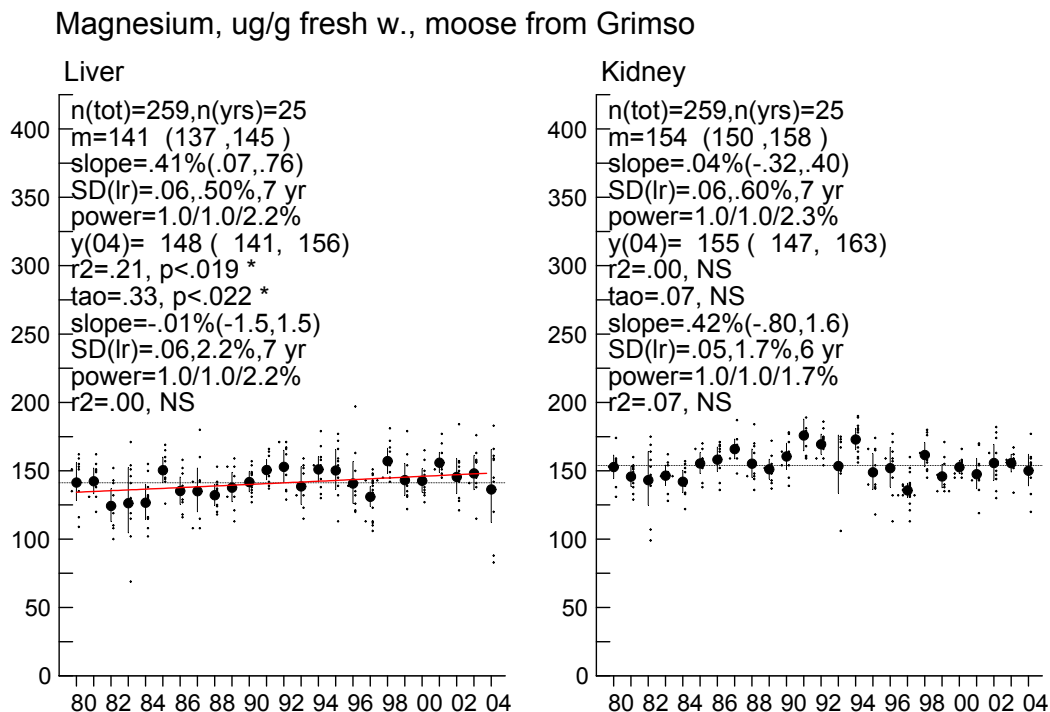
The overall geometric mean value of magnesium in liver and kidney of moose from Grimsö was 141 and 154 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

Spatial variation

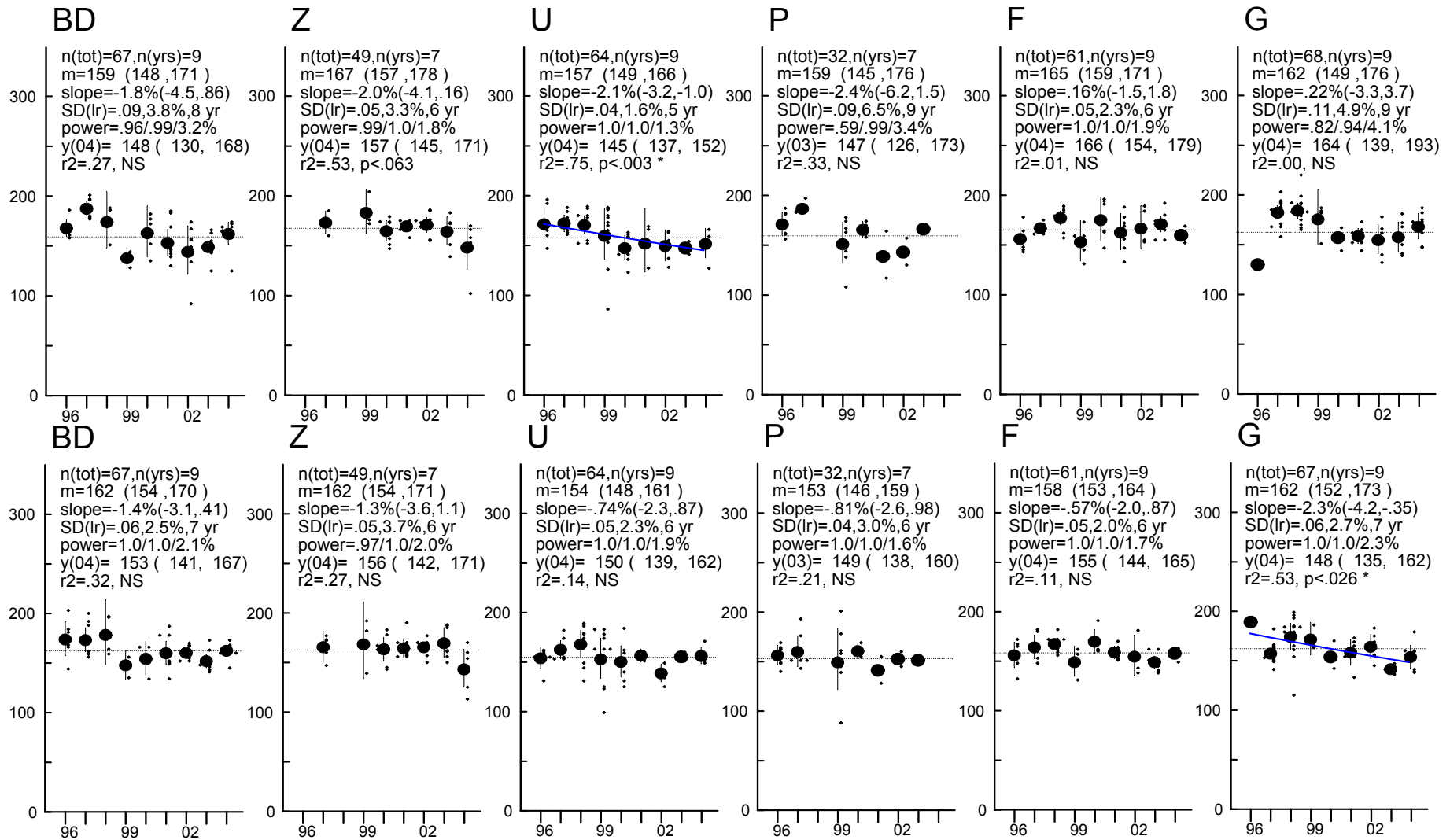
No significant differences in concentrations between the various counties were detected.

Differences between analysed tissues

No significant differences in concentrations between liver and kidney were shown.



Magnesium, ug/g fresh w., moose liver (above) / kidney (below)



Manganese Mn

Temporal variation

A significant positive time trend was detected for manganese concentrations in liver from Grimsö (annual increase 0.74%, $p < 0.007$) for the period 1980-2004. A significant decreasing trend was found for manganese in liver of moose from Kronoberg county (G), (annual decrease 2.8 %, $p < 0.028$).

The number of years required to detect an annual change of 5% was 8 years both for liver and kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 3% in both liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 7 and 12 years for liver tissue and between 9 and 15 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 3 % and 7 % in liver and between 3% and 9 % in kidney.

The overall geometric mean value of manganese in liver and kidney of moose from Grimsö was 3.64 and 4.53 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

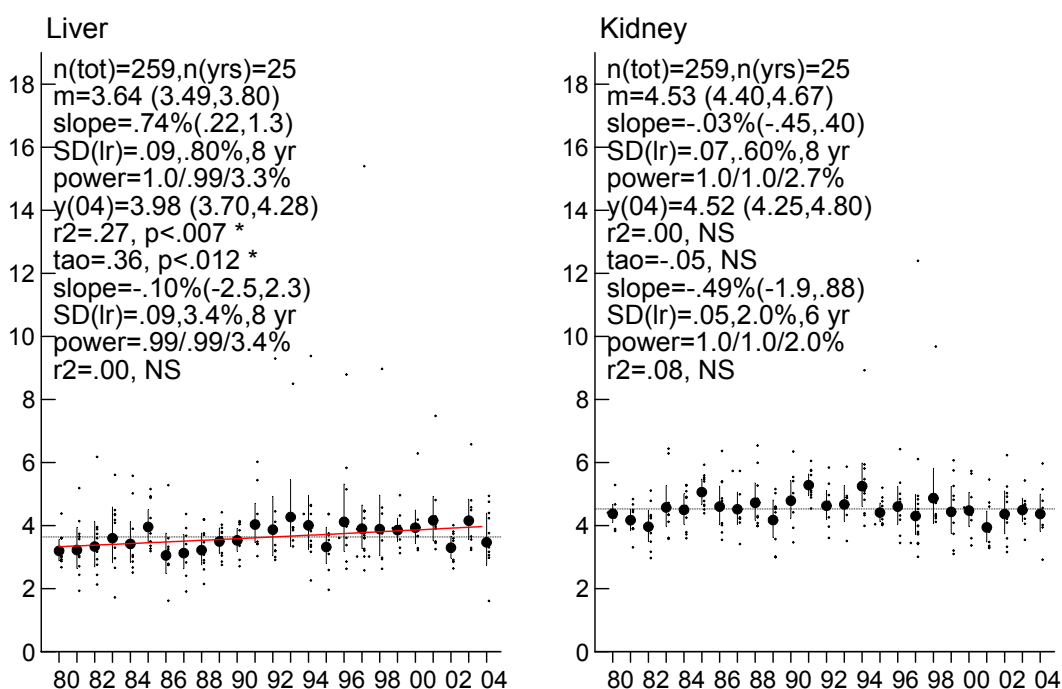
Spatial variation

No significant differences in concentrations between the various counties were detected.

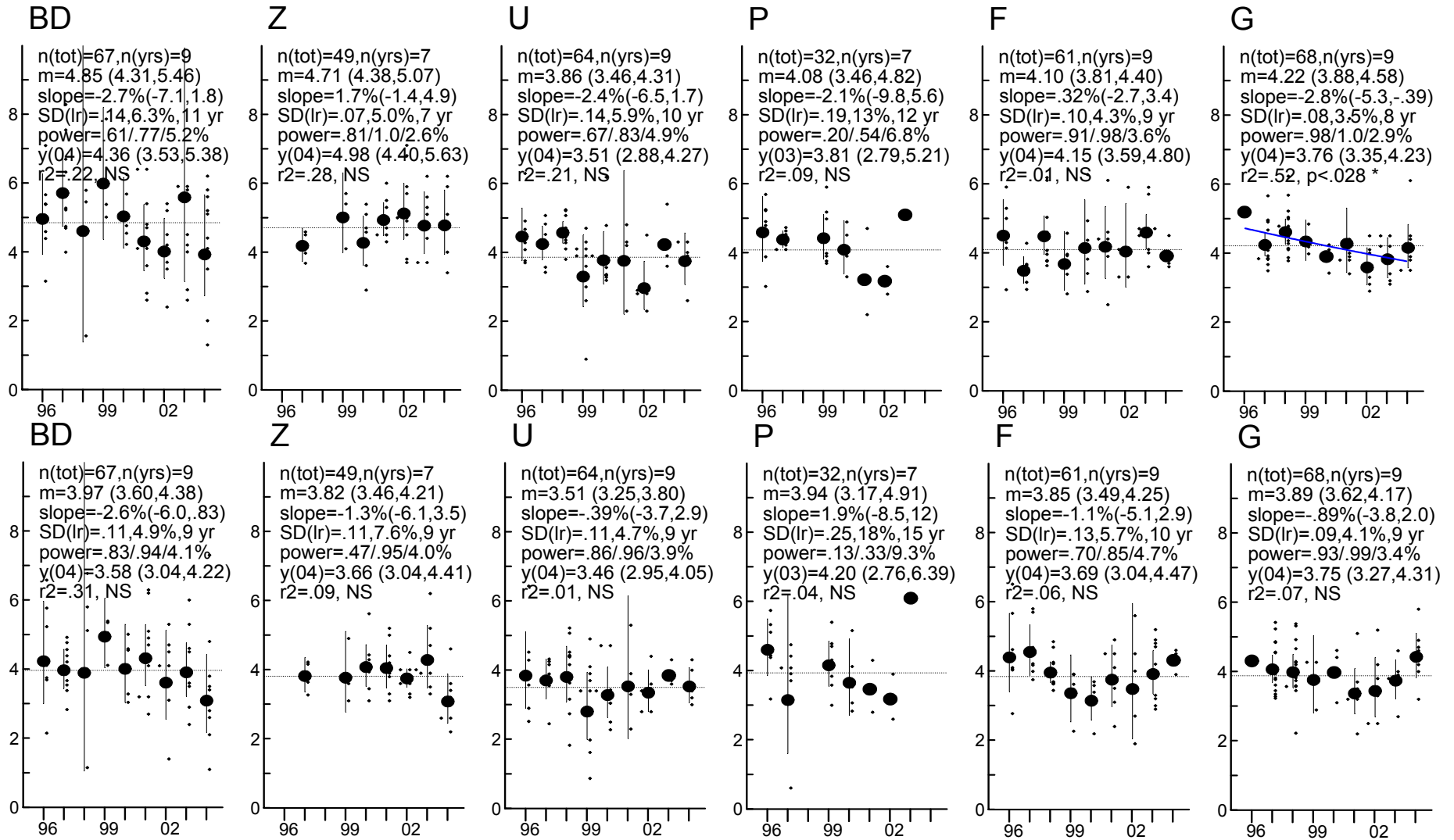
Differences between analysed tissues

There are differences between the analysed tissues but they are small and not consistent between the regions

Manganese, $\mu\text{g/g}$ fresh w., moose from Grimsö



Manganese, ug/g fresh w., moose liver (above) / kidney (below)



Molybdenum Mo

Temporal variation

A significant positive time trend was detected for molybdenum concentrations in both liver and kidney tissue from Grimsö during the period 1980-2004 (annual increase 2.6%, $p < 0.001$, liver; 3.8%, $p < 0.001$, kidney). A significant positive trend was also shown for concentrations in kidney of moose from Grimsö during the period 1995-2004 (annual increase 4.8%, $p < 0.039$).

The number of years required to detect an annual change of 5% was 11 years for liver tissue and 12 years for kidney tissue of moose from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 6% in both liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years are yet available the number of years required detecting an annual change of 5% varied between 8 to 19 years for liver tissue and between 10 and 18 years for kidney tissue. These time series are likely to detect an annual change of between approximately 3 and 15 % in liver and 5 and 13 % in kidney.

The overall geometric mean value of molybdenum in liver and kidney of moose from Grimsö was 0.733 and 0.254 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

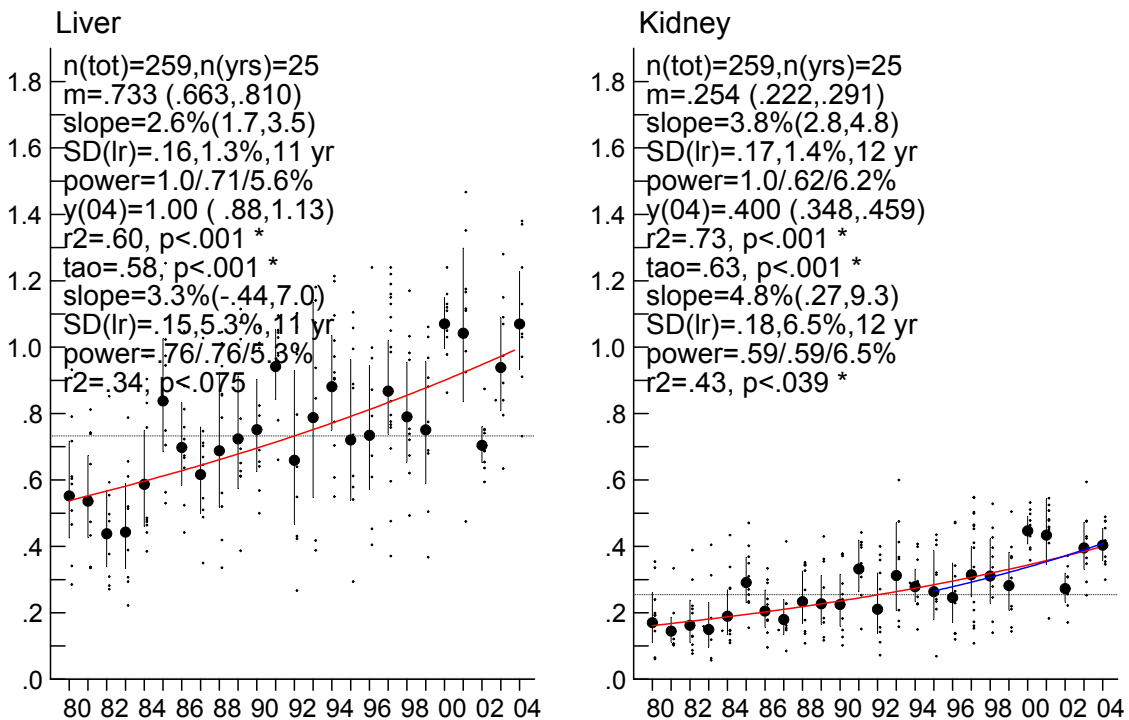
Spatial variation

No significant differences in concentrations between the various counties were detected.

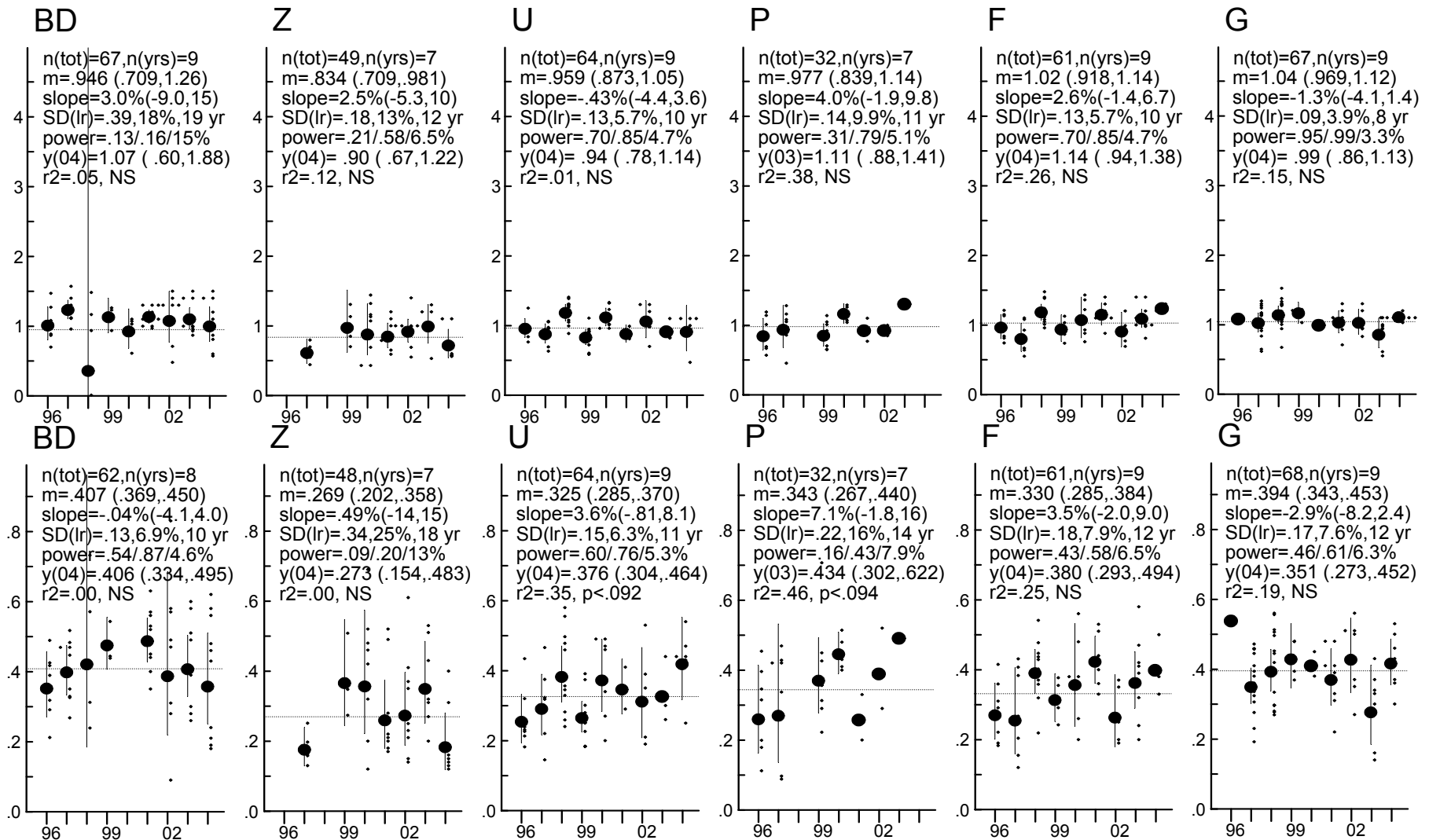
Differences between analysed tissues

The molybdenum concentration in liver is significantly higher compared to kidney, between 2 and 3 times.

Molybdenum, $\mu\text{g/g}$ fresh w., moose from Grimso



Molybdenum, ug/g fresh w., moose liver (above) / kidney (below)



Nickel Ni

The nickel concentrations measured, in liver and kidney tissue of moose, are close to the limit of detection. Hence, the analytical precision plays a larger role for the unexplained between-year variation in nickel compared to most of the other investigated trace metals.

Temporal variation

In the analysis of nickel in liver samples from Grimsö, 26 % of the samples fell below the limit of detection. For kidney the number was 17 %. This, of course, makes it very hard to detect any time trends. Approximately the same situation can be seen in the shorter time series.

The number of years required detecting an annual change of 5% were 30 years for liver and 38 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. The time series are likely to detect an annual change of about 31% and 49% respectively in liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, too many values are below the LOD to make statistics in liver tissue meaningful. The number of years required detecting an annual change of 5% varied between 12 and 17 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 6 and 12 % in kidney.

The overall geometric mean value of nickel in liver and kidney of moose from Grimsö was 0.009 and 0.017 µg/g (fresh weight), respectively for the period 1980-2004.

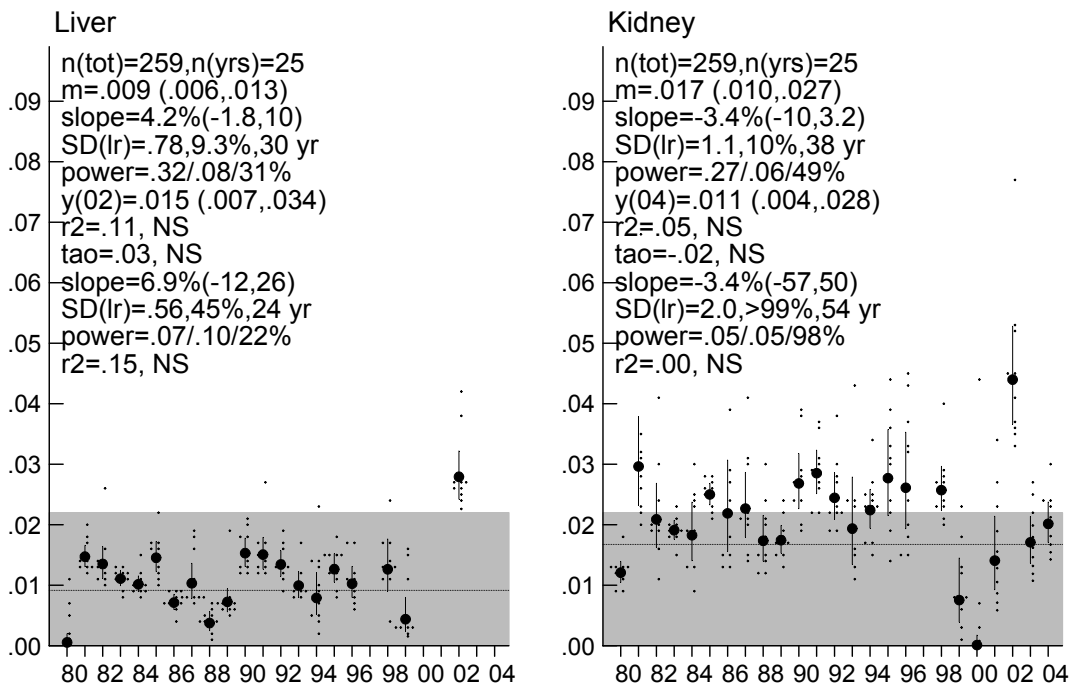
Spatial variation

No significant differences in concentrations between the various counties were detected.

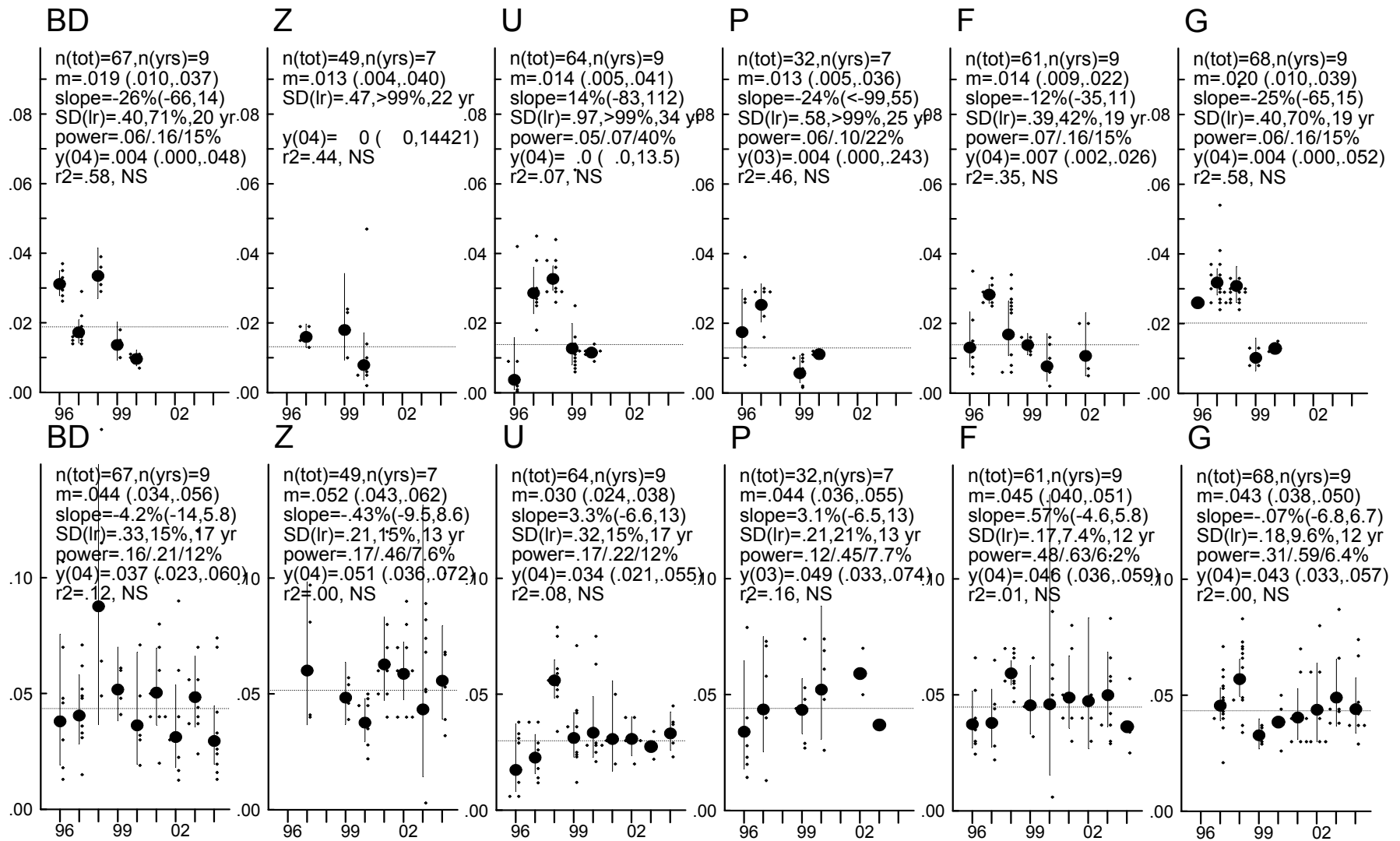
Differences between analysed tissues

The nickel concentration in kidney is significantly higher compared to liver for most of the time series, 1 to 3 times.

Nickel, ug/g fresh w., moose from Grimso



Nickel, ug/g fresh w., moose liver (above) / kidney (below)



Lead Pb

Temporal variation

For lead, only a very small number of samples fall below the limit of detection, and the analysis is most likely not affected at all.

Significant negative time trends were detected for lead concentrations in both liver and kidney tissue from Grimsö (annual decrease -5.8 %, $p < 0.001$, liver; -6.2 %, $p < 0.001$, kidney) for the period 1980-2004. Significant negative trends have also been detected in the shorter time series for liver from Västmanland county (U) (annual decrease 21 %, $p < 0.012$) from Jönköping county (F) (annual decrease 12 %, $p < 0.047$) and from Kronoberg county (G) (annual decrease 23%, $p < 0.009$).

The number of years required to detect an annual change of 5% was 16 years for liver and 14 years for kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 10 and 8 %, respectively in liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years yet are available, the number of years required detecting an annual change of 5% varied between 19 and 25 years for liver tissue and between 19 and 22 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 14 and 23 % in liver and 10 and 18 % in kidney.

The overall geometric mean value of lead in liver and kidney of moose from Grimsö was 0.040 and 0.060 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

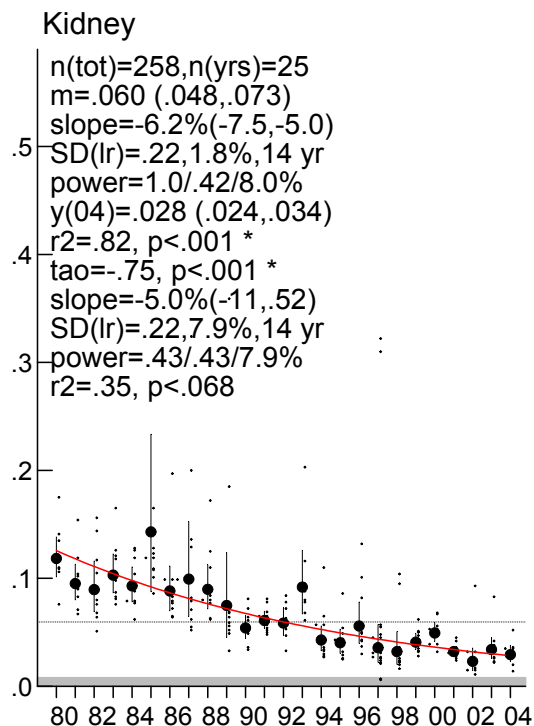
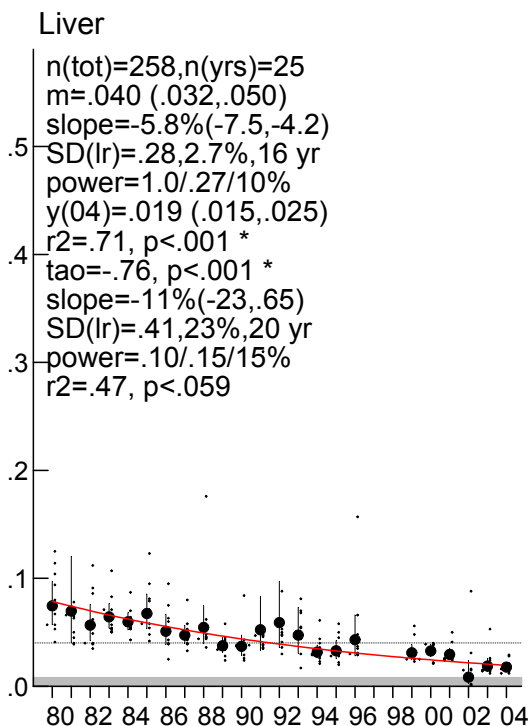
Spatial variation

No significant differences in concentrations between the various counties were detected.

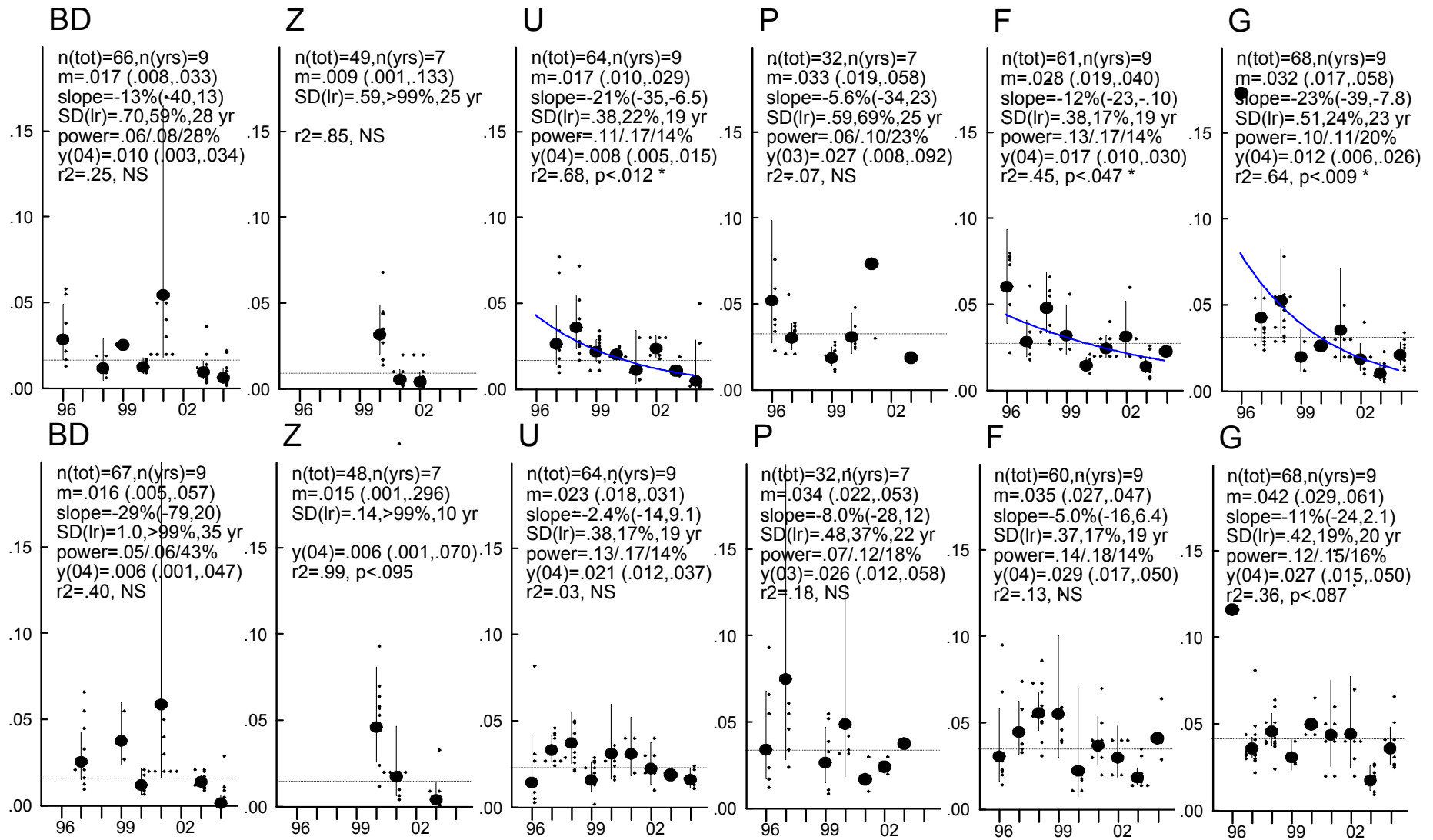
Differences between analysed tissues

There are no significant differences between the concentrations in liver and kidney.

Lead, $\mu\text{g/g}$ fresh w., moose from Grimso



Lead, ug/g fresh w., moose liver (above) / kidney (below)



Vanadium V

The vanadium concentrations measured in moose liver and kidney tissue are close to the LOD. Hence, the analytical precision plays a larger role for the unexplained between-year variation compared to most of the other investigated trace metals.

Temporal variation

For vanadium, 27 % and 22 % of the samples of liver and kidney respectively from Grimsö fell below the detection limit. This will affect the possibility to detect time trends but will probably not have a massive impact on the statistical analysis.

A significant decreasing log-linear trend was detected for vanadium concentrations in kidney from Grimsö (annual decrease -3.2 %, $p < 0.007$).

The number of years required to detect an annual change of 5% was 33 years for liver tissue and 18 years for kidney tissue in samples from Grimsö. These time series are likely to detect an annual change of about 38 and 13% respectively in liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series, too many values are below the LOD to make statistics meaningful.

The overall geometric mean value of vanadium in liver and kidney of moose from Grimsö was 0.003 and 0.003 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

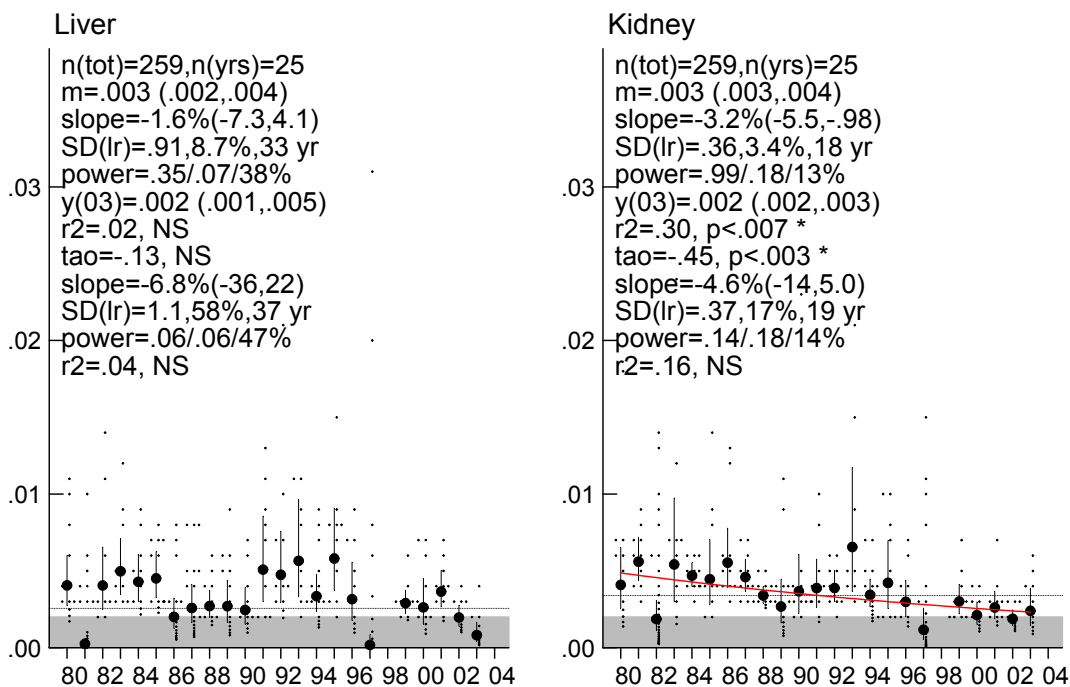
Spatial variation

No significant differences in concentrations between the various counties were detected.

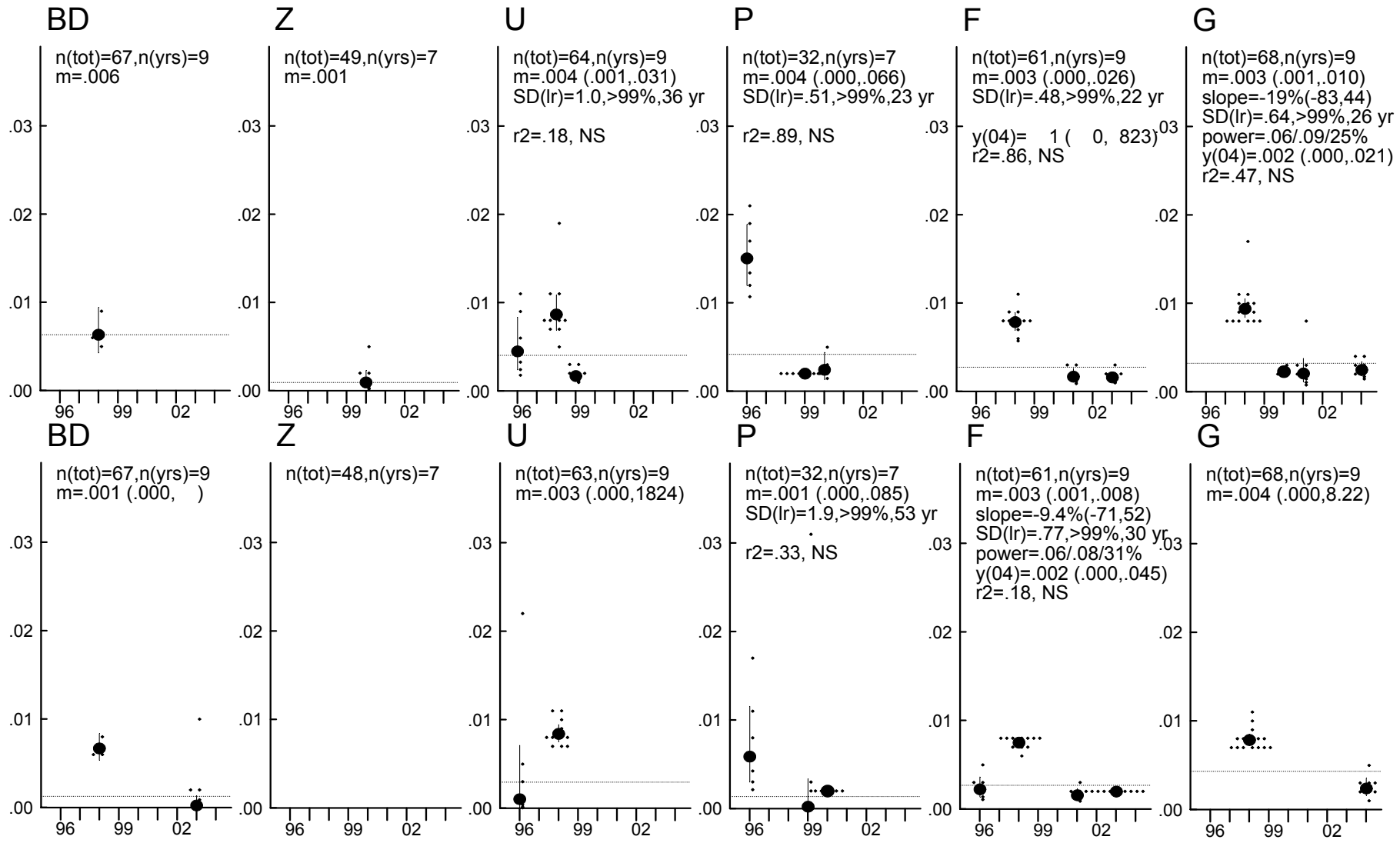
Differences between analysed tissues

There are no significant differences between the concentrations in liver and kidney.

Vanadium, $\mu\text{g/g}$ fresh w., moose from Grimsö



Vanadium, ug/g fresh w., moose liver (above) / kidney (below)



Zinc Zn

Temporal variation

A significant positive trend was detected for zinc concentrations in liver tissue from Grimsö (annual increase 0.47%, $p < 0.031$). A significant negative trend was found in kidney from Norrbotten county (BD) (annual decrease 2.9 %, $p < 0.019$).

The number of years required to detect an annual change of 5% was 8 and 6 years respectively for liver and kidney tissue in samples from Grimsö where 25 years of analyses are available. These time series are likely to detect an annual change of about 3 % and 2 % respectively in liver and kidney tissue, provided that the power is fixed to 80% and the significance level is set to 5%. For the shorter time series where only seven to nine years are yet available the number of years required detecting an annual change of 5% varied between 9 and 16 years for liver tissue and between 7 and 10 years for kidney tissue. Time series of ten years are likely to detect an annual change of between 4 to 11 % in liver and 3 and 5 % in kidney.

The overall geometric mean value of zinc in liver and kidney of moose from Grimsö was 22.5 and 23.6 $\mu\text{g/g}$ (fresh weight), respectively for the period 1980-2004.

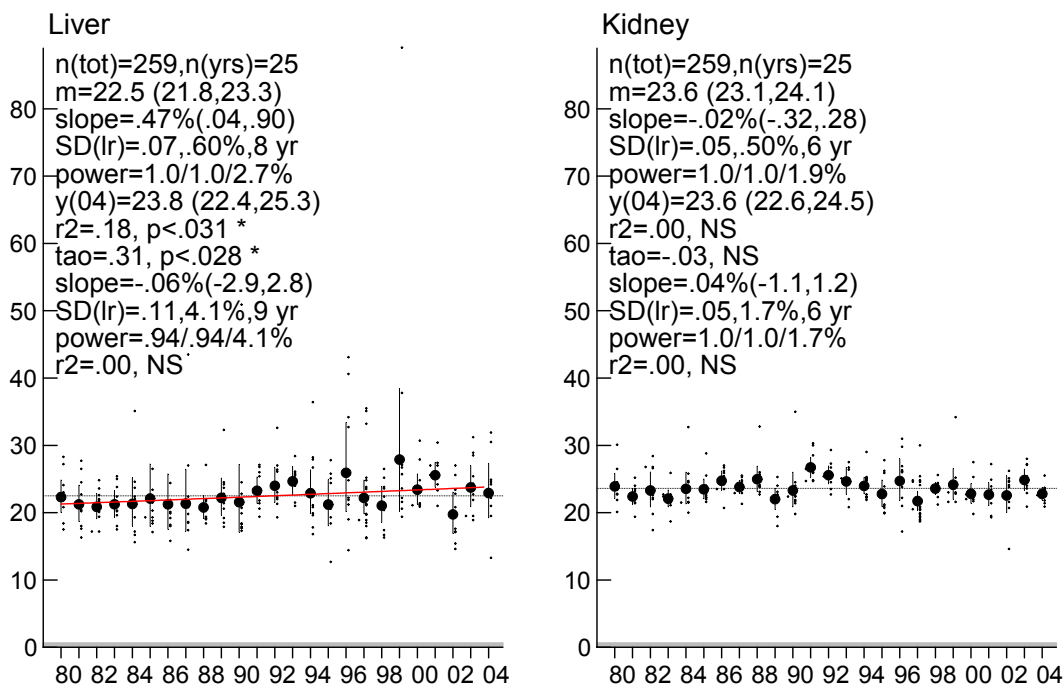
Spatial variation

No significant differences in concentrations between the various counties were detected.

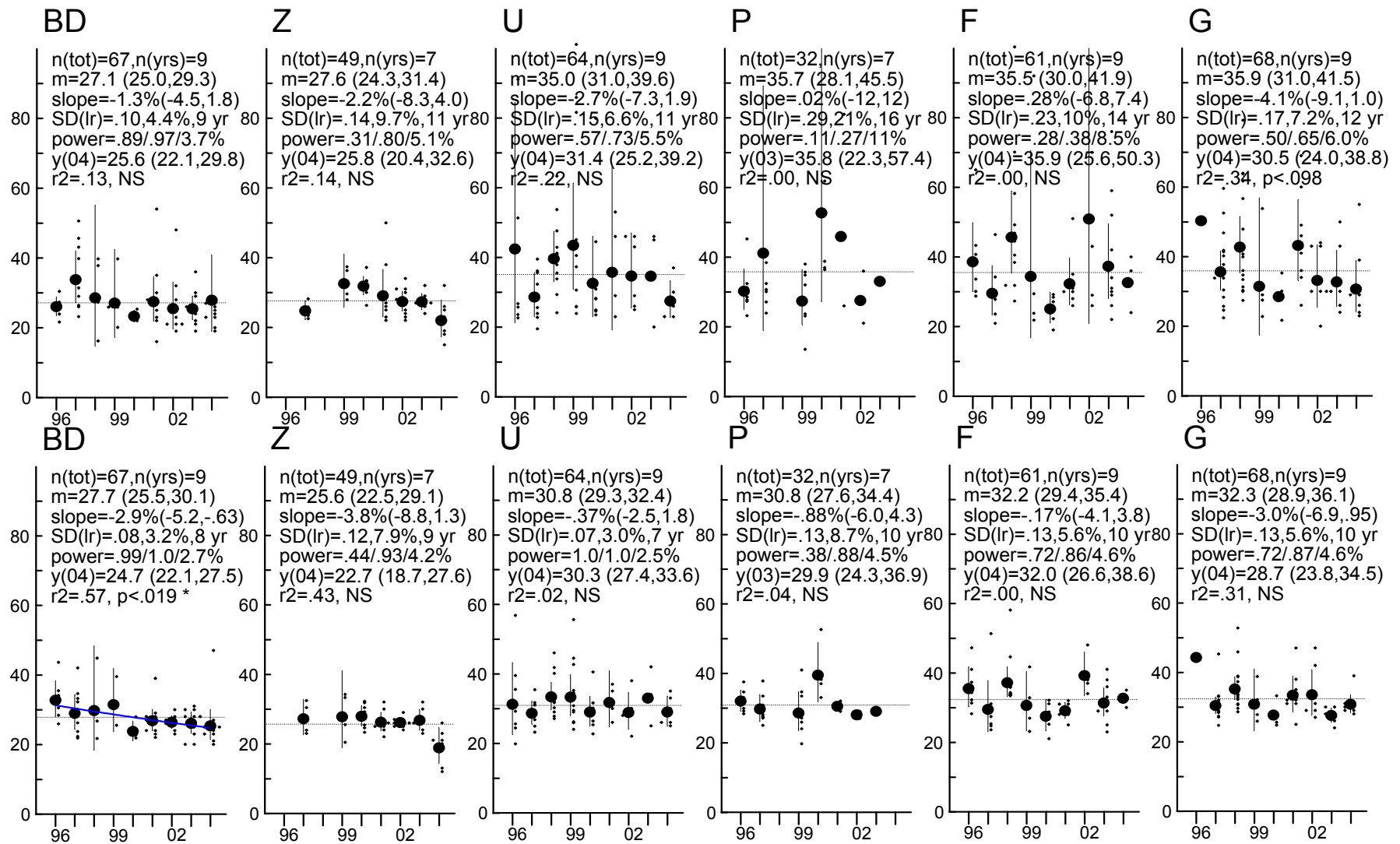
Differences between analysed tissues

There are no significant differences between concentrations in liver and kidney.

Zinc, $\mu\text{g/g}$ fresh w., moose from Grimso



Zinc, ug/g.fresh w., moose liver (above) / kidney (below)



Selenium Se

Temporal variation

For 2002 and 2004, analyses of selenium were only carried out in muscle and liver tissues of moose from Grimsö.

No significant trends were detected in the time series of selenium concentration in muscle and liver.

For the time series where only five to seven years are yet available the number of years required to detect an annual change of 5% varied between 15 to 25 years for liver tissue and between 11 and 17 years for muscle tissue, provided that the power is fixed to 80% and the significance level is set to 5%. Time series of ten years are likely to detect an annual change of between 6 and 23%.

The overall geometric mean value of selenium in muscle and liver of moose from Grimsö was 50.3 and 218 µg/g (fresh weight), respectively for the period 1999-2004.

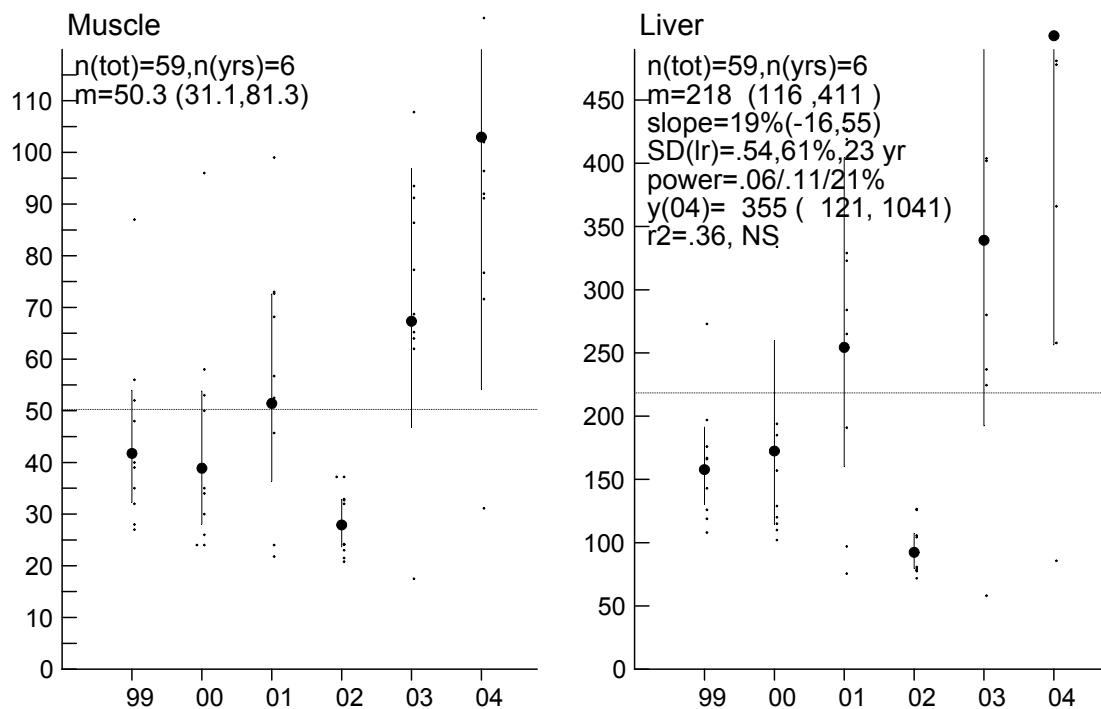
Spatial variation

No significant differences in concentrations between the various counties were detected.

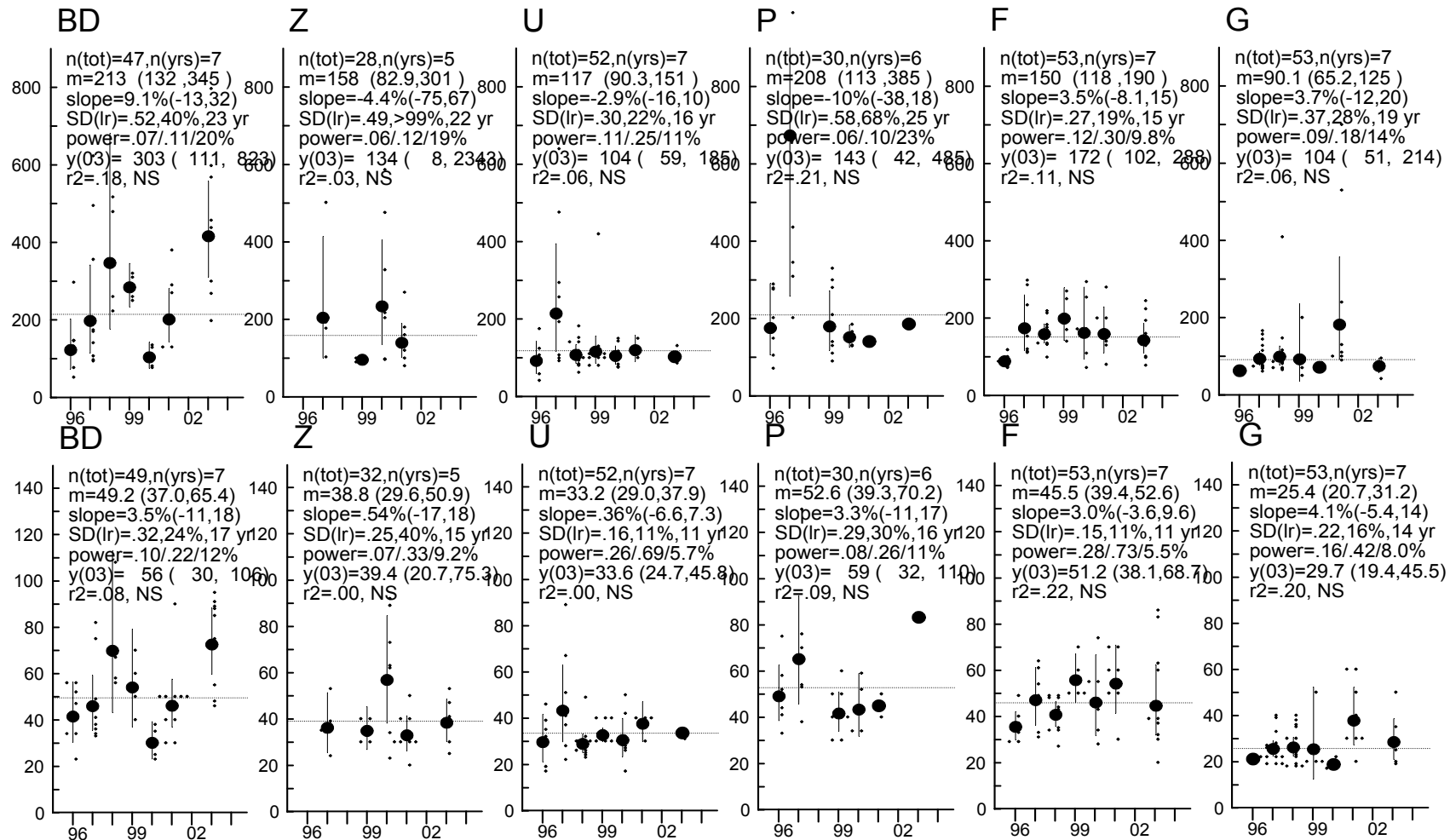
Differences between analysed tissues

There are significant differences between the concentrations of selenium in the analysed tissues. The concentrations are 3-4 times higher in liver than in muscle.

Selenium in muscle and liver, ng/g fresh w., moose from Grimsö



Selenium, ng/g fresh w., moose liver (above) / muscle (below)



Mercury Hg

Temporal variation

For mercury in muscle tissue from Grimsö, 50 % of the samples fall below the limit of detection (LOD). For mercury in liver only 8.5% were lower than LOD. This implies that liver may be a matrix more suited for mercury analyses.

No significant trends were found for mercury in liver. However, in muscle significant positive time trends were detected in four counties (annual increase 10 %, $p < 0.002$, Norrbotten county (BD); annual increase 16 %, $p < 0.019$, Västmanland county (U); annual increase 16 %, $p < 0.037$, Jönköping county (F); annual increase 15 %, $p < 0.001$, Kronoberg county (G)).

For the time series where only seven to nine years are yet available the number of years required to detect an annual change of 5% varied between 13 and 21 years for liver tissue and 9 and 23 years for muscle, provided that the power is fixed to 80% and the significance level is set to 5%. Time series of ten years are likely to detect an annual change of between 8 to 17 % in liver and 4 and 20 % in muscle.

The overall geometric mean value of mercury in muscle and liver of moose from Grimsö was 0.876 and 3.01 $\mu\text{g/g}$ (fresh weight), respectively for the period 1999-2004.

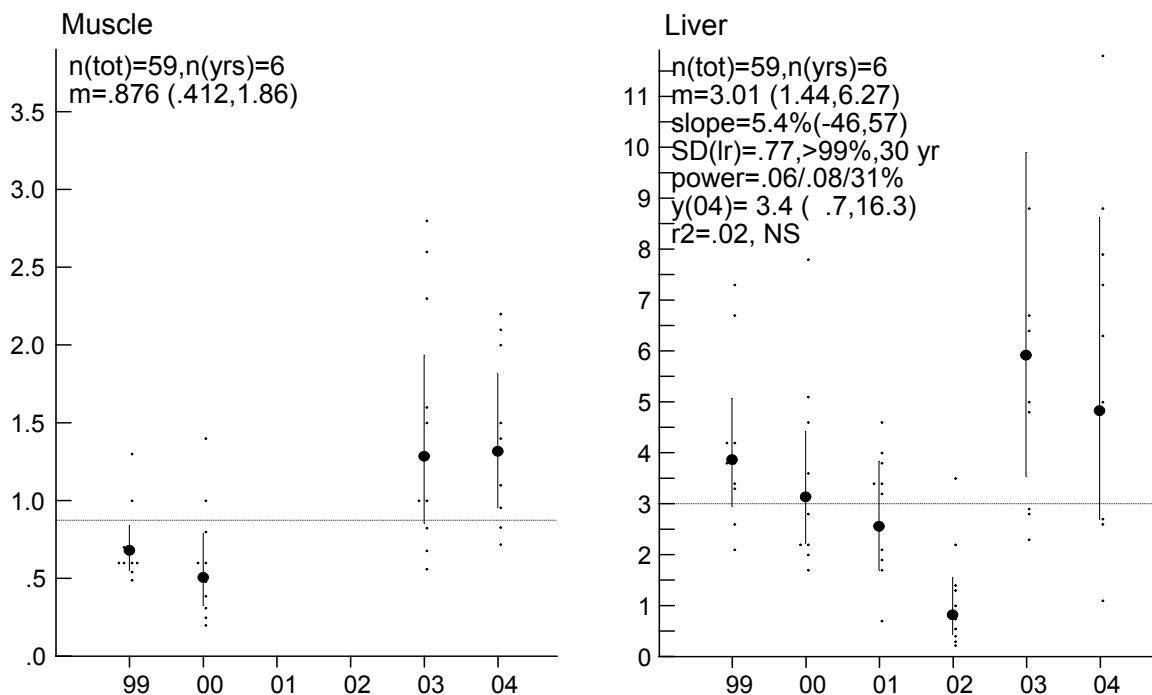
Spatial variation

No significant differences in concentrations between the various counties were detected.

Differences between analysed tissues

The measured concentrations in liver are significantly higher compared to the concentrations measured in muscle, in general 5 to 6 times.

Mercury in muscle and liver, ng/g fresh w., moose from Grimso



Mercury, ng/g fresh w., moose liver (above) / muscle (below)

