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Diplomarbeit

Determination of Correlations between Element Distributions in Bone Samples Measured with High Resolution X-ray Element Imaging

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Physikalische Energie- und Messtechnik

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Abstract

The aim of this thesis was to provide methods of correlation and co-localization to investigate element compositions of human bone sample. The μ-XRF generated images were collected at the ESRF and Diamond synchrotron radiation facilities and depict the distributions of the elements Ar, Ca, Cr, Fe, Co, Ni, Cu, Zn, Gd and W which are given with an approximated image size between 10 μm (ESRF) and 500 μm (Diamond) and a resolution of 50 nm and 500 nm respectively. The whole data set consists of 31 single scan locations on the bone samples, taken from five patients suffering from tumorous or osteoporotic diseases.

The main elements of interest are Ca, Cr, Fe, Ni, Cu, Zn and Gd where the origin is very well known. Ca, Cr, Fe, Ni, Cu and Zn are natural or age-related (due to the environment in the life of the patient) elements of bone tissue. Possible Gd accumulations are explored about used Gd based contrast agents for magnetic resonance imaging investigations during patient treatments.

The investigated methods are based on image correlation and overlap measurements. To obtain a value quantifying the correlation, the Pearson's Correlation Coefficient (linear correlation) and the Spearman's Correlation Coefficients (monotonic correlation) were determined. Furthermore, the Manders Overlap Coefficient was determined as a measure of the overlap between two element distributions.

Furthermore, to get a qualitative overview, correlation graphs and overlap images are presented for better interpretations. Statistical methods were used to show element correlations and co-localizations for the whole data set. Finally, the statistical methods are applied to compare two different patients and extract differences between them for the correlation and overlap coefficients.

Not surprisingly, Ca was found in each pixel for all samples. Between Ca and Zn, a high correlation and high overlap was found. Regarding GD1 (osteoporotic) and GD5 (tumorous), the Ca-Fe relationship for overlap measurements shows less Fe for patient GD1 compared to GD5. Due to the small number of samples (n= 7) for patient GD1 and n= 15 for patient GD5, more data would be required to solidify the results.

Concerning the overlap coefficients M1 and M2, the following element combinations are significant different between patient GD1 and GD5:
-) For GD5, Ca overlaps Fe to 53% more (median), in compare to GD1. Fe overlaps Ca to 100% for GD1 and GD5. The co-localization is significant higher for GD5 in compare to GD1.
-) For GD5, Fe overlaps Ni to 71% less (median), in compare to GD1. The co-localization is significant higher for GD1 in compare to GD5.
-) For GD5, Gd overlaps Fe to 55% more (median), in compare to GD1. The co- localization is significant higher for GD5 in compare to GD1.
-) For GD5, Gd overlaps Ni to 44% less (median), in compare to GD1. The co-localization is significant higher for GD1 in compare to GD5.
-) For GD5, Zn overlaps Fe to 51% more (median), in compare to GD1. The co-localization is significant higher for GD5 in compare to GD1.

Kurzfassung

Das Ziel dieser Arbeit soll Methoden ausfindig machen, damit Korrelationen und Co-Lokalisationen für die Untersuchung von menschlichen Knochen und deren Elementzusammensetzungen gemessen werden können. Die durch μ-XRF erzeugten Bilder wurden am ESRF Synchrotron und am Diamond Synchrotron erzeugt und geben die Elemente Ar, Ca, Cr, Fe, Co, Ni, Cu, Zn, Gd und W wieder. Die Bildgröße befindet sich im Bereich zwischen 10 μm (ESRF) und 500 μm (Diamond), beziehungsweise die Auflösung befindet sich zwischen 50 nm und 500 nm. Der Datensatz umfasst 31 einzelne Messpositionen von fünf verschiedenen Patienten, bei denen eine Tumorerkrankung oder Osteoporose bekannt ist.

Die Elemente auf denen das Hauptaugenmerk liegt und deren Herkunft bekannt ist, sind Ca, Cr, Fe, Ni, Cu, Zn und Gd. Ca, Cr, Fe, Ni, Cu und Zn sind natürliche oder altersbedingte (aufgenommen durch Umwelteinflüsse während der Lebenszeit eines Patienten) Elemente, die möglicherweise im Knochengewebe gefunden werden. Mögliche Gd-Ansammlungen können über verwendete Kontrastmittel, die während einer Magnetresonanzbildgebung verabreicht wurden, erklärt werden.

Die untersuchten Methoden basieren auf Korrelations- und Überlappungsmessungen. Die Korrelationsmessungen bestehen aus folgende Koeffizienten: Pearson's correlation coefficient (lineare Korrelation) und Spearman's correlation coefficient (monotone Korrelation). Der Mander's overlap coefficient wurde verwendet, um Überlappungen zwischen zwei Elementverteilungen zu bestimmen.

Des Weiteren geben Korrelationsgraphen und Überlappungsbilder eine Übersicht und sorgen für eine einfache Interpretationsmöglichkeit. Statistische Methoden machen Korrelationen und Co-Lokalisationen für den gesamten Datensatz sichtbar. Schließlich ermöglichen die statistischen Methoden, Unterschiede in den Korrelationen und Co-Lokalisationen zwischen zwei Patienten sichtbar zu machen.

Wenig überraschend konnte das Element Ca in jedem Bildpunkt für sämtliche Proben gefunden werden. Zwischen Ca und Zn wurde eine hohe Korrelation und eine hohe Überlappung gemessen. In der Betrachtung des Patientenvergleichs zwischen GD1 (Osteoporosepatient) und GD5 (Tumorpatient), zeigte die Ca-Fe-Beziehung für die Überlappungsergebnisse weniger Fe-Vorkommen in GD1 (Osteoporosepatient), verglichen zu GD5 (Tumorpatient). Aufgrund der kleinen Probengröße von n= 7 für Patient GD1 und n= 15 für Patient GD5 sind mehr Daten (Messpositionen und Patienten) notwendig, um die Ergebnisse zu untermauern.

Betreffend der Überlappungskoeffizienten M1 und M2 zeigen die folgenden Elementkombinationen zwischen GD1 und GD5 signifikante Unterschiede auf:
-) Für GD5, Ca überlappt Fe zu 53% mehr (median), verglichen zu GD1. Fe überlappt Ca zu 100% für GD1 und GD5. Die Co-Lokalisation ist signifikant höher für GD5, verglichen zu GD1.
-) Für GD5, Fe überlappt Ni zu 71% weniger (median), verglichen zu GD1. Die Co-Lokalisation ist signifikant höher für GD1 im Vergleich zu GD5.
-) Für GD5, Gd überlappt Fe zu 55% mehr (median), verglichen zu GD1. Die Co-Lokalisation für GD5 ist signifikant höher, verglichen zu GD1.
-) Für GD5, Gd überlappt Ni zu 44% weniger (median) im Vergleich zu GD1. Die Co-Lokalisation ist signifikant höher für GD1, verglichen zu GD5.
-) Für GD5, Zn überlappt Fe zu 51% mehr (median), verglichen zu GD1. Die Co-Lokalisation ist signifikant höher für GD5 im Vergleich zu GD1.

Contents

1 Introduction

The aim of this work is to find suitable assessment methods for image correlation and co-localization to investigate element distributions of bone tissue. The founded methods should help to understand which element correlations and co-localizations can be typically found for a defined patient group where the medical past (diseases) is known.
The μ-XRF generated images for the elements Ar, Ca, Cr, Fe, Co, Ni, Cu, Zn, Gd and W are given with an approximated size between 10μm and 500μm. The whole data set consists of 31 single scan locations. The original use for the generated data set was the investigation of Gd occurrences, corresponding to known magnetic resonance imaging investigations in combination for used Gd based contrast agents during patient treatments.
Normally the data output is taken for interpretations, where single scan locations can be used to compare element distributions. This qualitative view for data evaluation can generate basic statements about the localization and occurrence of main elements and trace elements. To develop the data evaluation, alternative methods are needed. The implemented methods are very well established in the field of biological histology. The proposed methods are based on correlation and overlap measurements. To measure the correlation, the Pearson's Correlation Coefficient (linear correlation) and the Spearman's Correlation Coefficients (monotonic correlation) were taken. The Manders Overlap Coefficient was used to measure the overlap between two element distributions seen in the images. Furthermore, to get a qualitative overview correlation graphs and overlap images are also presented. Statistical methods are used, to show element correlations and co-localizations for the whole data set. Finally, the statistical methods are applied to compare two different patients and extract those differences for the correlation and overlap coefficients.[1]

2 Theoretical Background

The following chapter describes the theoretical background which is needed to understand how the data was generated. Furthermore, the theoretical knowledge for the data evaluation process is also described.

2.1 μ-XRF

X-Ray fluorescence is nowadays a well established analytical method to measure the elemental compositions of solid materials, bulk materials, powdered and liquid specimens and further coating systems. The measurement technique itself is non-destructive and the sample can be reused for further studies. However, the smooth and even surfaces of the sample is desirable. This is achieved by grinding or polishing the sample which makes the method partly destructive.
μ-XRF compared to XRF, provides a sufficient method to investigate the inhomogeneous composition of a specimen and for getting informations about the element distribution in two dimensions and for special specimens even in three dimensions.
To get a sufficient signal intensity, the excitation radiation has to be concentrated to a small area. The availability of X-Ray optics makes it possible to reach the required characteristics of an intense and satisfying signal. The X-Ray optics must be chosen in a prudent way, because of the given beam shape functions and in addition the optics are also influences the spectral distribution of the X-rays.
Furthermore the positioning of the sample must also be observed.[2]

2.2 Interaction Between X-rays and Matter

The principle of the XRF technique is to excite the atoms of the analyzed matter (sample). The sample is exposed to sufficiently energetic X-rays/gamma-rays or charged particles which is shown in Figure 1 (image a) and the photo ionization process (Figure 1 image b) being the desired effect for XRF analysis. In short, this effect removes an inner electron from an atom of interest, frequently an electron from an outer shell takes up the free position. Through the fact of higher binding energies for inner electrons in comparison to outer electrons, the energy difference devolve into X-rays which are different between all the elements and discrete energy transitions.
Image d in Figure 1 shows the electron transfer from the L_{II} shell to the K shell represents the K_{α_2} transition and creates a photon with the energy of the different binding energies between the outer L_{II} shell and the inner K shell. Image c in Figure 1 describes the Auger Emission, which is described in section 2.2.2 in more detail.
[3]

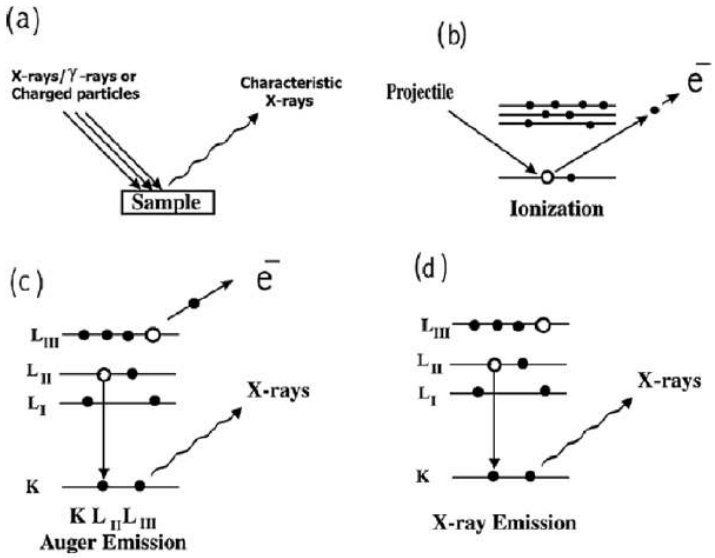

Figure 1: a) How photon emission principle works. b) The ionization process creates a vacancy by kicking out inner shell electron. c) Auger emission process. d) $K_{\alpha II}$ emission process.[3]

At the end, the spectrum should represent the investigated elements. Figure 2 shows an example for the measured energy [keV] distribution in counts per 100 seconds. One can see a peak of nearly 550 counts/ 100 seconds for the $Ca - K_\alpha$ transition, which is the energy transition for the L shell to the K shell. Furthermore, the element Ca shows a lower peak representing the energy transition between the M shell and the L shell (nearly 100 counts/ 100 seconds), which is called $Ca - K_\beta$ transition.

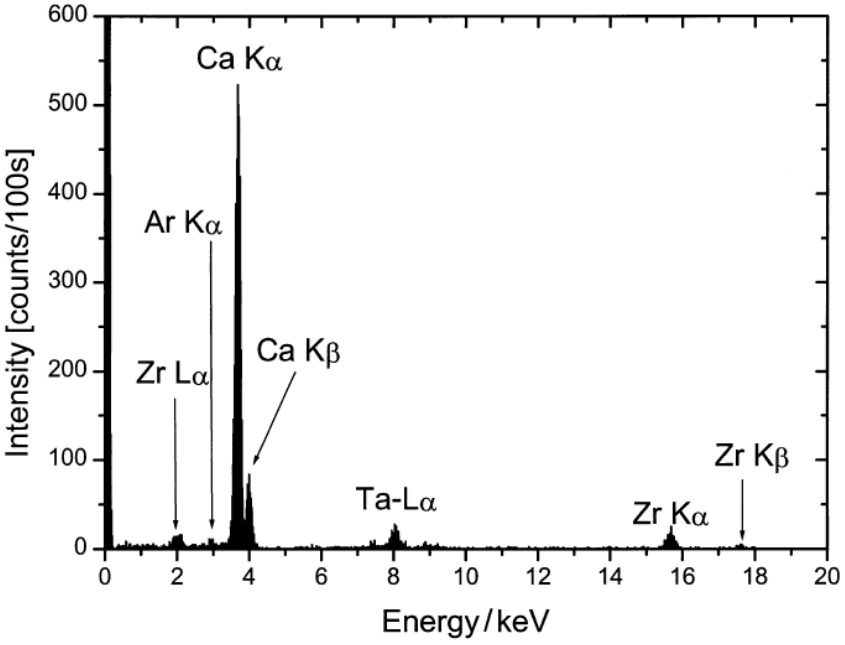

Figure 2: Generic spectrum. X-Axis: Energy [keV] and Y-Axis: Intensity [counts/100 seconds].[4]

Due to the short wavelength (X-rays: 10 pm - 10 nm), in the range of atomic distances in solid and liquid material, X-rays can be used for material investigations. Absorption, scattering, diffraction, refraction and emission form the physical explanations. To show the different phenomena, Figure 3 is given and a general overview of the interactions is described in the following chapters.[3, 2]

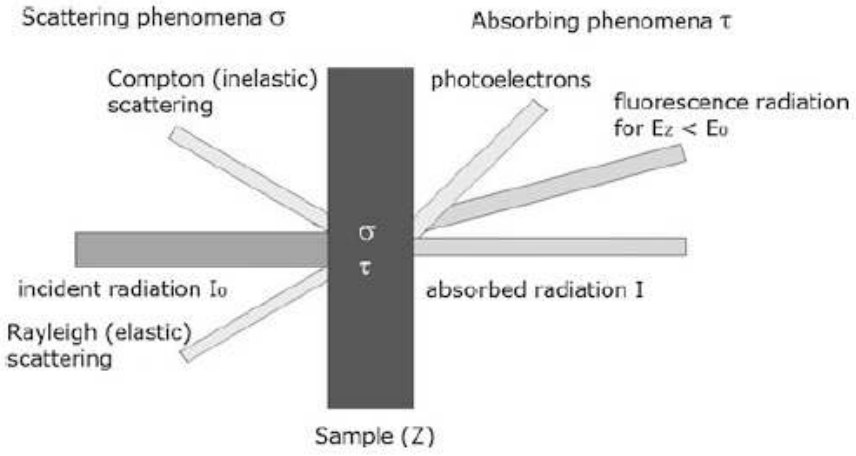

Figure 3: Interaction phenomena between X-rays and Matter.[2]

2.2.1 Absorption

To describe the absorption of X-rays in matter the Lambert-Beer-law is used (equation 1). This law describes an exponential function, which gives the intensity (I) of X-rays after passing a material layer

with the defined density (ρ) and the layer thickness (t). The absorption also depends on the mass attenuation coefficient (μ). I_0 sets the initial intensity for the X-rays.

$$I = I_0 * exp(-\mu * \rho * t) \tag{1}$$

Lambert-Beer-law for the intensity I with I_0: initial intensity, μ: mass attenuation coefficient, ρ: density, t: layer thickness.

The coefficient μ normally consists of the absorption (τ) and the scattering (σ) (equation 2). If we compare τ with σ, we can determine σ is significantly smaller than τ. The approximated relationship is shown in equation 3.

$$\mu = \tau + \sigma \tag{2}$$

Mass attenuation μ with τ: absorption and σ: scattering.

which can be approximated to:

$$\mu \approx \tau \tag{3}$$

Approximation of the mass attenuation μ with τ: absorption.

2.2.2 Emission

The absorption of X-rays in matter can conduct to phonons. It is due to the mechanism of enhance the oscillation for the lattice or over excite atoms by the emission of photo electrons. Due to the high energy of the absorbed X-rays, this electron can even escape from the inner shells of the atom. If the hole in this shell is filled by an outer electron, the atom enters the ground state and energy can be emitted in condition to electromagnetic radiation, shown in Figure 4.

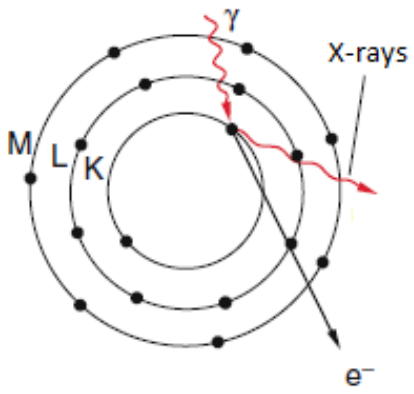

Figure 4: Photo effect as absorption of X-rays by bound atom-electrons. [5]

If the inner shell of the atom is filled up by an outer electron, the atom (which is presently in the ground state) emits the resulting energy differential via a photon. This radiation can be normally located in the range of X-rays. Due to the discrete energy levels the X-rays are called characteristic radiation or fluorescence. The Moseley's law, which is a relation between the energy E (characteristic radiation energy) and the atomic number Z (the atom which is emitting the energy), is given in equation 4. The constants C_1 and C_2 are dependent on the affected electron shell.

$$E = C_1 * (Z - C_2)^2 \tag{4}$$

Moseley's Law for the characteristic radiation energy E with C_1: constant 1, Z: atomic number and C_2: constant 2.

The characteristic radiation can be divided into different radiation expressions: The first three series are

the K-, L- and M-radiation, beginning from the highest frequency. The schematic expression is given in Figure 5.

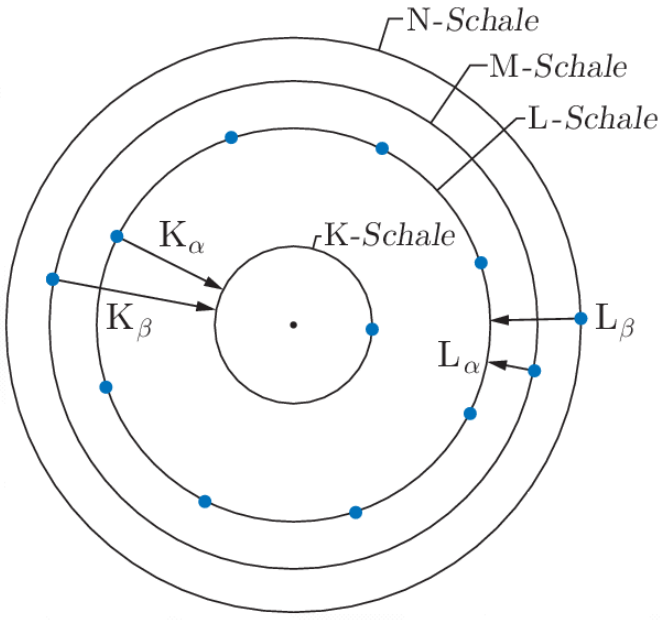

Figure 5: Exploring characteristic radiation for the K, L, M and N shell.[6]

The different Energy-Atomic number dependencies can be shown in Figure 6 where the graph presents the energy series for the inner shells, where the K transition defines the highest frequency or energy (energy-frequency relation: $E = h * \upsilon$ [7]) depending on the atomic number Z.

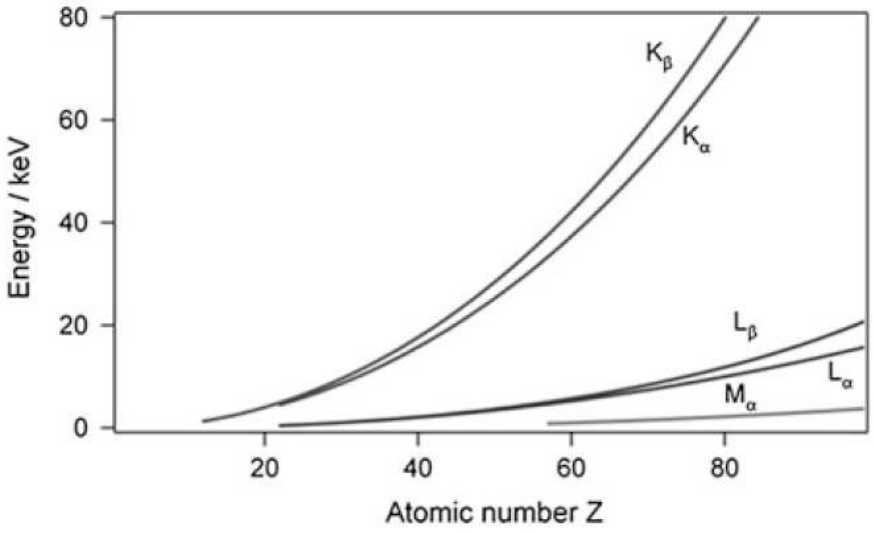

Figure 6: Relation between atomic number Z and energy E.[2]

The characteristic radiation caused by the energy differences ($E_{diff} = E_{vacancy} - E_{outer}$) is one source of energy emission. Another source of energy emission is the Auger-electron where the emitted radiation caused by the photo effect removes an other electron of the same atom. The Auger-electron has the energy: $E_{Auger} = E_{diff} - E_{binding}$. Only one process is possible for energy emission: Characteristic radiation or Auger-electron.[2]

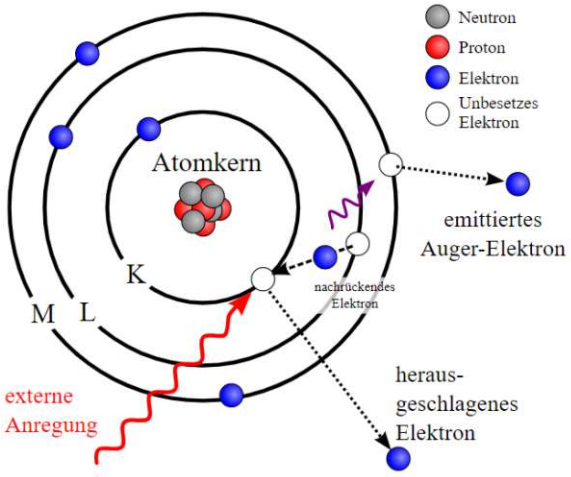

Figure 7: The emergence of an auger-electron. [8]

The sum of the probability between Auger-electron and characteristic radiation is defined as follows: $p_{Auger} + p_{R-ray} = 1$. The fluorescence yield (ω) gives the probability of characteristic radiation. In Figure 8, the dependency between the fluorescence yield and the atomic number is illustrated. The emitted characteristic radiation will be used to investigate the element composition of samples. Also coating systems in reference to their thickness and composition can be determined.[2]

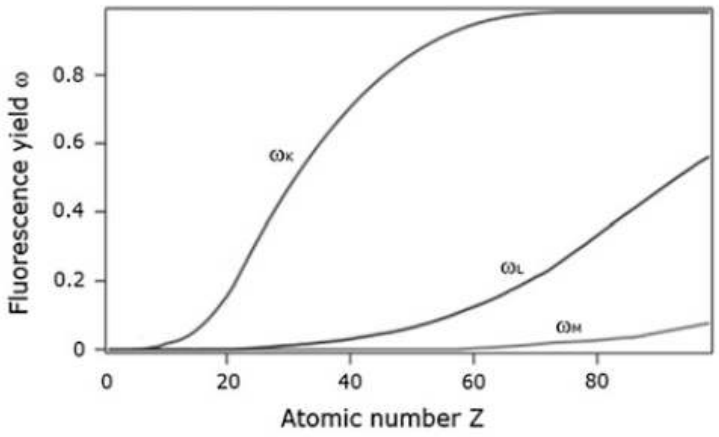

Figure 8: Fluorescence yield ω (Y-Axis) for the shells: K,L and M depending on the atomic number Z (X-Axis). [2]

The spectrum for wavelengths distribution consists of the characteristic spectrum and the continuous spectrum which is shown in Figure 9. The continuous spectrum is called bremsstrahlung, which is generated by decelerated electrons in matter, shown in Figure 10. [7, 2]

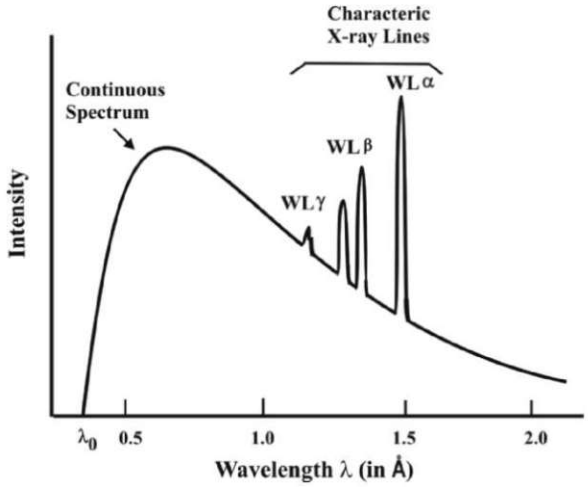

Figure 9: Characteristic ($74Wtarget$) and continuous spectrum, emitted by X-ray tube. [3]

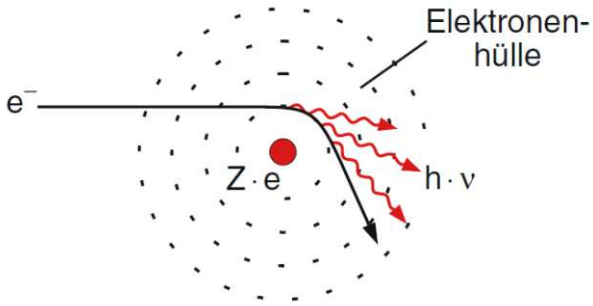

Figure 10: The emergence of bremsstrahlung. [7]

2.2.3 Elastic Scattering

Elastic scattering is described in equation 5 where it is shown that the intensity of the scattered photons depends on the angle υ.

$$I_{scat} = I_0 \frac{1}{r^2} \left(\frac{e^2}{m_0 c^2} \right)^2 * \left(1 + cos^2 \upsilon\right) \tag{5}$$

Scattered intensity I_{Scat} with I_0: primary intensity, r: distance to the observation point, e: charge of an electron, m_0: mass of an electron, υ: scatter angle.

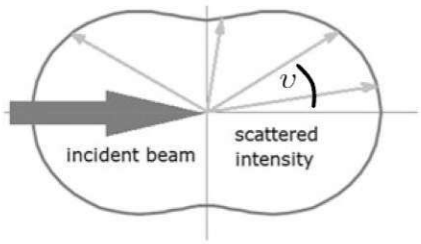

Figure 11: Incidence beam and scattered intensity, which depends on angle υ. [2]

The interaction is without a change in energy, this type of scattering is also called Rayleigh or coherent-scattering. The elastic scattering shows the maximum at 0° and 180° and the minimum at 90° and 270°, which is shown in Figure 11. [2]

2.2.4 Inelastic Scattering

For inelastic scattering phenomena the dependency of the scattering angle υ can be described in equation 6 (wave length shift) and in equation 7 (energy transfer) and it can also be known as Compton or incoherent-scattering.

$$\lambda_{Scatt} = \lambda_0 + \lambda_C * (1 - cos\upsilon) \tag{6}$$

Scattered wave length λ_{Scatt} with $\lambda_C = h/mc$: Compton wavelength, m: mass of the scattering particle(electron), λ_0: wavelength of the incident photon, υ: scatter angle

$$E_{Scatt} = \frac{E_0}{1 + \frac{E_0}{mc^2} * (1 - cos\upsilon)} \tag{7}$$

Energy of the scattered photon E_{Scatt} with E_0: The energy of the incident radiation, m: mass of the scattering particle(electron) and υ: scatter angle.

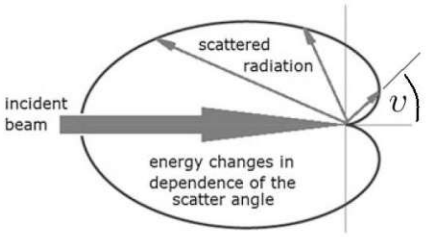

Figure 12: Dependence of the scattered wave length on the angel υ. [2]

According to the loss of energy an increase of the wave length for the scattered photon follows. The energy change, which depends on the angle υ is displayed in Figure 12. For the scattering angle 0°, the loss of energy is 0 i.e. the photons do not hit the electron. By increasing the scattering angle υ, the loss of energy also rises, at 180° the transferred energy reaches the maxima.

2.3 General Design of SR μ-XRF Synchrotrons

The necessary analytical procedure defines the chosen components of the SR-μ-XRF, which are presented in the following chapter. A general overview is shown in Figure 13. The description will follow the course of the beam line.[2]

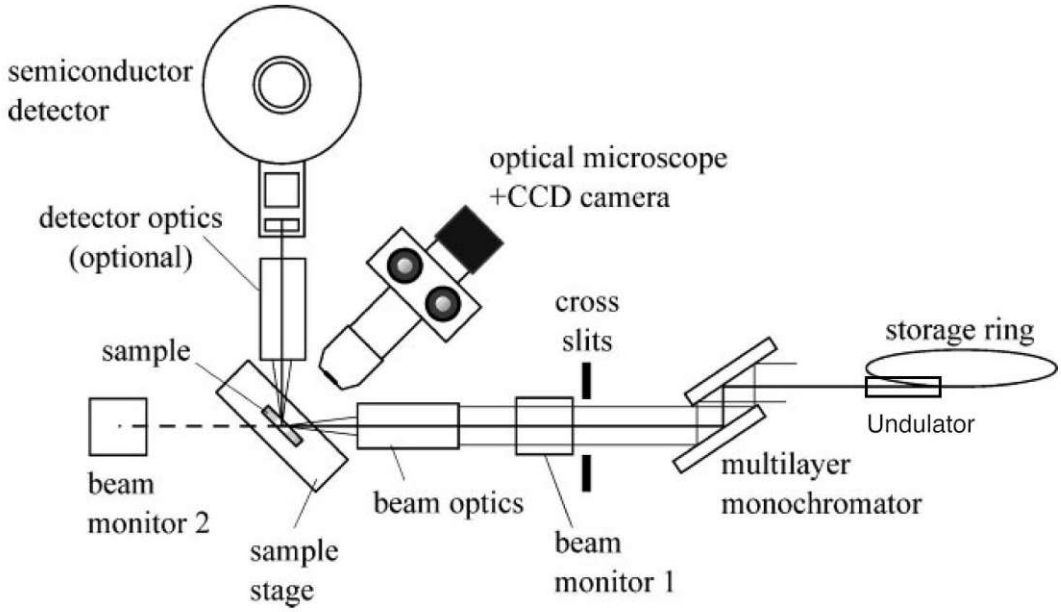

Figure 13: Setups at synchrotron radiation facilities. [9]

2.3.1 Excitation Source

A synchrotron is a highly sophisticated device to generate X-rays. The place of X-Ray production in the synchrotron is the bending magnet, wiggler or undulator, where the accelerated particles (e.g. electrons) moving at a speed close to that of light, emit energy in the form of X-rays.

In bending magnets, the trajectory of the electrons can be described as a circular path, which produces a smooth X-Ray spectrum. The angular distribution $\Delta\Phi$ is inversely proportional to the electron energy γ $(m * c^2)$ which is described in Figure 14.

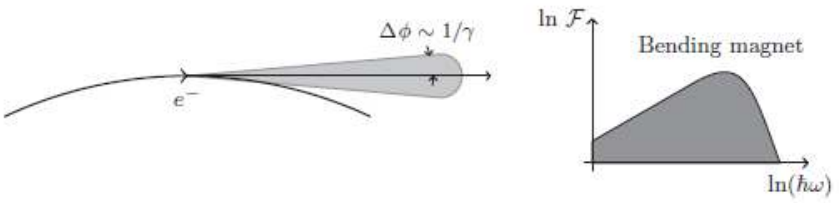

Figure 14: The radiation angular distribution and spectral flux characteristics for bending magnets. [10]

Wigglers consist of periodic alternated magnets, which are designed to bend charged particles back and forth over a nominally rectilinear trajectory. The radiation characteristics are similar to spectrum produced by bending magnets. The intensity increment is proportional to the number of magnetic poles. For 2 N_u poles, the intensity enhancement becomes 2 N_u-fold. The angular distribution is inversely proportional to the electron energy γ $(m * c^2)$, shown in Figure 15.

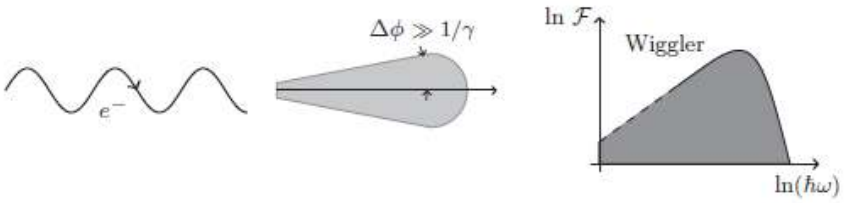

Figure 15: The radiation angular distribution and spectral flux characteristics for wigglers. [10]

An undulator can be understood as an N_u-period magnetic structure which is able to produce a gentle, periodic shell. Undulator radiation is characterised through a discrete spectrum where a narrow beam angle is given. The angular distribution $\Delta\Phi$ is described in Figure 16 by N_u for the number of magnetic undulator periods and γ for the electron energy $(m * c^2)$.[10]

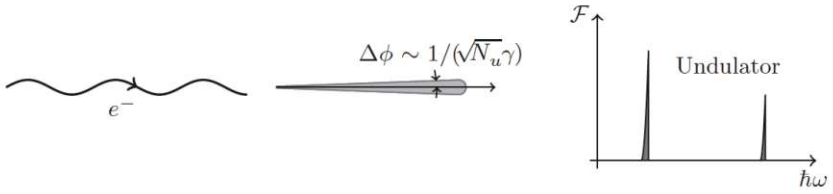

Figure 16: The radiation angular distribution and spectral flux characteristics for undulators. [10]

2.3.2 Primary Optics

Primary optics are able to shape (collimators/cross slits) and focus the beam, in addition to a change of the energy distribution by using filters, secondary targets or monochromators. The beam optics is able to focus the beam to a small spot (for example 20 μm). The intensity of the incoming beam is monitored by beam monitor 1 Figure 13.

2.3.3 Sample Stage

To investigate a small area of a sample which has to be positioned in the beam trajectory, the sample stage is often motorized for the X and Y direction. To localize the position of the sample a microscope is normally used.[11, 12, 10, 2, 9]

2.3.4 Detectors

For detection of X-rays in terms of the energy and intensity diverse detector principles and designs are available. Since the first usage of plates or film detectors a big development has been occurred, especially for semiconductor based detectors. The following example (Figure 17) describes a SDD (Silicon Drift Detector).

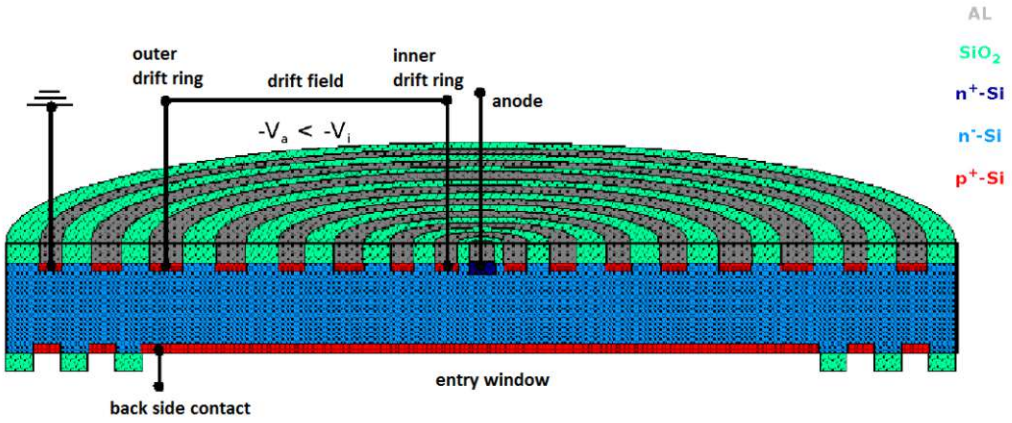

Figure 17: Schematic design of a Silicon Drift Detector. [13]

The entrance window in our example is doped with boron (heavily positive doped) and serves as the income area for the X-rays. The anode at the opposite side of the detector is doped with phosphorus (heavily negative doped). The substrate in the middle region is a lightly negative doped silicone wafer with a resistance of approximately 3000 Ωcm. The centred anode is kept small in compare to the big entrance window and is encircled of a series of drift rings. To create an electric field parallel to the surface, a voltage has to be applied between the outer drift ring (high negative) and the inner drift rings (less negative) which causes to a drift of electrons to the anode. To collect the electrons at the anode efficiently, the concentric drift rings are connected over voltage dividers to generate a potential decrease to the anode (Figure 18). The detected electrons are created by X-rays which strike the detector. The energy of the X-rays lifts the electrons of the detector material from the valence band up to the conduction band where the electrons, caused by the potential difference, drift to the anode and can be read out.

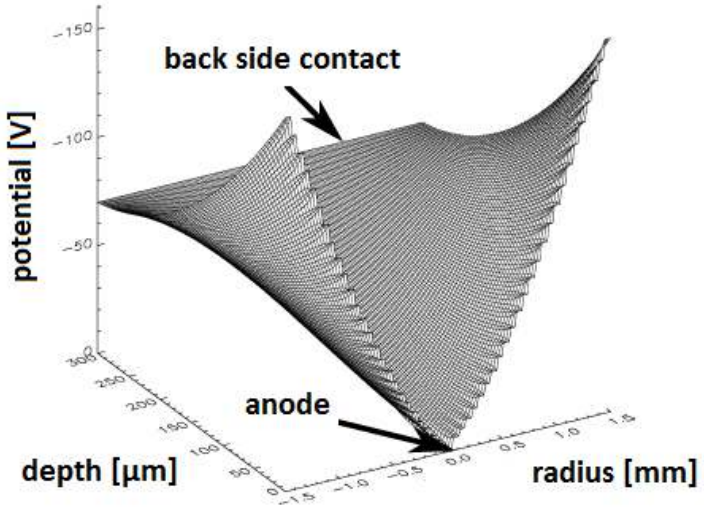

Figure 18: Potential curve in a SDD, caused by the drift ring design. [13]

[2, 13, 14]

2.4 Quantitative Assessments of the Data

To measure the similarity of the images the Pearson's Correlation Coefficient (PCC) and the Spearman's Correlation Coefficient (SCC) are used since they are suitable tools to quantify the correlation between two data distributions. To describe the overlap (co-occurrence) numerically, the Manders Overlap Coefficient (MOC) can be used. These methods (correlation and overlap) should not be confused with each other. To measure the similarity of two images, it is necessary to use both methods for comprehensive data analysis. To get reliable results pre operations have to be done, in relation to segmentation and thresholding.[15]
Correlation analysis can make the relationship between any pair of variables visible. Two commonly used measurement techniques are the Pearson and Spearman correlation, which are explained in the next sub chapters. In general, the output of these methods is a number between +1 and -1 which expresses a perfect relationship between the variables. The value 0 indicates that there is no relationship between the variables and +0.5 or -0.5 normally suggests a relationship.[16]
The Manders Overlap Coefficient (MOC) was introduced to avoid deficits in the PCC, principally because the PCC is not sensitive enough in differences of the signal intensity between two images (background sensitivity). In addition, the negative scale of the PCC is difficult to interpret, when the degree of overlap is the pursued quantity to be measured. The coefficient for the MOC is used to limit the range between 0 and +1, where 0 means 0% overlap and 1 indicates a perfect 100% overlap.[17]

2.4.1 Pearson's Correlation Coefficient (PCC)

The Pearson Correlation Coefficient (PCC) is a good method to show the degree of the linear correlation between two data series. A PCC of +1 indicates a full positive linear correlation and a PCC of -1 is defined for a perfect negative linear correlation. In general, the method is robust against background signal. For example, the first data series (Figure 19) has a perfect linear correlation for the first 10 data values. The background which is represented through the last five data values where the x-values are zero, has entries between 0 and 5 (y-values randomly set). The correlation behaviour shows a robust PCC of 0.9993.

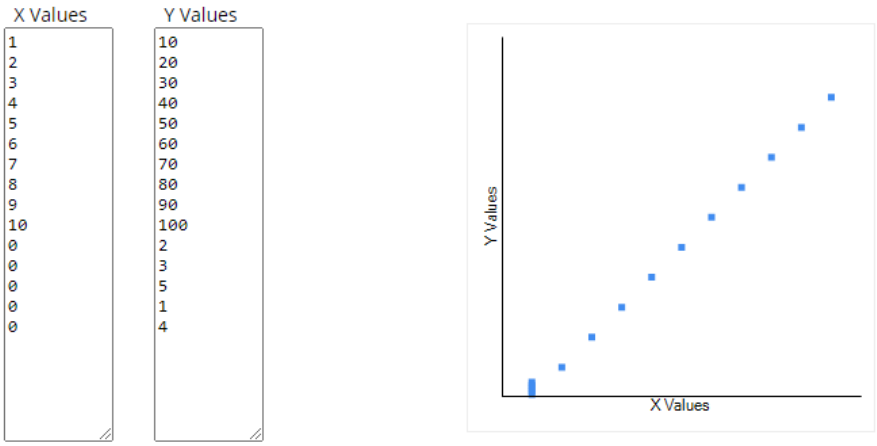

Figure 19: The tables to the left show data pints for the X-Axis and Y-Axis for the graph to the right. The PCC of 0.9993 shows a high robustness against background signal.

The second series has a step size of 10 (distance between the last five y-values) for the background, which is shown in Figure 20. In this case The PCC of 0.8488 can be viewed as less robust against background.

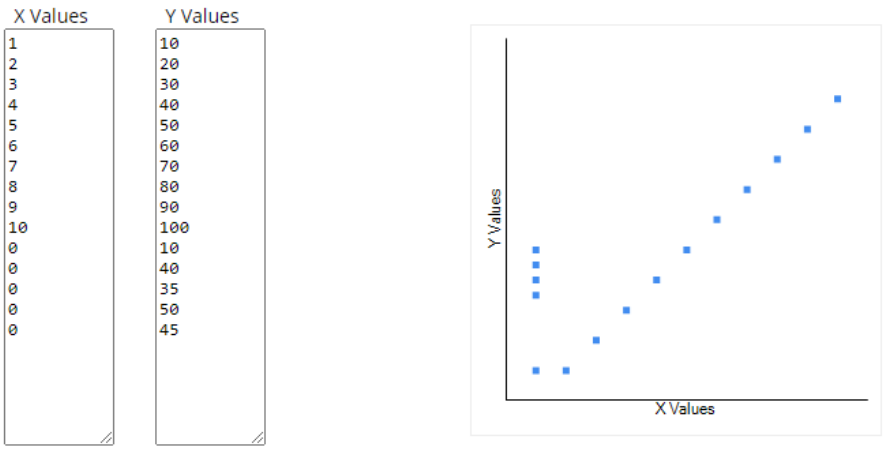

Figure 20: The tables to the left show data points for the X-Axis and Y-Axis for the graph to the right. The PCC of 0.8488 shows less robustness against background signal in compare to Figure 19.

Under some circumstances, the PCC can not measure image characteristics at some points, which is shown in Figure 21. The calculation of the PCC for these image series come out with no change, where the background (green signal dots) increases.

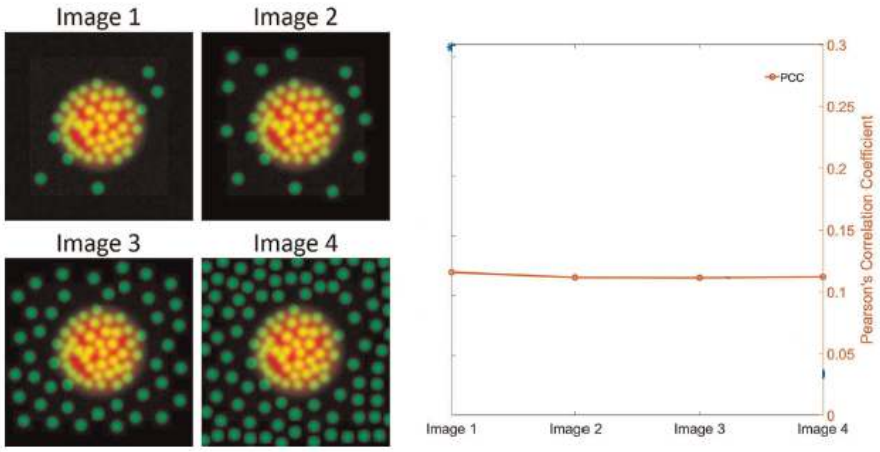

Figure 21: The PCC are measured from image one to image four, where an increase of one signal (green dots) can be observed. The graph plots the four images against the PCC's. The change of image characteristics can not be made visible over the PCC. [17]

[15, 17, 16]

In mathematical terms, the PCC sets the covariance between two images, normalized by the product of their calculated standard deviations. Equation 8 uses the variables $C1_i$ and $\overline{C1}$, which represent the i^{th} pixel intensity and the average pixel intensity of image 1. The variables $C2_i$ and $\overline{C2}$ are the corresponding pixel values of image 2. The total number of pixels is represented by the value of n. It has to be noted that image 1 and image 2 needs the same number of pixels (data points).

$$PCC = \frac{\sum_i \left(C2_i - \overline{C2}\right) \cdot \left(C1_i - \overline{C1}\right)}{\sqrt{\sum_i (C2_i - \overline{C2})^2 \cdot \sum_i (C1_i - \overline{C1})^2}} \tag{8}$$

Pearson's Correlation Coefficient (PCC)

[1]

2.4.2 Spearman's Correlation Coefficient (SCC)

The Spearman's Correlation Coefficient (SCC) is used to measure correlations between two series, where the scatter plot shows a monotonic function behaviour. This can help to make a relationship between two series visible where the PCC gives unexpected results, which includes situations where two series show a good correlated function but are not linearly correlated. The SCC is equivalent to the PCC, but the SCC method is based on ranked pixel intensity and the intensities themselves.
If it is possible to arrange the measured series in serial order (ranks), we can implement the Spearman's Rank Difference Method, to get the SCC. For the following calculations, the SCC is defined by r_s.

$$r_s = 1 - \frac{6 \cdot \sum_i \cdot D_i^2}{N \cdot (N^2 - 1)} \tag{9}$$

Spearman's Correlation Coefficient (SCC) r_s, with $D = R_x$ - R_y: difference of ranks of two series, N: series size

The following example shows the working method of the SCC. X and Y stand for the existing values in both data series. R_x and R_y build the rank for X and Y and D shows the calculated difference of the rank. The rank sets for each X value an associated number, to get a descending sequence of numbers where R_x= 1 is connected to the biggest value of X and R_x= 10 (N= 10) is connected to the smallest value of X.

X= 1	R_x	Y	R_y	D= R_x - R_y	D^2
20	10	16	9	+1	1
22	8	15	10	-2	4
24	7	20	6	+1	1
25	6	21	5	+1	1
30	3	19	7	-4	16
32	2	18	8	-6	36
28	4	22	4	0	0
21	9	24	2	+7	49
26	5	23	3	+2	4
35	1	25	1	0	0
N = 10	N = 10				$\sum D^2 = 112$

$$r_s = 1 - \frac{6 \cdot \sum \cdot D^2}{N \cdot (N^2 - 1)} = 1 - \frac{6 \cdot 112}{10 \cdot (10^2 - 1)} = 0.32 \tag{10}$$

Spearman's Correlation Coefficient (SCC) r_s, with $D = R_x$ - R_y: difference of ranks of two series, N: series size

[18, 1]
For the calculation of a Spearman's correlation, a complete monotonic relationship is not necessary. The Spearman's correlation can also be used to make monotonic components visible. It must be noted that a full SCC value of +1 or -1 can only be reached if the function is hundred percent (strongly) monotonic. (Figure 22) shows a perfect quadratic function where the SCC is 0 but a relationship clearly exists.

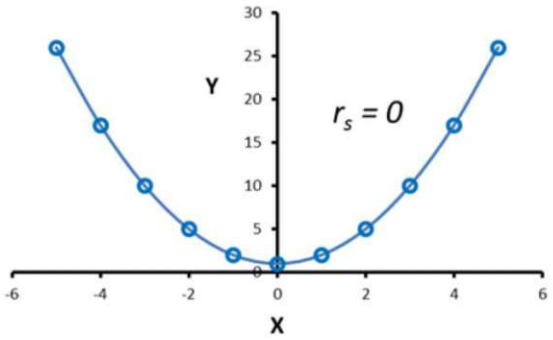

Figure 22: Quadratic function, which shows a SCC(r_s) = 0.[19]

The following Figures (Figure 23 and Figure 24) show examples for the SCC in comparison to the PCC. Figure 23 presents a perfect monotonic function, where the SCC is exactly 1. The PCC shows also a high correlation but also the disadvantage in comparison to the SCC. In Figure 24 an example is given where the SCC shows a higher resilience against strong outliers compared to the PCC. The PCC of 0.67 is strongly impaired by the five outliers (5 data points around x=6) of the whole data set, the SCC of 0.84 is more undismayed of it.

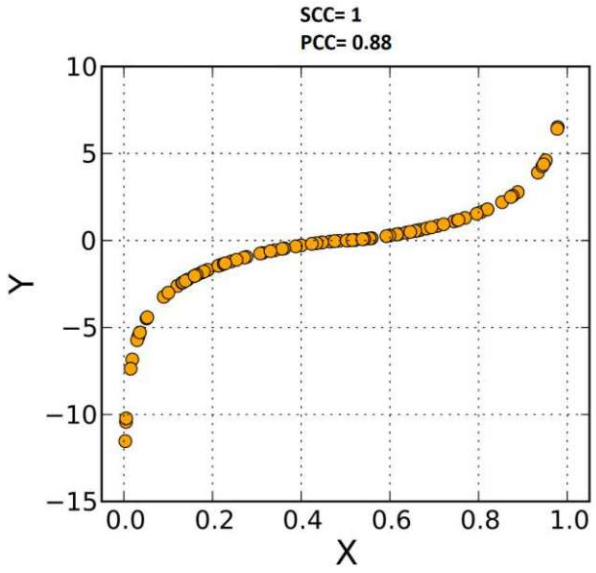

Figure 23: The graph shows a 100% monotonic function, which is shown by the SCC of 1. The corresponding PCC is 0.88 [20]

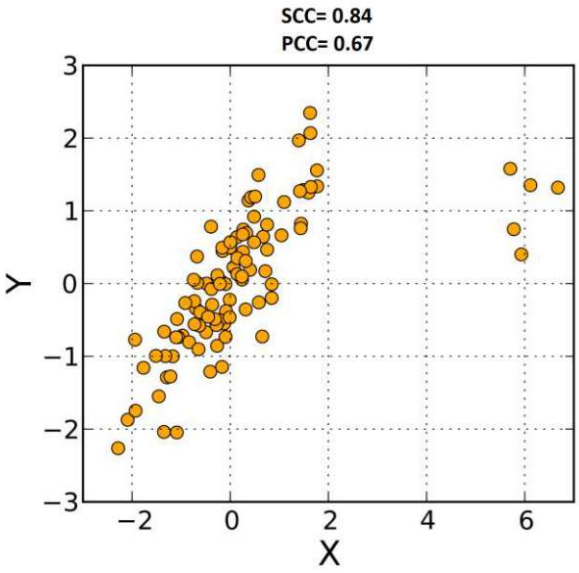

Figure 24: The graph shows a partly monotonic function, which is shown by the SCC of 0.84. The corresponding PCC is 0.67 [20]

[19, 20]

2.4.3 Measuring Overlap by using Manders Overlap Coefficient (MOC)

The Manders overlap coefficient (MOC) is used to determine the overlapping of two images. The MOC operates in a way where the pixel values are divided into two groups. Before the MOC equation can be applied, the images have to be segmented. The pixel values under the threshold limit will be set to 0 and the pixel values over the threshold limit will be set to 1 (or for the following equations (12 and 14) $>$ 0 is also sufficient). The MOC consists of three coefficients. The first coefficient M1 shows the overlap where the pixels of image one (x_i values) overlap those of image two (y_i values), given by equation 11. The number of pixels is defined by the value n.

$$M_1 = \frac{\sum_{i=1}^{n} = x_{i,coloc}}{\sum_{i=1}^{n} = x_i} \tag{11}$$

with:

$$x_{i,coloc} = \begin{cases} x_i & \text{if } y_i > 0 \\ 0 & \text{if } y_i = 0 \end{cases} \tag{12}$$

The second coefficient M2 shows the overlap where the pixels of image two (y_i values) overlap those of image one (x_i values) presented by equation 13.

$$M_2 = \frac{\sum_{i=1}^{n} = y_{i,coloc}}{\sum_{i=1}^{n} = y_i} \tag{13}$$

with:

$$y_{i,coloc} = \begin{cases} y_i & \text{if } x_i > 0 \\ 0 & \text{if } x_i = 0 \end{cases} \tag{14}$$

The third coefficient is the combination of M1 and M2 and is called the overall Manders Overlap Coefficient (MOC), where x_i, y_i and n are defined like before.

$$MOC = \frac{\sum_{i=1}^{n} x_i * y_i}{\sqrt{\sum_{i=1}^{n} x_i^2} \sqrt{\sum_{i=1}^{n} y_i^2}} \tag{15}$$

Both images consist of n pixels. $x_{i,coloc}$ and $y_{i,coloc}$ only have non-zero values if the corresponding x_i and y_i values have a value which is higher than the set threshold. Normally, the MOC equation gives greater importance to brighter pixels (high count rates) and lower weight to pale pixels (low count rates). It has to be noted that our threshold mask sets the pixel value to 0 or to 1. That means, our pixel values are weighted equally.
The following example (Figure 25) shows a plot for the MOC corresponding to four images where an increase of one signal (green dots) can be observed. The change of image characteristics from image 1 to image 4 can be measured with the MOC. For the PCC, it is not possible to make the change in these image characteristics visible. [1, 17]

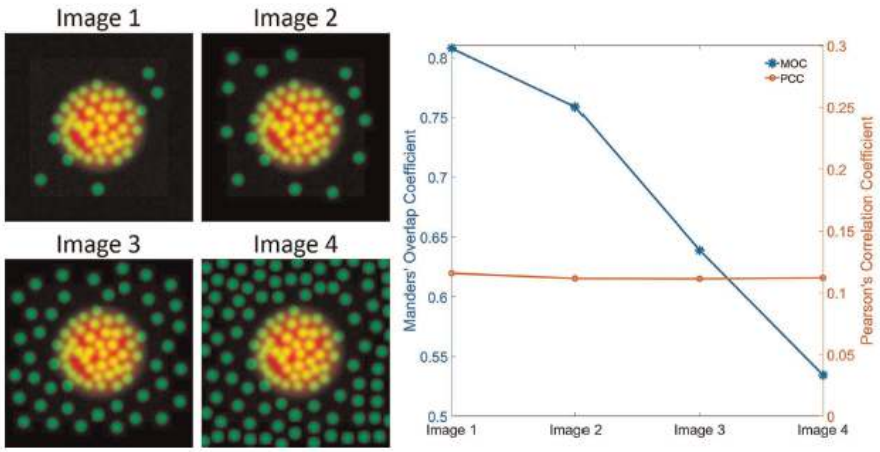

Figure 25: The PCC and the MOC are measured about image one to image four, where an increase of one signal (green dots) can be observed. The graph plots the four images against the PCC's and MOC's. The change of image characteristics can not be made visible over the PCC. The MOC can measure the change of image characteristics.[17]

3 Methods

The following chapter describes the data source and which data sets are selected for the investigations. The methods show all tasks and calculations to get satisfactory results. Furthermore, all necessary steps for the used software and programming are given. The scripts can be explored in section 7.1 (Python code) and section 7.2 (ImageJ macro code).

3.1 Data Source

The data sets were produced by two synchrotrons. The ESRF (European Synchrotron Radiation Facility, 71 avenue des Martyrs, CS 40220 ,38043 Grenoble Cedex 9) and the Diamond synchrotron (Diamond Light Source Ltd, Diamond House, Harwell Science and Innovation Campus, Didcot ,Oxfordshire ,OX11 0DE). The following section should give an overview about the chosen adjustments and used beam lines. The main information about the accelerators can be read up on their websites. The detailed adjustments were taken from the log books of the measured data sets: Logbook ESRF 13-18 July 2017 Gadolinium (Gd) in bone LS-262 and Logbook Diamond 6-12.9.2017.
The obtained data include a number of 31 single scan locations from 5 different patients (GD1, GD2, GD3, GD4 and GD5). Some patients were measured in both synchrotrons, where different sample locations were investigated. The medical background is known for all patients. GD1 has a osteoporotic background and GD2, GD3, GD4 and GD5 presented tumorous activity (osteosarcoma).
[21, 22]
The measured elements are separated in: elements of interest Ca, Cr, Cu, Fe, Gd, Ni and Zn and remaining elements Ar, Co and W. The origin for those elements of interest can be explored as follows: Ca forms the main element for bones, Zn and Fe serves as important components of bone tissue, which are natural presented. Cr, Cu and Ni can also be found and explored naturally or due to age-related behaviours through environmental factors in the life of the patients. The occurrence of Gd can be explained by Gd based contrast agents for magnetic resonance imaging investigations during patient treatments.[23, 24, 25, 26]

The remaining elements Ar, Co and W are present because of there known origins: Ar is out of the surrounding air. Co and W comes from blade contaminations, during sample preparation.

3.1.1 ESRF

The data set was obtained between 13 and 18 July 2017 and investigated with the ID 13 microfocus beamline (Figure 26). The excitation energy was set to 12.7 keV and the optics consists of multilayer-Laue-lenses where the beam size was set to 50 nm furthermore, the detector Vortex (single element) was a little bit declined (Angle Detector-Sample-Beam was more than 90°).

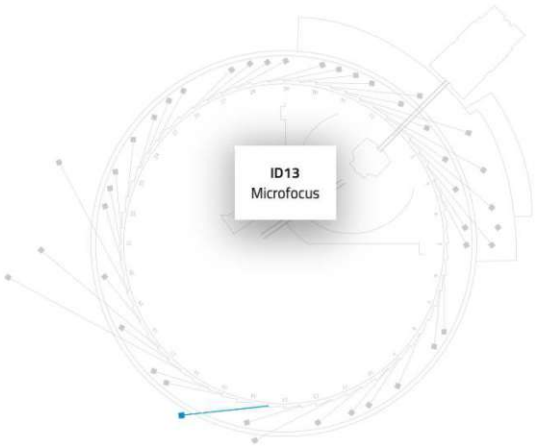

Figure 26: The synchrotron at the ESRF shows the used beam line ID 13 (micro focus) marked.[21]

The collected data for the ESRF synchrotron are listed in Table 1, the total number of single scan locations is n=13.

patient (disease)								
GD1 (osteoporotic)	scan10	scan22						
GD4 (tumorous (osteosarcoma))	scan18	scan17	scan35					
GD5 (tumorous (osteosarcoma))	scan18	scan21	scan27	scan28	scan32	scan34	scan37	scan38

Table 1: The patients are listed in combination to the single scan locations.

3.1.2 Diamond Light Source

The data set was obtained between 6 and 12 September 2017 at the test beam line B 16 (Figure 27) where the excitation energy was set to 12.7 keV. The beam size was set to 500 nm. Furthermore, the detector Vortex is defined by a single element and 150 eV @ Mn-Kα.

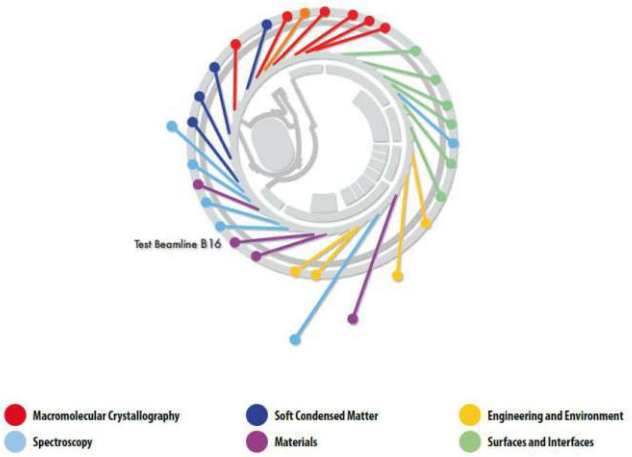

Figure 27: The accelerator at the Diamond shows the used beam line B 16 (Test Beamline) marked.[22]

The collected data for the Diamond synchrotron are listed in Table 2, the total number of single scan locations is n=18.

patient (disease)							
GD1 (osteoporotic)	183278	183279	183280	183242	183309		
GD2 (tumorous (osteosarcoma))	183287	183288	183247	183248			
GD3 (tumorous (osteosarcoma))	183270						
GD4 (tumorous (osteosarcoma))	183258						
GD5 (tumorous (osteosarcoma))	183293	183294	183297	183298	183300	183302	183303

Table 2: The patients are listed in combination to the single scan locations.

3.1.3 Spectrum Translation

The following graph in Figure 28 presents a sum spectrum which is defined as a summation of the whole data output for one sample. The spectrum shows the characteristic energy transitions for the specific elements, sum peaks (two energy events are measured as one event as they come within a time interval that cannot be separated), escape peaks (the energy of one event minus the energy of fluorescence radiation caused by the detector material, for Si= 1,74 keV). The background radiation is mainly caused by elastic and inelastic scattering through the normal atmosphere.

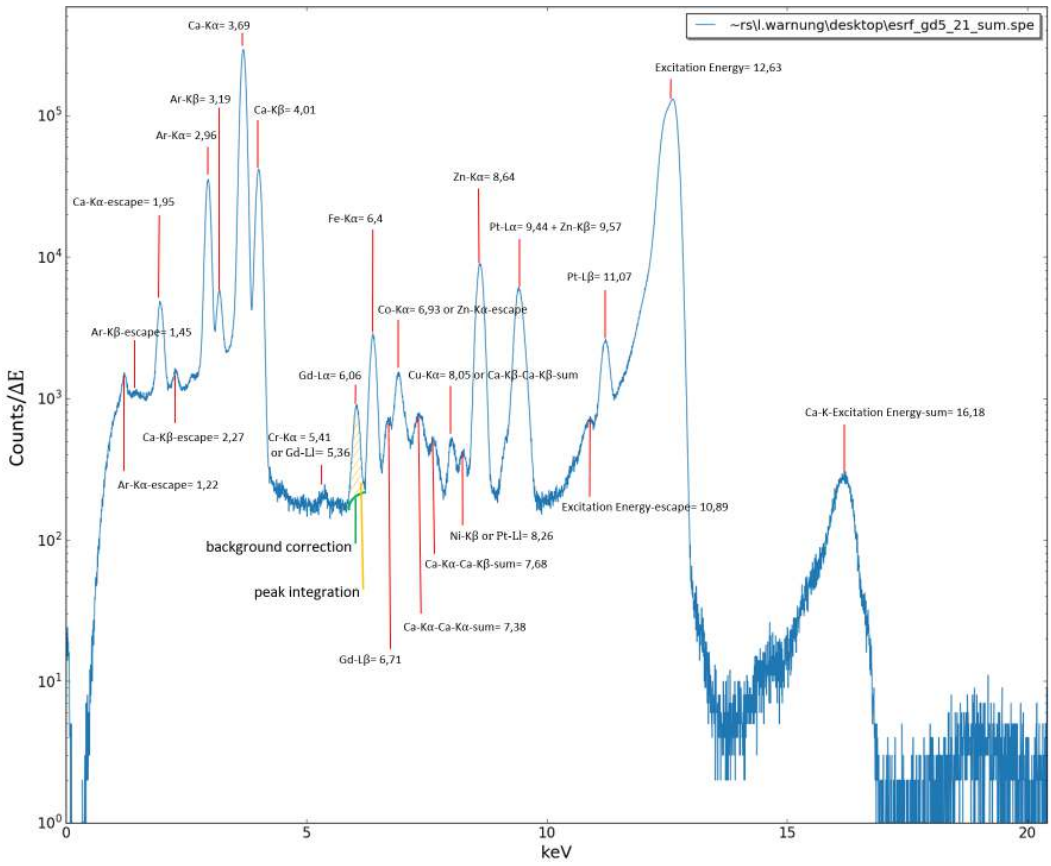

Figure 28: Sum spectrum GD5 scan 21 shows all the characteristic energies for the founded elements, sum peaks, escape peaks and the peak for the inelastic and the elastic scattered peak of the excitation energy. For Gd-Lα the peak integration is shaded and limited by the background correction appreciated for demonstrations.

To get the element maps in Figure 29, the spectrum for each pixel has to be evaluated. For the evaluation, a fit has to be applied for the spectrum where a peak integration for each element peak brings the whole number of counts. The graph in Figure 28 shows an example for Gd-Lα where the peak integration is shaded and limited by the background correction approximated for demonstrations. It should be noted that a total spectrum is only sufficient to get an overview of the possible element occurrences in a sample. Overlapping peaks in the sum spectrum must be examined separately pixel by pixel in order to be able to exclude any elements that may be found.
At the end, the element maps has to be normalized by the measuring time to get the counts per time unit.

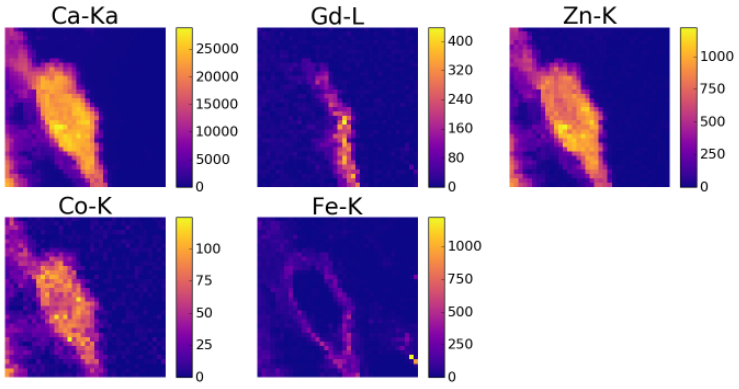

Figure 29: Element maps GD5 scan21 present the characteristic energy distributions for Ca-Kα, Gd-L, Zn-K, Co-K and Fe-K.

[27, 28, 29]

3.2 Data Evaluation

In the following section the whole of image processing scheme and data evaluation is shown. To calculate the correlations (PCC and SCC) Python is used to handle the data sets. For the overlap measurements, ImageJ was defined as a suitable tool. The supplied macro language of ImageJ makes it possible to get quick work-flows and out-puts, which is necessary for processing the whole data set in a manageable period.

3.2.1 Correlation

To calculate the PCC and SCC between two images, python functions were used. The input data forms two XRF images for example Ca and Gd, which is presented in Figure 30 and Figure 31 where the intensity of the pixels reflects the count rats.

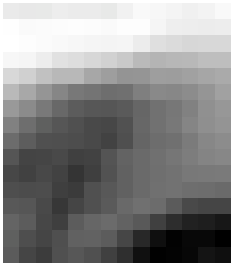

Figure 30: Ca distribution measured by μ-XRF imaging.

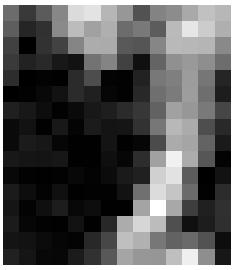

Figure 31: Gd distribution measured by μ-XRF imaging.

To visualize the correlation, X-Y plots are implemented in combination to a linear fit, which is done by the python function. To verify the relationship between two different elements, the slope is also calculated using a python function. The output file is shown in Figure 32.

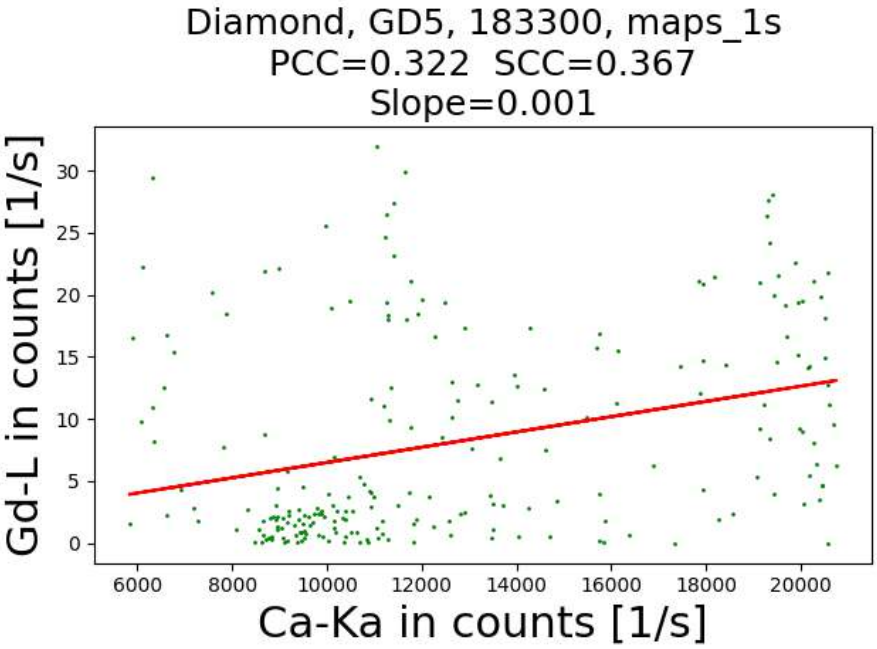

Figure 32: The headline of the correlation graph shows the first line the used synchrotron (Diamond), the patient (GD5), the scan location (183300) and which normalized maps are used (maps_1s). The second line presents the PCC (0.322) and the SCC (0.367) value. In the third line, the value for the slope (0.001) is set. The X axis and the Y axis show which elements are merged for measuring correlations and how great the bandwidth is for the number of counts. Each data point of the graph presents one pixel corresponding to the number of counts. Finally the correlation line to the corresponding slope is indicated.

The output file for the correlation coefficients presents the linear correlation (PCC) and the monotonic correlation (SCC) between two element distributions in addition the slope is also stated.

3.2.2 Overlap

The calculation of the overlap coefficients M1, M2, MOC and the overlap images are done via the program ImageJ by the plugin JACoP. Before the overlap coefficients can be calculated, a threshold level (which is equal for all element images) has to be set. The given example (Figure 33) shows the work flow for adding the defined threshold level of 10 counts/second.

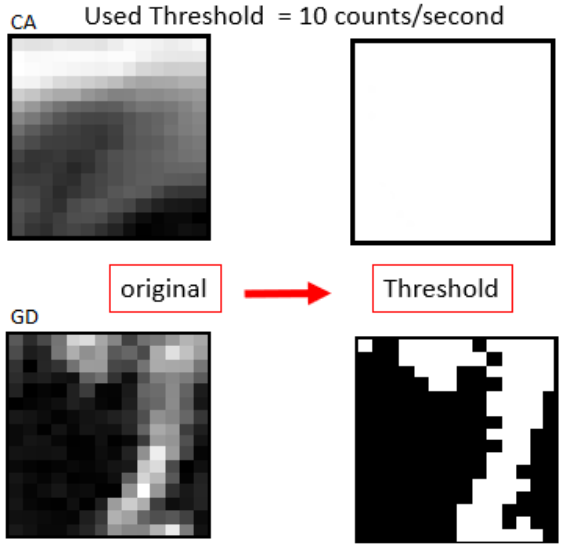

Figure 33: Work flow for adding threshold. Left: original Ca and Gd image. Right: processed images via threshold.

Due to the unknown exact noise level, which is estimated between 0 and 5 counts/second, it has to be noted that a small amount of information gets lost when the threshold level is set to a level of 10 counts/second. Figure 34 presents an XRF image for Cu to the left and Ca to the right. Where the Ca image presents a intuitive distributed structure, is the Cu image more than less randomly distributed. Due to the small count rates, which are shown in the middle image in Figure 34, the Cu image could not be taken as clear information. Therefore a threshold level of 10 counts/second keeps the results free of noise approximately.

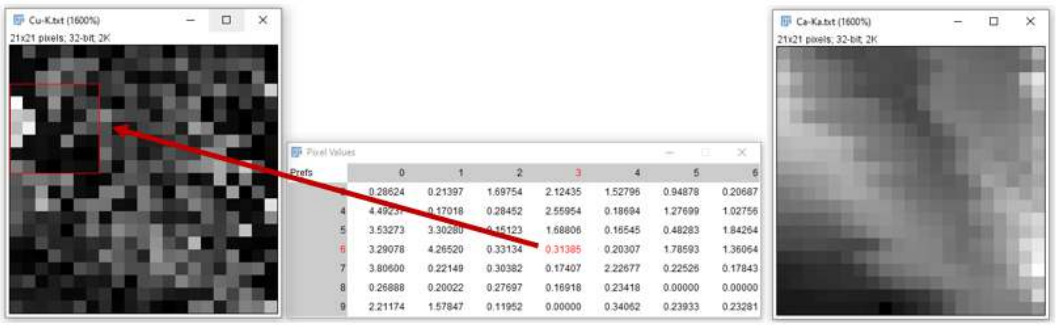

Figure 34: The XRF image to the left shows count rates under 5 counts/second. The XRF image to the right gives the corresponding image for the Ca-K counts (around 10 000 counts/second max.)

After setting the threshold level, the pixel values consist of 0 (black/ no signal) and 1 (white/ signal). That means, for the overlap calculations, that each pixel has the same weight of information. The overlap is calculated by the ImageJ tool JacoP (Figure 35) where the generated out-put text file delivers the coefficients M1, M2 and MOC (marked with a red box).

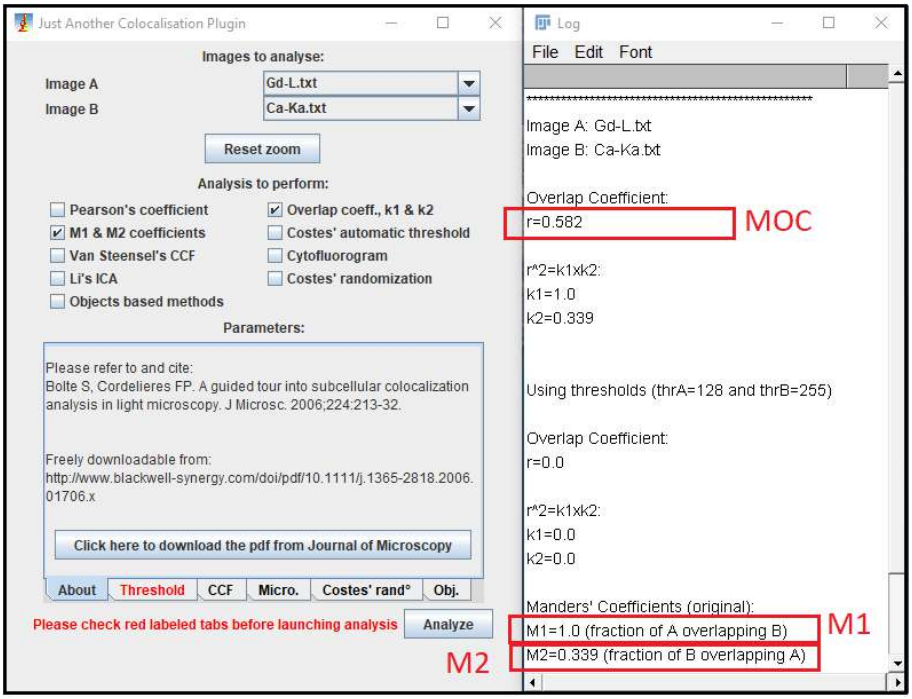

Figure 35: Plug-in program JACoP (ImageJ). Left: windows for program settings. Right: Log file for out-put parameters: MOC, M1 and M2 (marked by red rectangles).

Furthermore the program calculates the overlapping images for the intensity maps (Figure 36), without added threshold and the overlap images, where the pixel values are set to 0 or 1 by the threshold of 10 counts/second (Figure 37). The overlapping images consist of the stand alone areas, where element 1 (red) is measured in absence of element 2 (blue) and the overlap areas, where both elements are measured (magenta).

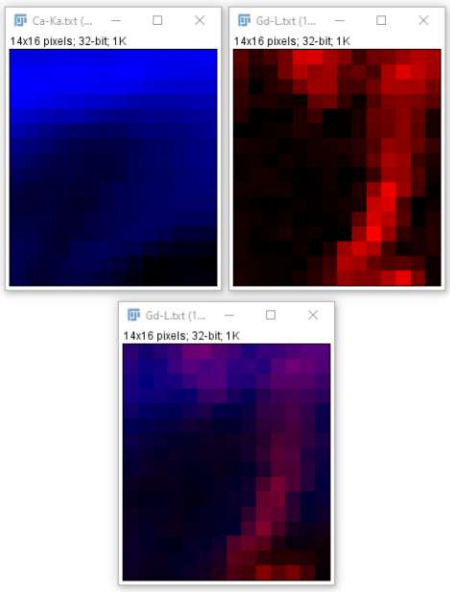

Figure 36: Top: Input images (left: Ca, right: Gd). Bottom: Out-put image for the intensity overlap without threshold.

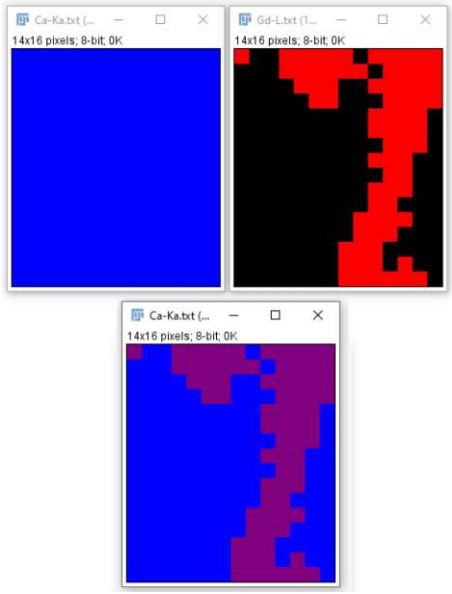

Figure 37: Top: Input images (left: Ca, right: Gd). Bottom: Out-put image for the overlap with threshold.

The count rates are presented in the correlation graph, which are linked to the overlap images in the multi plot slides (Figure 39). For the intensity maps the color intensity is independent of the chosen element, which is important for interpretation. For the given example (Figure 36), the maximum counts are detected for Gd by approximate 30 counts/second and for Ca approximate 20000 counts/second (Figure 32).

For the overlap images where the threshold is set, the whole element areas are visible and the interpretation is more vivid corresponding to overlap informations. This circumstances are recognizable if the comparison between Figure 36 and Figure 37 will be considered, where the presence of Ca is not fully visible for the intensity images (Figure 36).

3.2.3 Output Example For Single Location Interpretation

To get an overview for one scan location, the following images are given (Figure 38): The qBEI (quantitative Backscattered Electron Imaging) image, where the surface of the bone sample is shown. The orange rectangle shows the location for the rough XRF scan and the small green rectangles inside the rough scan show the location for the μ-XRF images, which are presented for the locations # 183293 and # 183294.

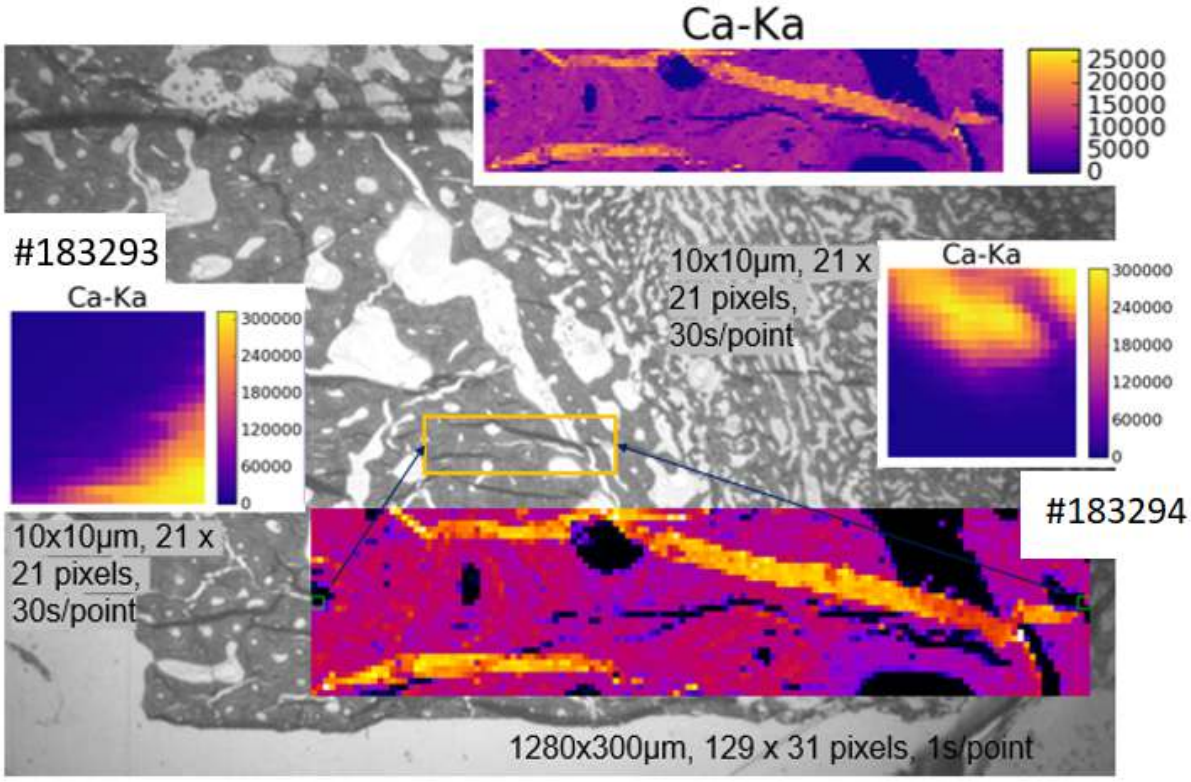

Figure 38: Original data overview, which shows the qBEI image (background image in a gray scale), the located rough scan (orange rectangle), scan locations (# 183293 and # 183294) for μ-XRF images marked by green squares.

The multi plot (Figure 39) provides an overview to interpret the correlation and overlap data in one image. At the left side (correlation graph) of the y-axis (Gd counts) is the original Gd map located. Under the x-axis of the correlation graph, the original Ca map is located. In the lower left corner, the link between the colors and the elements is given. Furthermore, the overlap coefficients M1, M2 and MOC (M) are given. The image to the right of the correlation graph presents the overlap intensity image, where no threshold is set. Finally the right lower corner presents the overlap image, where the threshold of 10 counts/second is set.

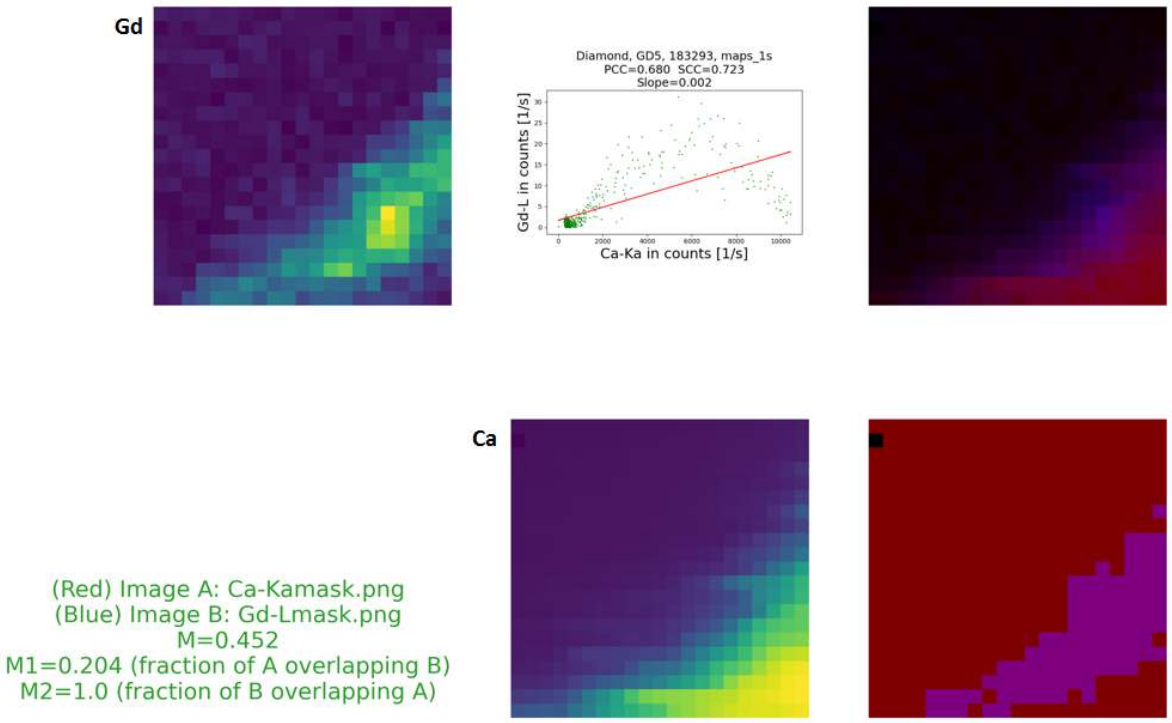

Figure 39: First line from left to right: 1. original Gd map, 2. correlation graph and correlation coefficients, 3. overlap without threshold. Second line from left to right: 1. Element-color legend and overlap coefficients, 2. original Ca map, 3. overlap with threshold

3.2.4 Statistics

For statistic evaluations, all patients (GD1,GD2,GD3,GD4 and GD5) and scan locations (n=31) were plotted in one distribution to calculate the PCC, SCC, Slope, M1, M2 and M for each element comparison, the following example in Figure 40 is given for explanations.
The main title (Ca-Ka) sets the first element for correlation and overlap calculations. On the left side of the graph, all elements are listed. The (SCC) degree of relationship between two elements, is defined between -1 and +1 under the specific relationship coefficient for correlations. For the overlap coefficients, the scale reaches from 0 to +1. Each element combination presents the mean value (mean), the standard deviation (std) and the median. The box plots consist of the quartiles (inner 50% in total), the whiskers (outer 50% in total), the marked median lines and outliers (small circles).

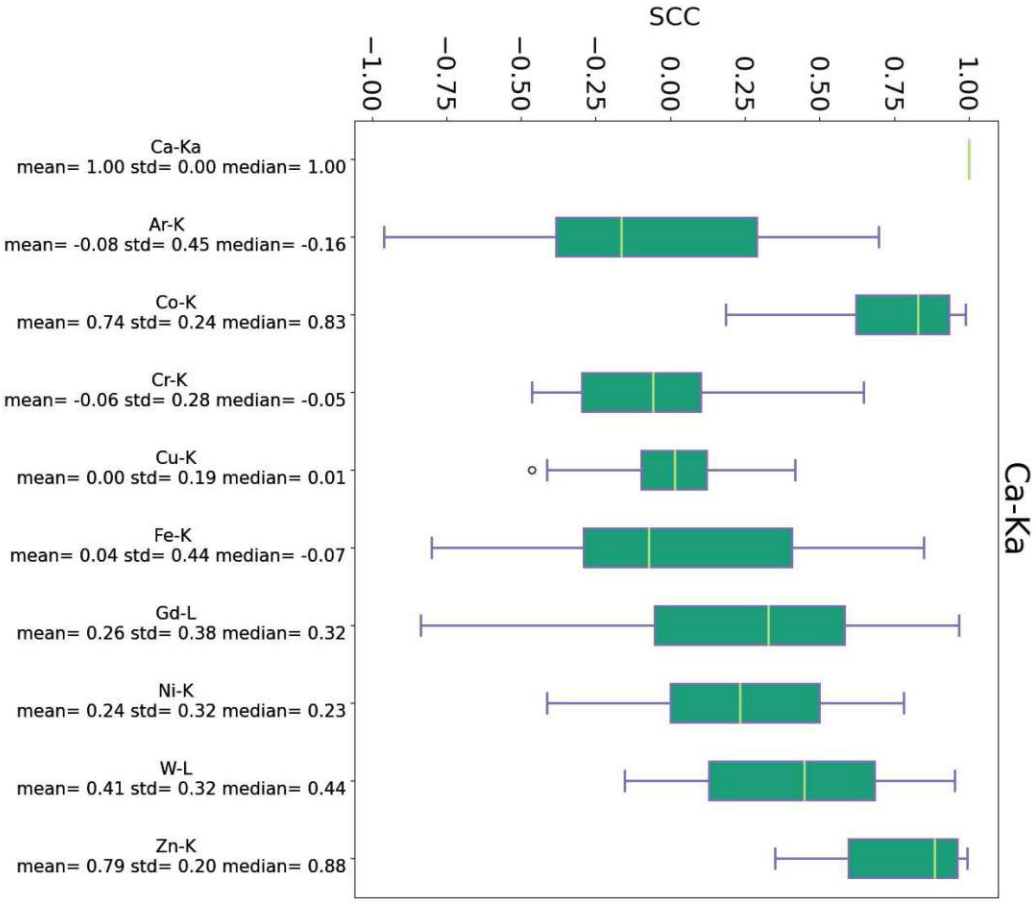

Figure 40: Monotonic correlation (SCC) between Ca (head line) and other elements. The mean value (mean), standard deviation (std) and median is given for each element composition.

For statistical evaluations, in terms of comparing groups, only the patients GD1 (n=7) and GD5 (n=15) were used. Due to the small sample sizes for GD2 (n=4), GD3 (n=1) and GD4 (n=4), those patients could not be used for further comparison investigations.
The following figure (Figure 41) presents a box plot example to compare patient GD1 and GD5 via the corresponding SCC.
The main title (Ca-Ka) sets the first element for correlation or overlap calculations. On the left side, the graph all elements are listed. The (SCC) degree of the relationship between two elements is defined between -1 and +1 under the specific relationship coefficient for correlation. For the overlap coefficients, the scale reaches from 0 to +1. Each element combination presents two groups (GD1 and GD5). The p-value, the mean value (mean) of GD5, the standard deviation (std) of GD5 and the median of GD5 are given in the first line under each element designation. The effect size Hedges' g (hedges-g), the mean value (mean) of GD1, the standard deviation (std) of GD1 and the median of GD1 are given in the second line under each element designation. The box plots for the two groups (GD5: blue plot and GD1: green plot) consist of the quartiles, the marked median lines and outliers (red crosses).

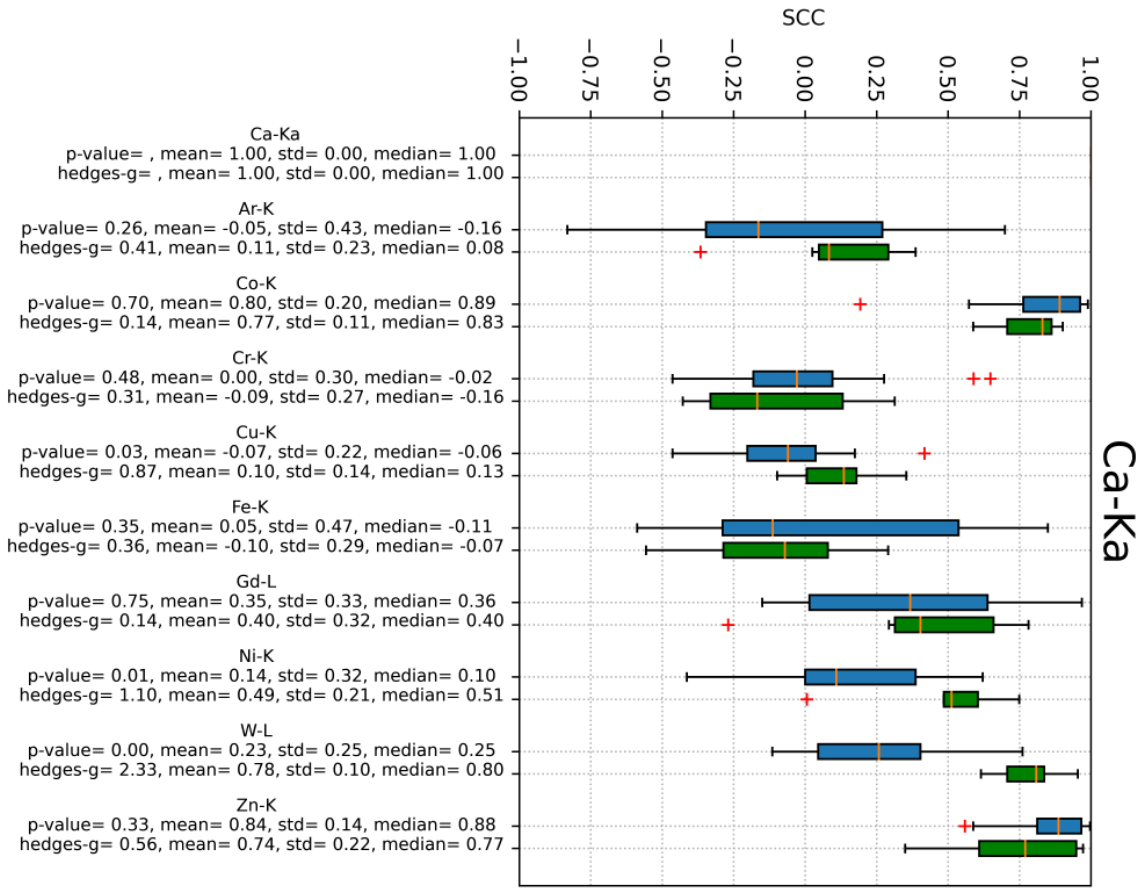

Figure 41: Monotonic correlation (SCC) between Ca (head line) and other elements. The mean value (mean), standard deviation (std) and median is given for each element composition. Furthermore the data set consists of two groups. The blue bar describes GD5 and the green bar describes GD1. The two groups are compared to each other. The p-value and hedges-g are given for comparison.

To get informations about significance, in terms of group comparison, the p-value and the hedges-g are calculated. The p-value is calculated about the student t test, which is suitable for small sample sizes, the significance level was set to $\alpha = 0{,}05$ (p<= 0,05 means: The groups are significant different). Hedges-g is a method for effect size measuring to underline significance and to make the distinction between two groups visible. A high Hedges-g means great differences between two groups. A small Hedges-g means, the two groups are still significantly different but the difference can be yet low. The definition of a suitable Hedges-g value which interprets high, medium or low difference, is beyond the scope of this work. Nevertheless, Hedges-g is an important basement in statistic works to compare similar studies with each other. Hedges-g is the developed effect size for different sample sizes based on the effect size Cohen's-d. Cohen's-d is defined as the difference between the mean values of two groups over the common group standard deviation. The mathematical definition of Hedges-g is presented in 16 and 17. [30]

$$g = \frac{M_1 - M_2}{S} \tag{16}$$

Hedges-g with M_1: mean value of group 1, M_2: mean value of group 2 and S: common group standard deviation.

with

$$S = \sqrt{\frac{((n_1 - 1) * SD_1^2 + (n_2 - 1) * SD_2^2)}{n_1 + n_2 - 2}} \tag{17}$$

Common group standard deviation S with n_1: sample size of group 1, n_2: sample size of group 2, SD_1: standard deviation of group 1 and SD_2: standard deviation of group 2.

4 Results

In the following chapter, the results obtained with the implemented methods will be presented.

4.1 Measurement for Single Scan Locations

The measurement for a single scan location gives a qualitative overview about the behaviour between element combinations in one scan. Each one of the 31 scans can be interpreted separately. For that, images are created which describe the comparison of two elements. For one scan, 21 element combinations are possible for the elements of interest. To get the high correlated data, a filter was set by the PCC and SCC value, if the PCC or the SCC value is greater then 0.65, the data is considered meaningful. The value of 0.65 reduces the output data to a still manageable size and describes according to a rule of thumb the beginning of high correlated data. The whole data set is presented in the appendix (section 7.3). Nevertheless, the scan location # 183278 and its interpretation is given below as an example.
Figure 42 presents an overview image which shows the exact scan position of the μ-XRF image for the scan number 183278.
[31]

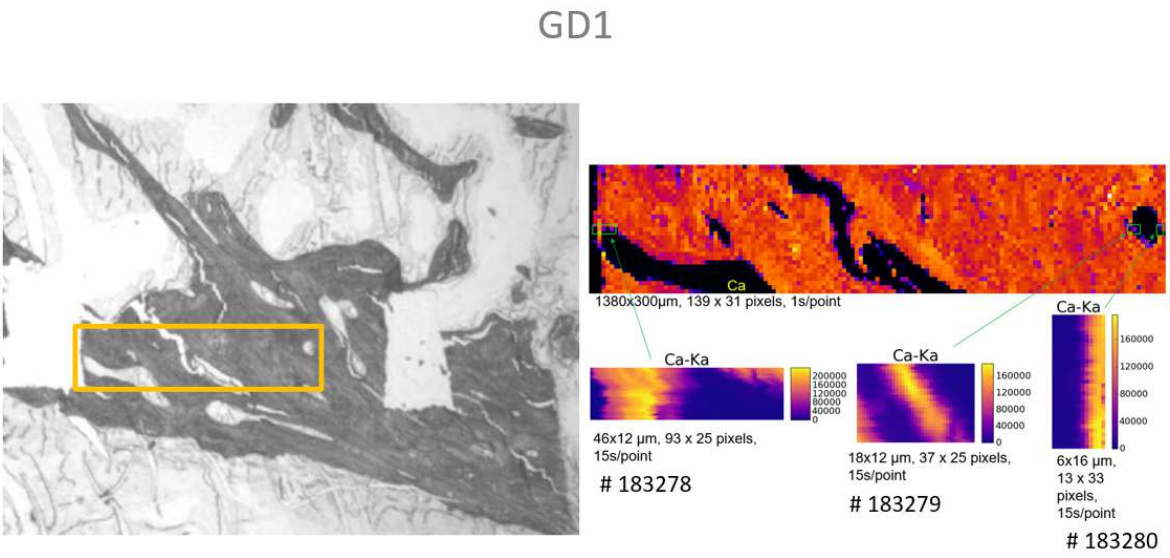

Figure 42: Original overview image, which shows the qBEI image (left image in a gray scale), the located rough scan (orange rectangle), scan locations (# 183278, # 183279 and # 183280) for μ-XRF images marked by green rectangles.

Around the zero position for the x-axis in the correlation graph (Figure 43), an accumulation of Gd counts can be located alongside a small linear correlation region at the end is to be found. The PCC= 0.727 and the SCC= 0.732. The minor slope of 0.003 is caused by the huge difference between the Ca and the Gd count rates. The M2 shows Gd overlaps Ca to 100%. Ca overlaps Gd to 42.1 %. The bilateral (combined overlap of M1 and M2) overlap M in percentage is 64.9 %.
The intensity of the Gd signal increases from the outer to the inner region which can be explained by the change of the color. The inner magenta region means a higher correlation between Ca and Gd in compare to the outer region where the color is red which implies a weak correlation between Ca and Gd. This behaviour is linked with the correlation graph.

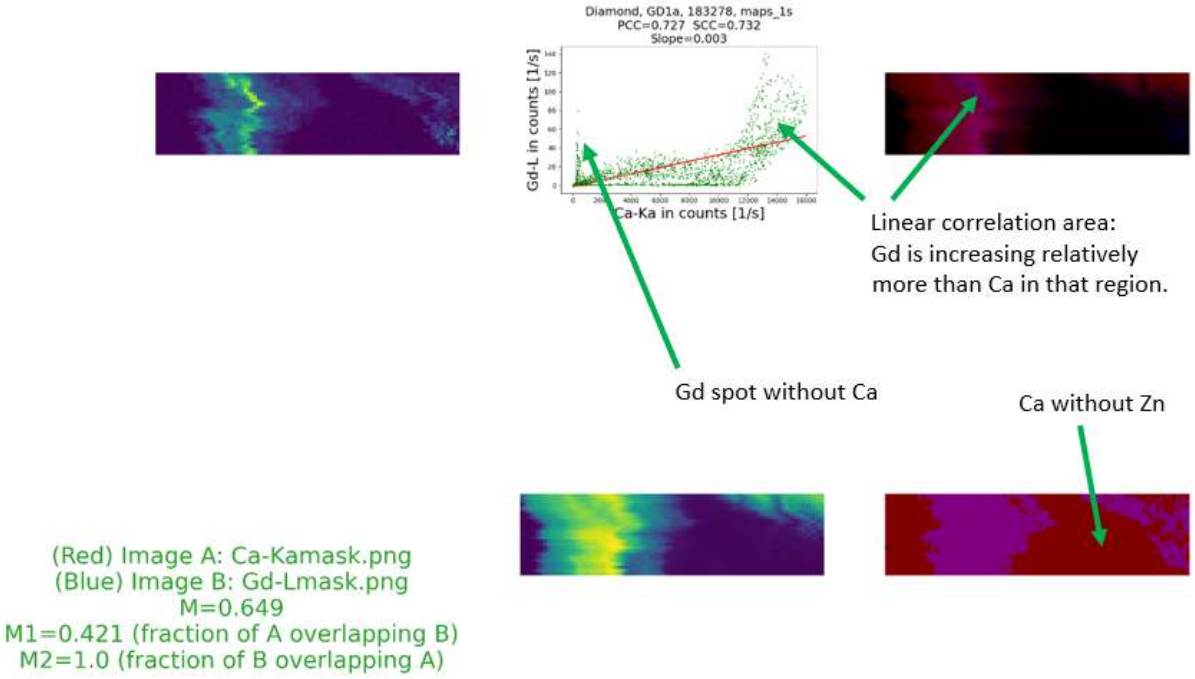

Figure 43: First line from left to right: 1. original Gd map, 2. correlation graph and correlation coefficients, 3. overlap without threshold. Second line from left to right: 1. Element-color legend and overlap coefficients, 2. original Ca map, 3. overlap with threshold

In Figure 44, a high linear correlation (PCC) of 0.929 can be observed and the SCC= 0.962 shows a small non linear but nevertheless monotonic behaviour in addition to the PCC. The slope of 0.023 is caused by the difference between the Ca and Zn count rates. Furthermore, Ca overlaps Zn to 100%, Zn overlaps Ca to 69.7% and the bilateral overlap (M) is 83.5 %.
The intensity of the signal increases from the outer to the inner region, which is specially present in the intensity overlap image.

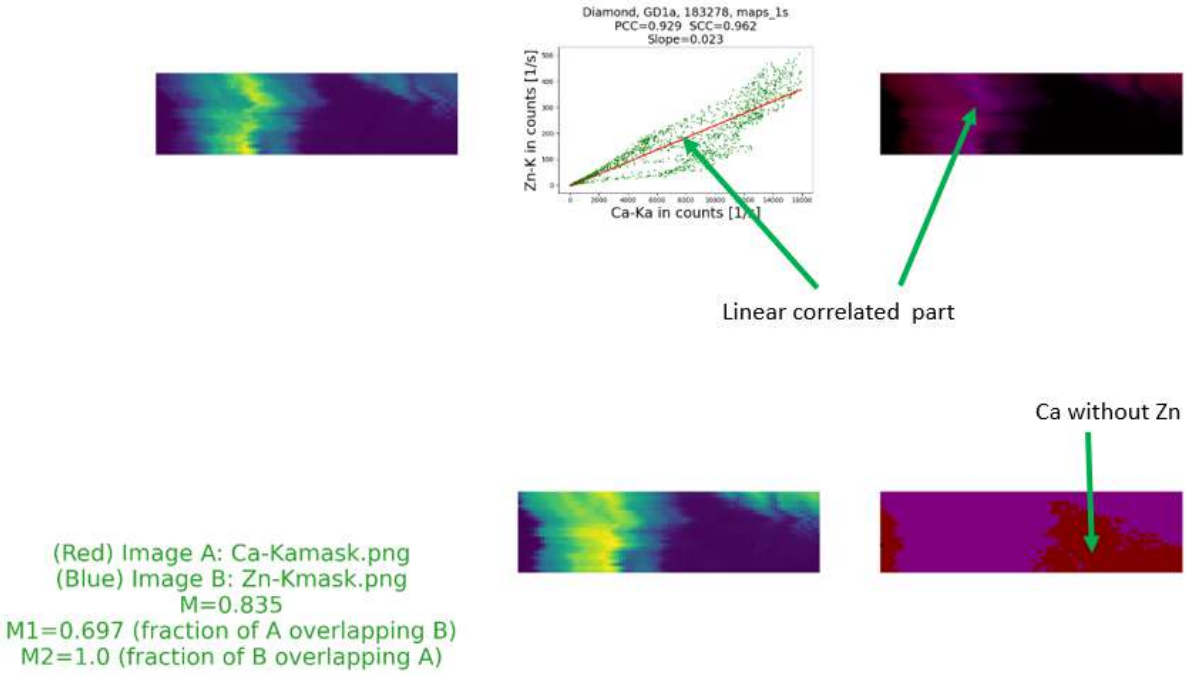

Figure 44: First line from left to right: 1. original Zn map, 2. correlation graph and correlation coefficients, 3. overlap without threshold. Second line from left to right: 1. Element-color legend and overlap coefficients, 2. original Ca map, 3. overlap with threshold

In Figure 45, the linear correlation (PCC) of 0.855 is high and around the zero position of the y-axis, a small Gd spot without Zn can be shown which caused a lower SCC of 0.8 in related to the PCC. The slope of 4.627 is caused by the higher count rate of Zn compared to Gd. Zn overlaps Gd to 57.4 %. Gd overlaps Zn to 95.1 % which can be linked to the small Gd spot where no Zn is measured. Zn without Gd can be located at the edge regions.

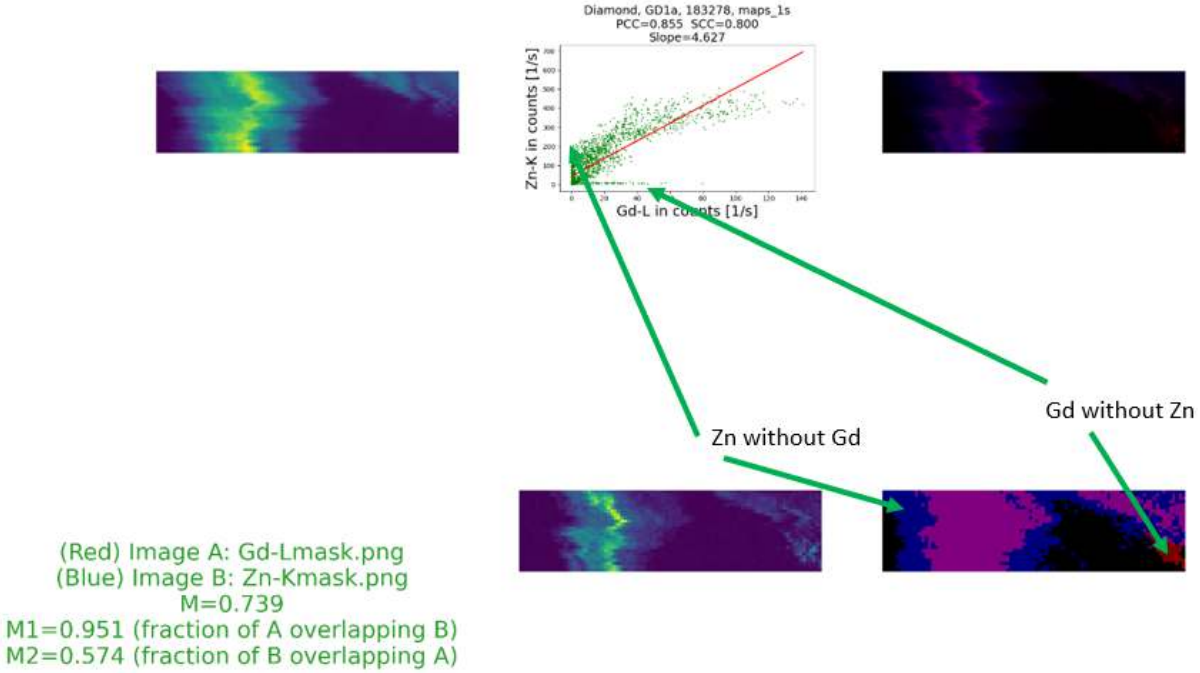

Figure 45: First line from left to right: 1. original Zn map, 2. correlation graph and correlation coefficients, 3. overlap without threshold. Second line from left to right: 1. Element-color legend and overlap coefficients, 2. original Gd map, 3. overlap with threshold

4.2 Measurement for One Group

To make the distribution for the whole data set visible, box plots were created for data interpretation. Each element is compared to all other elements for the coefficients: PCC, SCC, Slope, M1, M2 and M, which forms an accumulation of 42 images. The distribution of one bar is created with n= 31 scan locations (patients: GD1, GD2, GD3, GD4 and GD5). The main focus was set on the elements Ca, Zn, Ni, Fe and Gd. The elements Ar, Co and W are also displayed for the box plots. Due to the known source, those three elements are marked transparent. The elements Cr and Cu must be investigated separately because of the small count rates and the suspected high noise level. To reduce the results only the box plots for the SCC and M coefficient are shown, which present a good overview of the relations between the elements. The whole box plot set can be found in the appendix (section 7.3).
The monotonic correlation (Figure 46) between Ca and Zn is 0.86 for the median and the standard deviation is quite low (0.2) and even negative. Ca poorly correlates to all other elements. Compared to the linear correlation (section 7.3), the deviation is marginally higher for the SCC.

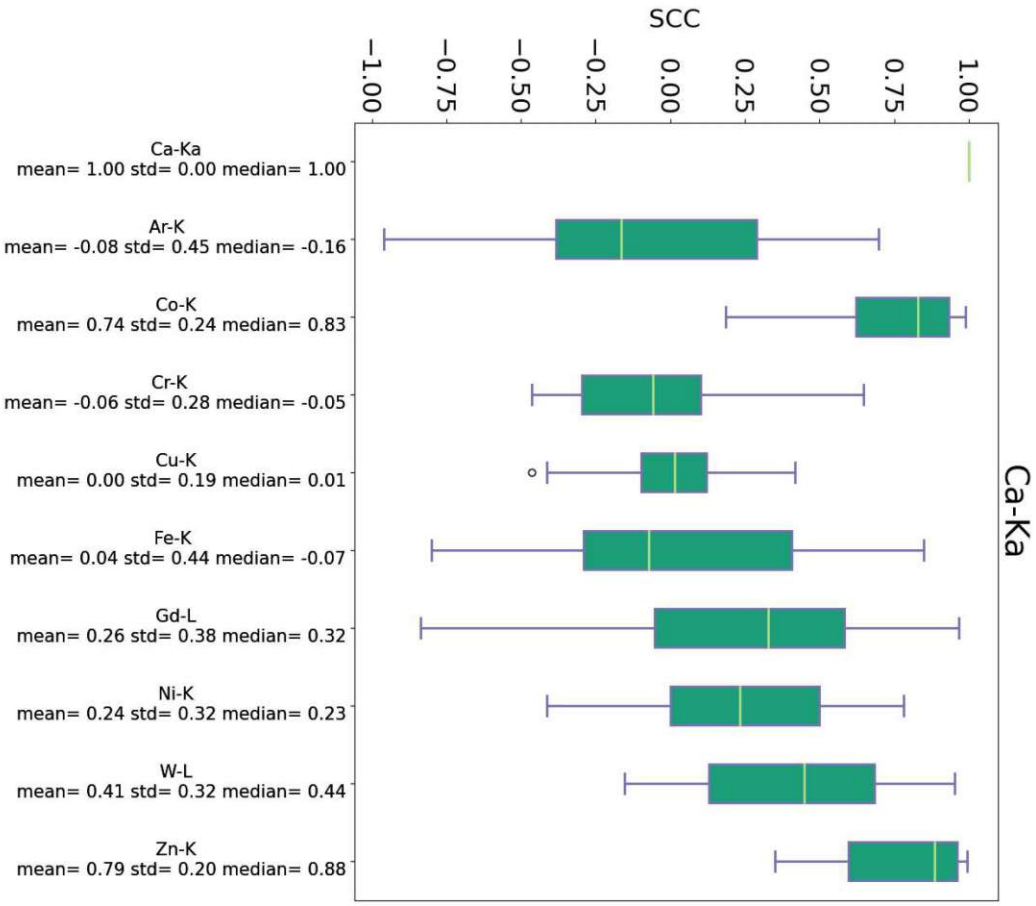

Figure 46: Monotonic correlation (SCC) between Ca and other elements.

In Figure 47, the overlap between Ca and Zn is very high, is 99% (median). The overlap between Ca and Fe is 76% (median) and the standard deviation shows a value of 0.26. The overlap between Ca and Gd is 54% (median) and the standard deviation of 0.29 can be considered high. The overlap of 29% (median) between Ca and Ni is low and the standard deviation of 0.29 is high. The overlap between Ca and Cr is 0% which can be explored by noise problems. The M1 (Ca overlaps Cr) coefficient shows a value of 0% (found in section 7.3), which means an almost zero Cr amount in the Ca regions. However the M2 (Cr overlaps Ca) coefficient gives a value of 31% and a deviation of 0.46 (found in section 7.3) which would mean, that in an average Cr region of 69% no Ca is localized. This behaviour suggests a high noise level for the count rates of Cr. The same noise explanation could be found for Cu.

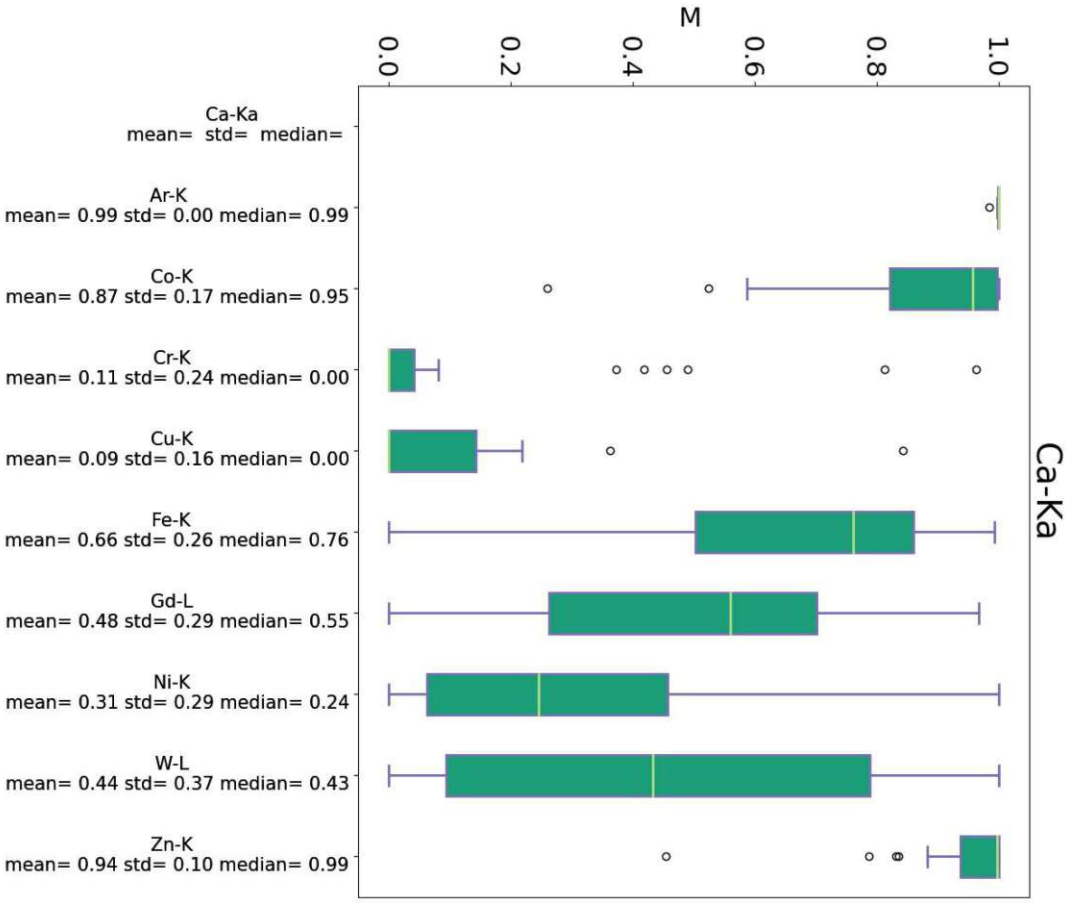

Figure 47: Overlap coefficient (M) between Ca and other elements.

The monotonic correlation in Figure 48 between Zn and Ca is 0.86 for the median (like for Figure 46). For all other elements, the monotonic correlation is quite low, the deviation is high in general and also negative.

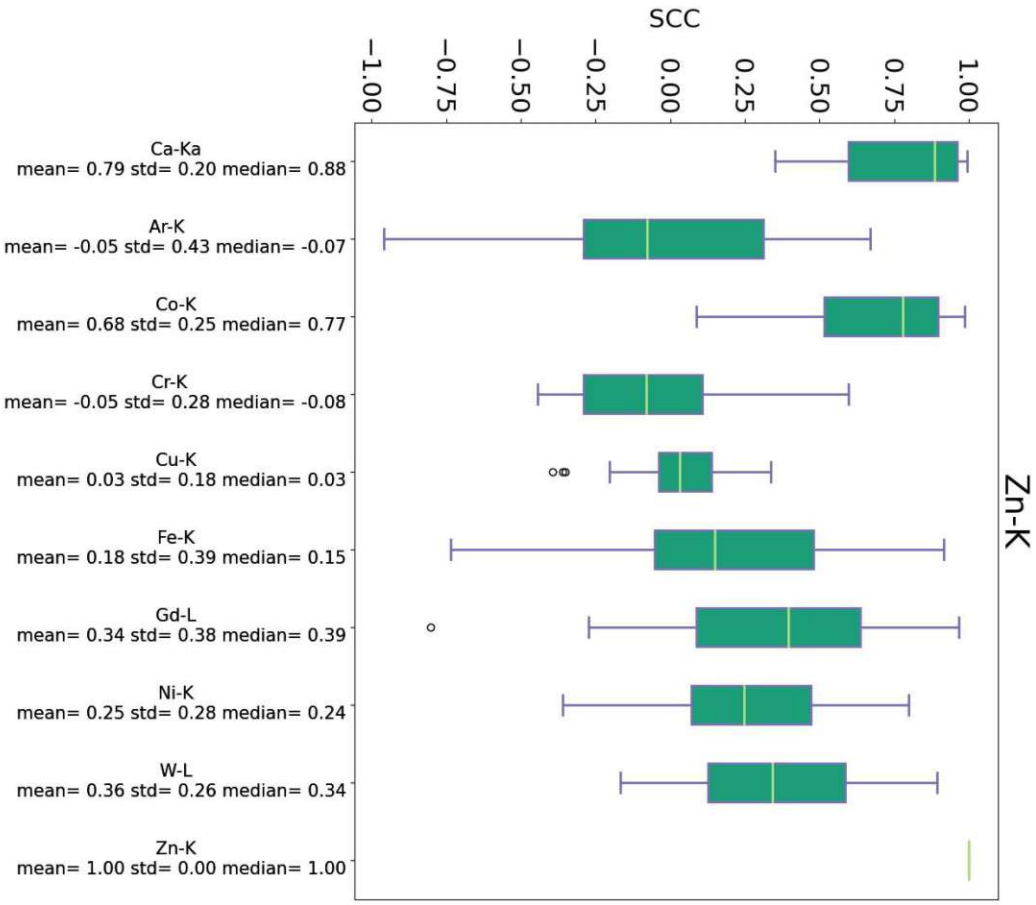

Figure 48: Monotonic correlation (SCC) between Zn and other elements.

The overlap between Zn and Ca in Figure 49 is 99% for the median (like for Figure 47) and the standard deviation is only 0.1. The overlap between Zn and Fe is 75% and the standard deviation is 0.26. The overlap between Zn and Gd is 58% (median) and the standard deviation is 0.3. The overlap between Zn and Ni is 0.29% (median) and the standard deviation is 0.3. The overlap between Zn and Cr and Cu is 0% for the median. The 0% overlap behaviour can be shown for Zn and Cu as well.

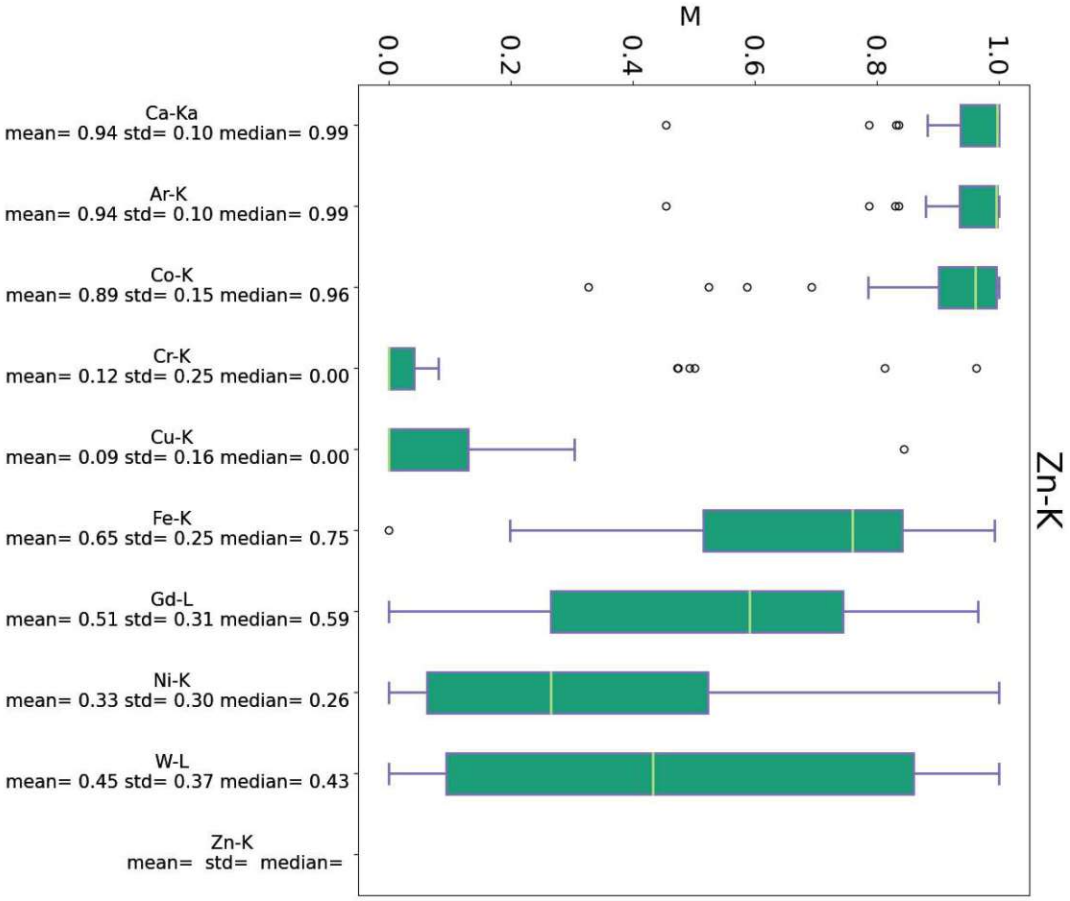

Figure 49: Overlap coefficient (M) between Zn and other elements.

In Figure 50, the monotonic correlation between Ni and other elements is very low, which is between 0 and 0.27 for the median. The standard deviation is very high. In general the medians and the deviations are leaning over more to the positive side.

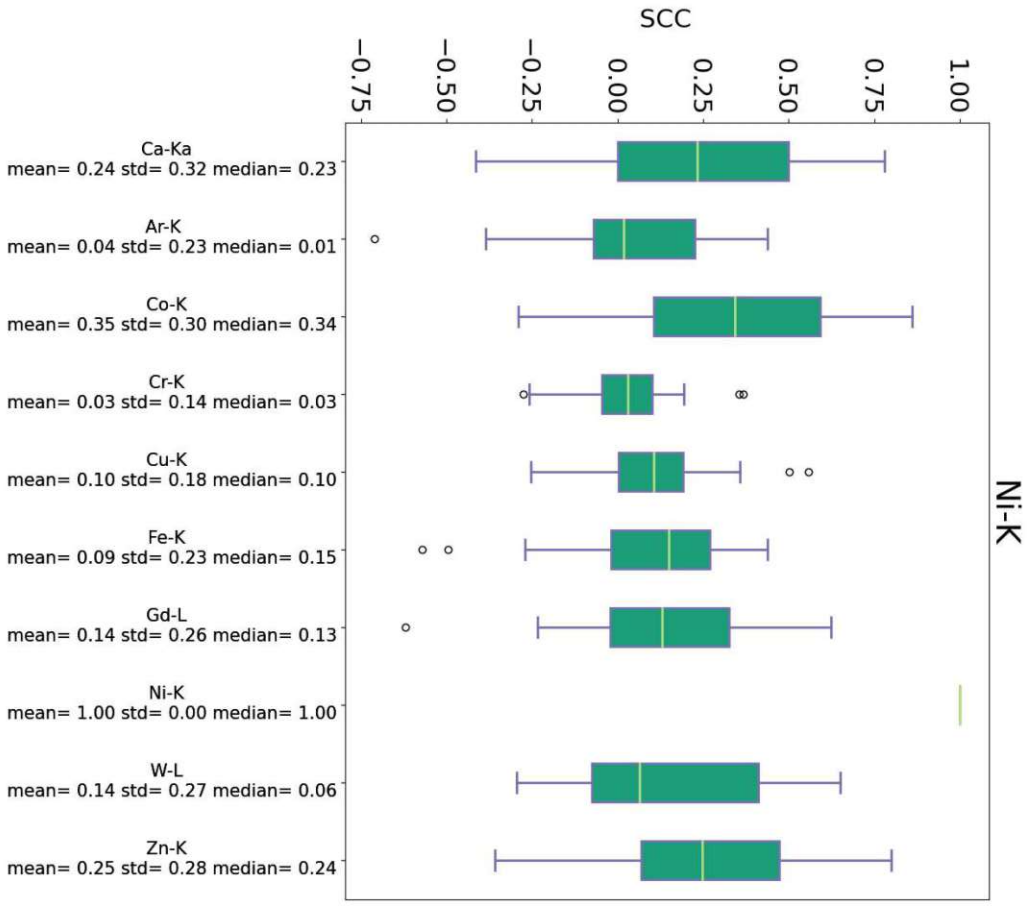

Figure 50: Monotonic correlation (SCC) between Ni and other elements.

The overlap in Figure 51 between Ni and Ca, Fe, Gd and Zn is between 20% and 30% in the median and the standard deviation is high. The overlap between Ni and Cr is 0% for the median, the same for Ni and Cu.

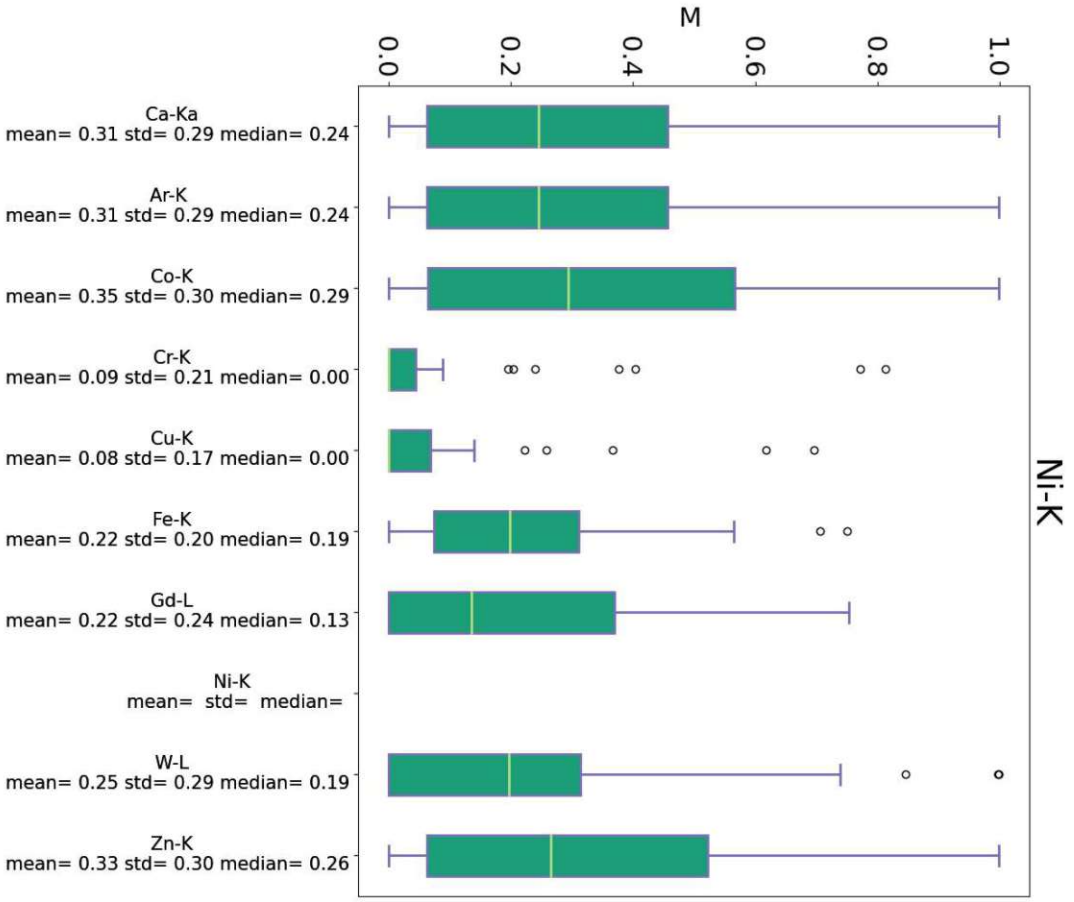

Figure 51: Overlap coefficient (M) between Ni and other elements.

The monotonic correlation between Fe and all other elements in Figure 52 is very low (between -0.04 and 0.2 for the median). The standard deviation is high and reaches the positive and the negative side. Gd shows a small divergence about a higher correlation and a distribution more on the positive side.

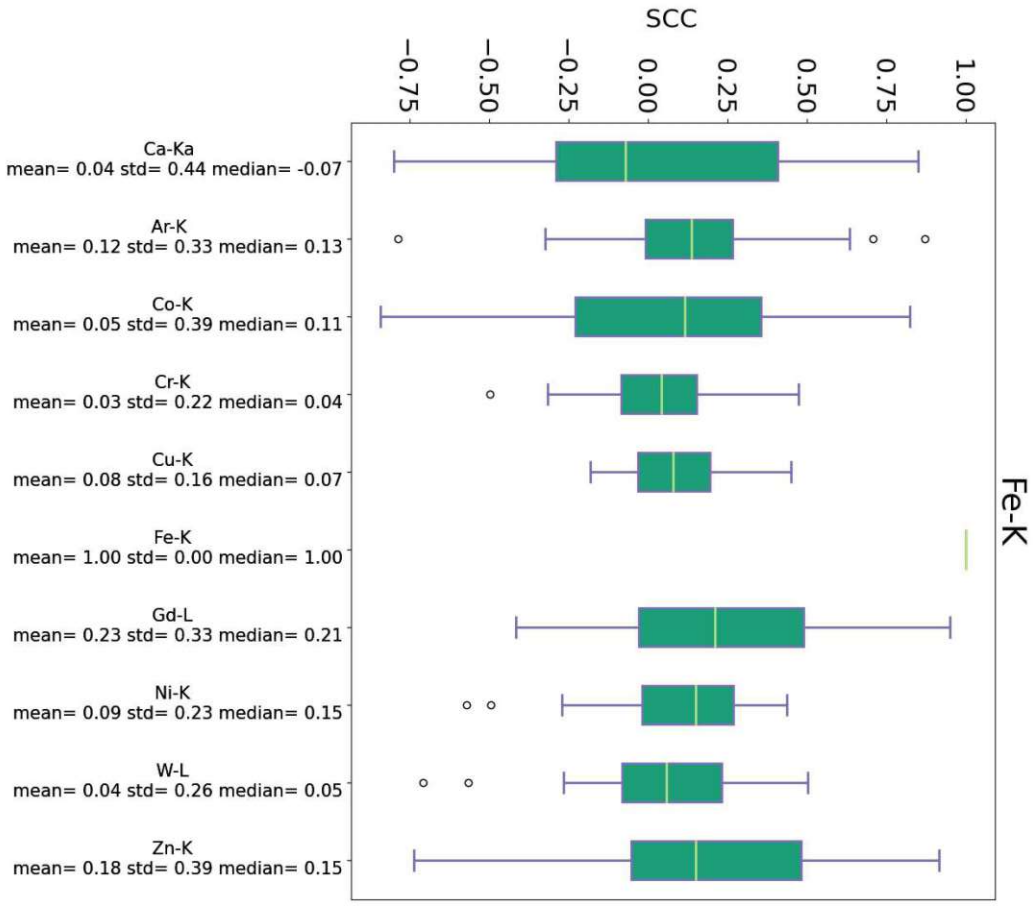

Figure 52: Monotonic correlation (SCC) between Fe and other elements.

The overlap between Fe and Ca in Figure 53 is 76% (median) and the standard deviation is 0.26. The overlap between Fe and Zn is 75% (median) and the standard deviation is 0.26. The overlap between Fe and Gd is 47% (median) and the standard deviation is very high (0.28). The overlap between Fe and Ni is low (20% in median). The overlap between Fe and Cr and Cu is 0% (median), which indicates a high noise level for the Cr and Cu signal.

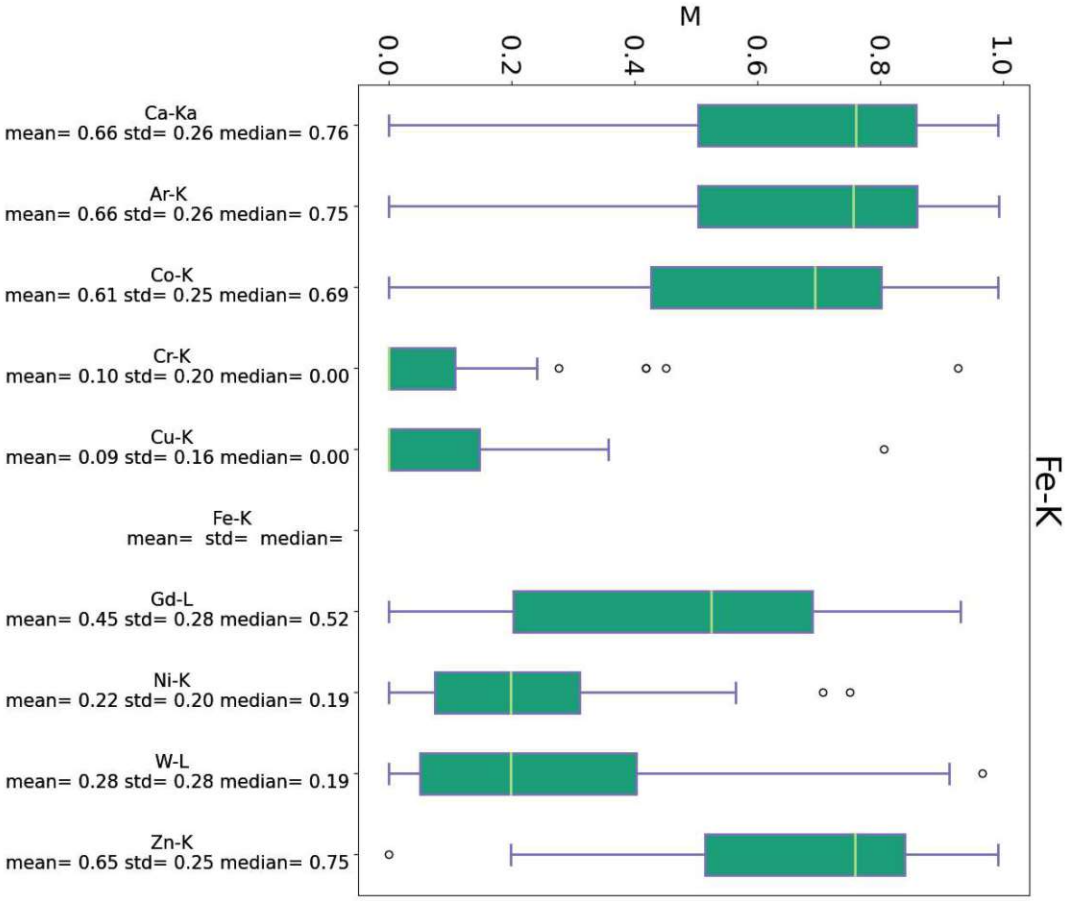

Figure 53: Overlap coefficient (M) between Fe and other elements.

In Figure 54, the monotonic correlation between Gd and Zn is 0.42 for the median and the standard deviation is 0.39. The correlation between Gd and all other elements is very low (between 0.07 and 0.33) and the deviation is very high and also negative (between 0.23 and 0.39). The data distribution is leaning over more on the positive side.

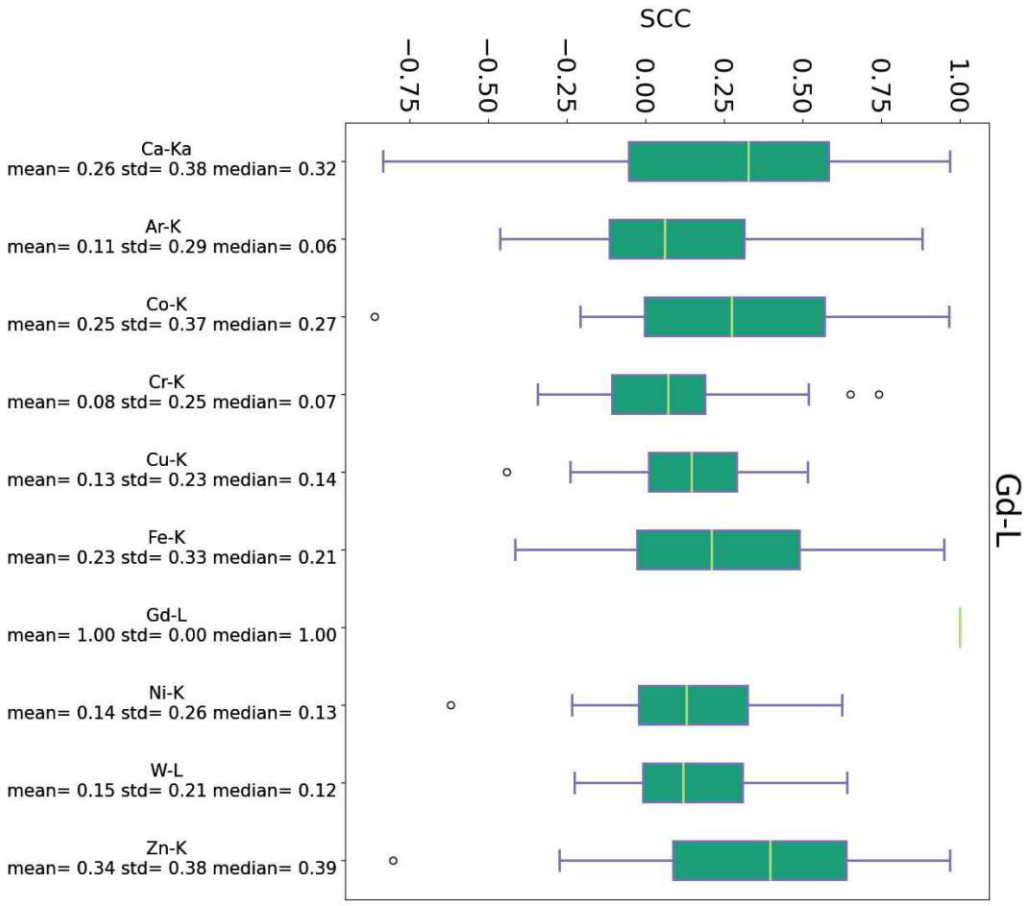

Figure 54: Monotonic correlation (SCC) between Gd and other elements.

The overlap between Gd and Ca in Figure 55 is 54% (median) and the standard deviation is 0.29. The overlap between Gd and Zn is 58% (median) and the standard deviation is 0.3. The overlap between Gd and Fe is 47% (median) and the standard deviation is 0.28. The overlap between Gd and Ni is low (23% in median, standard deviation = 0.23). The overlap between Gd and Cr respectively Cu is 0% (median), which indicates a high noise level for the Cr and Cu signal.

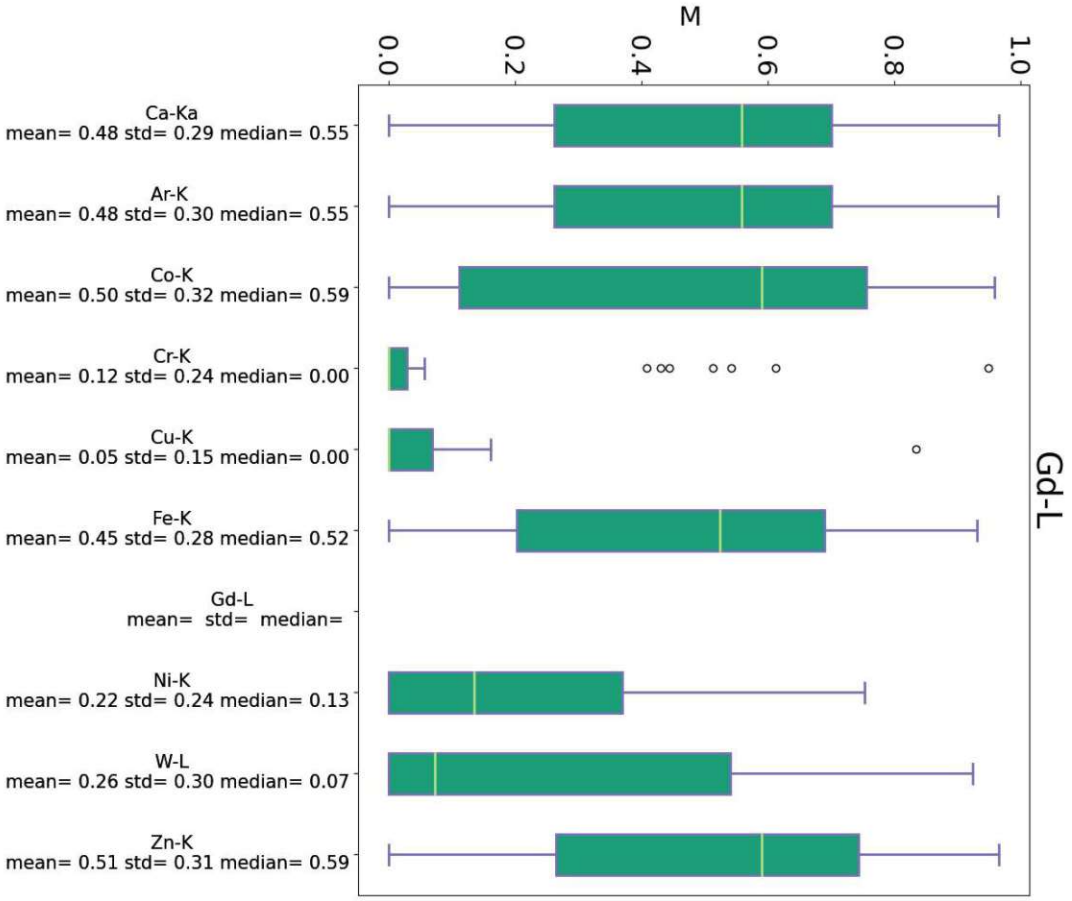

Figure 55: Overlap coefficient (M) between Gd and other elements.

4.3 Measurement for Two Groups (GD1 and GD5)

To make the deviations between two groups (GD1 and GD5) visible, box plots were set for data interpretation. Each element is compared to all other elements for the coefficients: PCC, SCC, Slope, M1, M2 and M, which forms a set of 42 images. The distribution of one element for group GD1 is built by 7 single scan locations (n=7) and for group GD5 by 15 single scan locations (n=15). The main results are presented at the end of these chapter. The graphs for PCC, Slope, M1 and M2 are listed in section 7.3. For the following explanations, the graphs for the coefficients SCC and M are presented.
The element combination Ca-Ni in Figure 56 shows a significant difference between GD1 and GD5 (p-value= 0.01) where GD1 shows a significant higher monotonic correlation compared to GD5. For Ca and Cu, GD1 shows also a higher monotonic correlation (p-value= 0.03) compared to GD5. For all other elements, no significant differences could be identified.

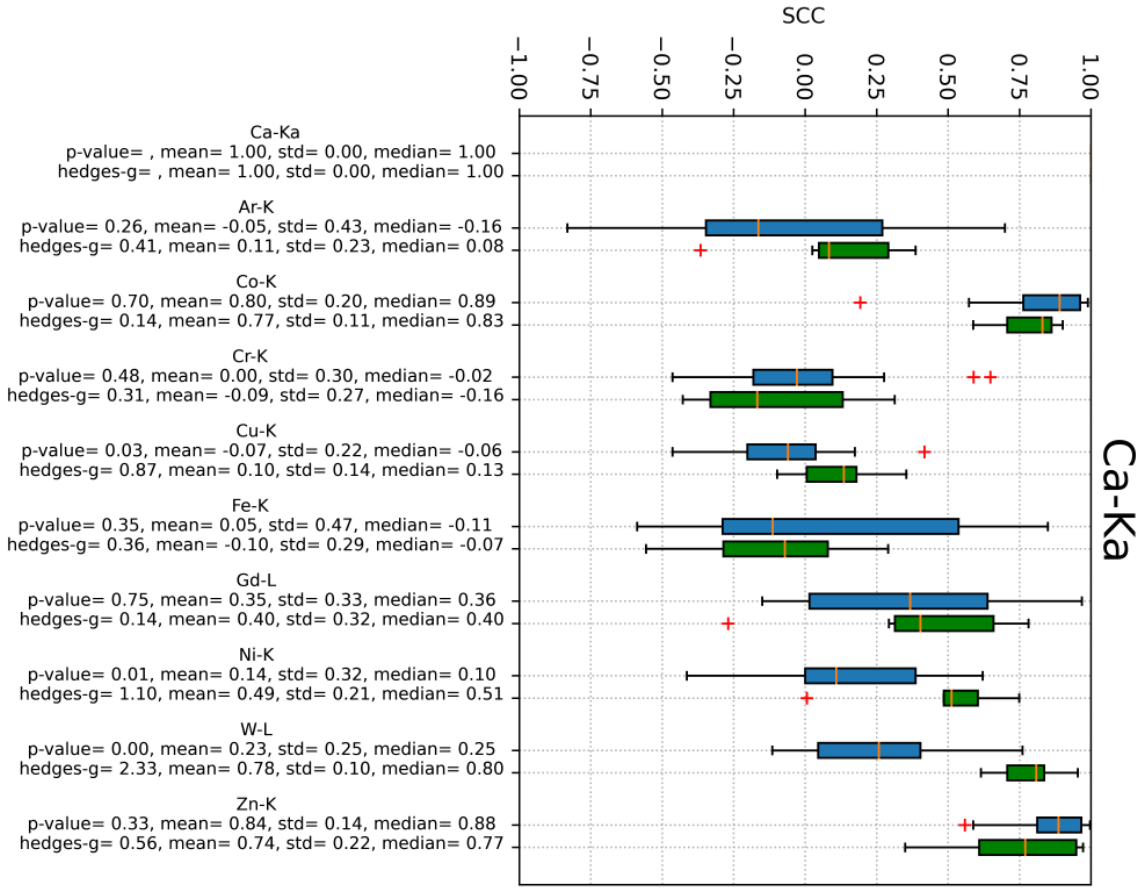

Figure 56: Monotonic correlation (SCC) between Ca and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

GD5 shows a significance difference (p-value= 0.01) to GD1 in Figure 57 for the element combination Ca and Fe. GD5 shows a higher overlap than GD1. The Ca-Ni interaction shows a significant higher overlap for GD1 compared to GD5 (p-value = 0.05). For all other elements, no significant differences between GD1 and GD5 could be found.

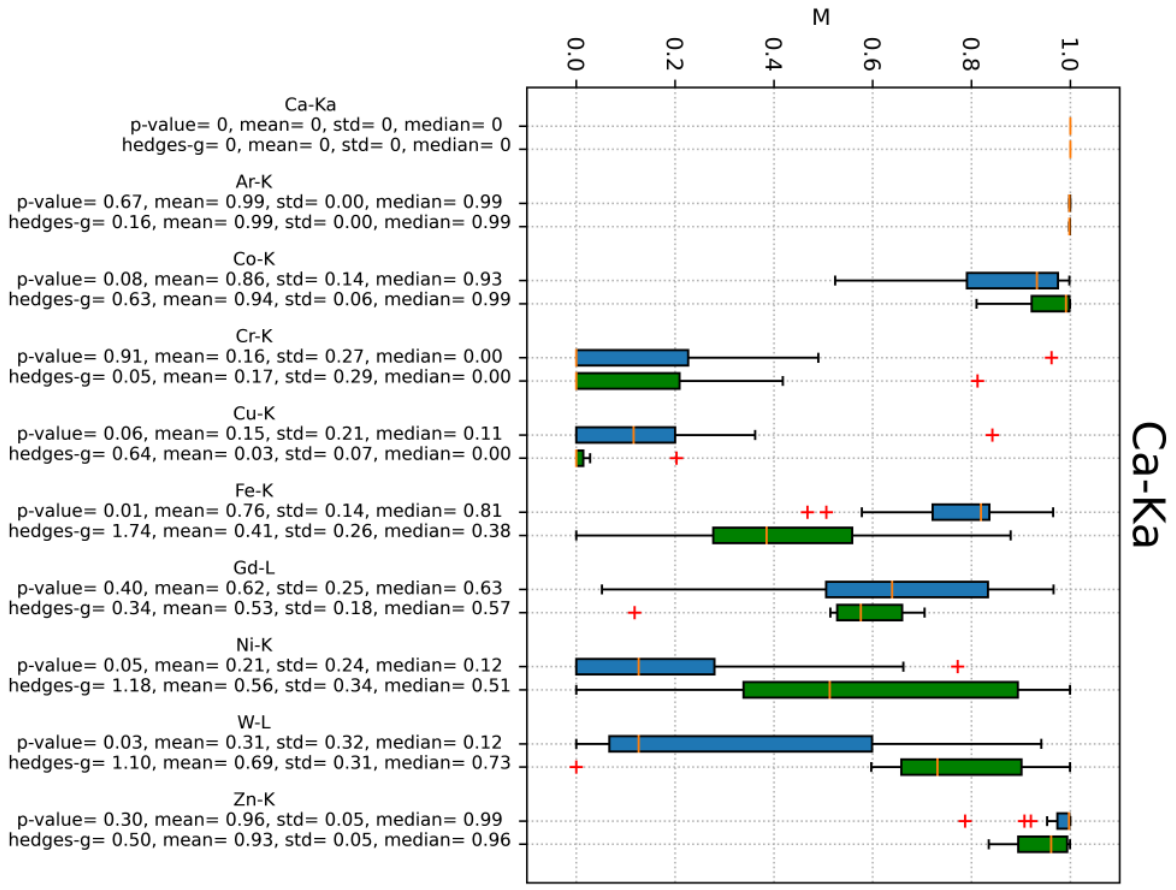

Figure 57: Overlap coefficient (M) between Ca and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The element combination Zn and Ni in Figure 58 shows a significant difference between GD1 and GD5 (p-value= 0.03), where GD1 shows a higher monotonic correlation. For Zn and Cu, GD1 also shows a higher monotonic correlation compared to GD5 (p-value= 0.01). The Zn and Fe ensemble shows an indicated higher overlap for GD5 compared to GD1 (p-value = 0.08). For all other elements, no significant differences were found.

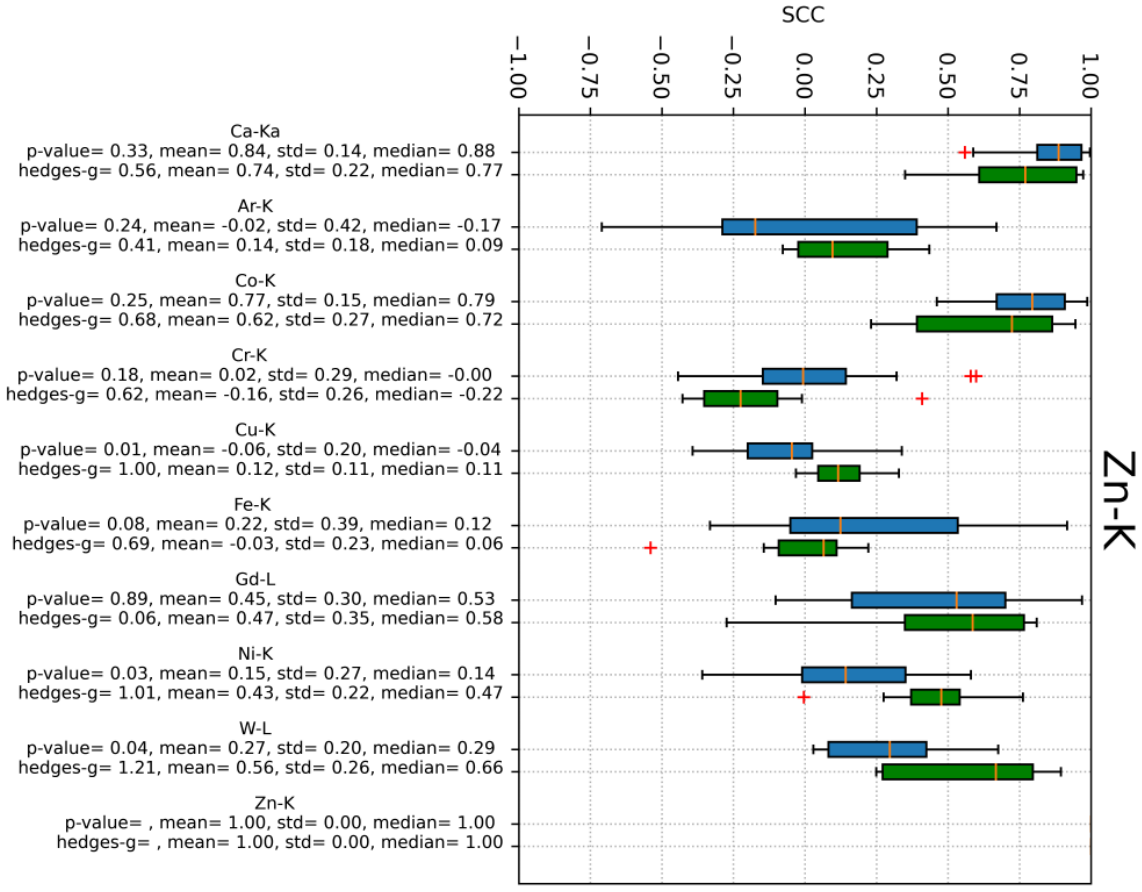

Figure 58: Monotonic correlation (SCC) between Zn and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

In Figure 59 GD5 shows a significant difference (p-value= 0.04) to GD1, for the element combination Zn and Ni, the overlap GD1 was located higher than that of GD5. The Zn-Fe interaction shows a significant difference, which shows an higher overlap for GD5 compared to GD1 (p-value = 0.01). For all other elements, no significant differences between GD1 and GD5 were found.

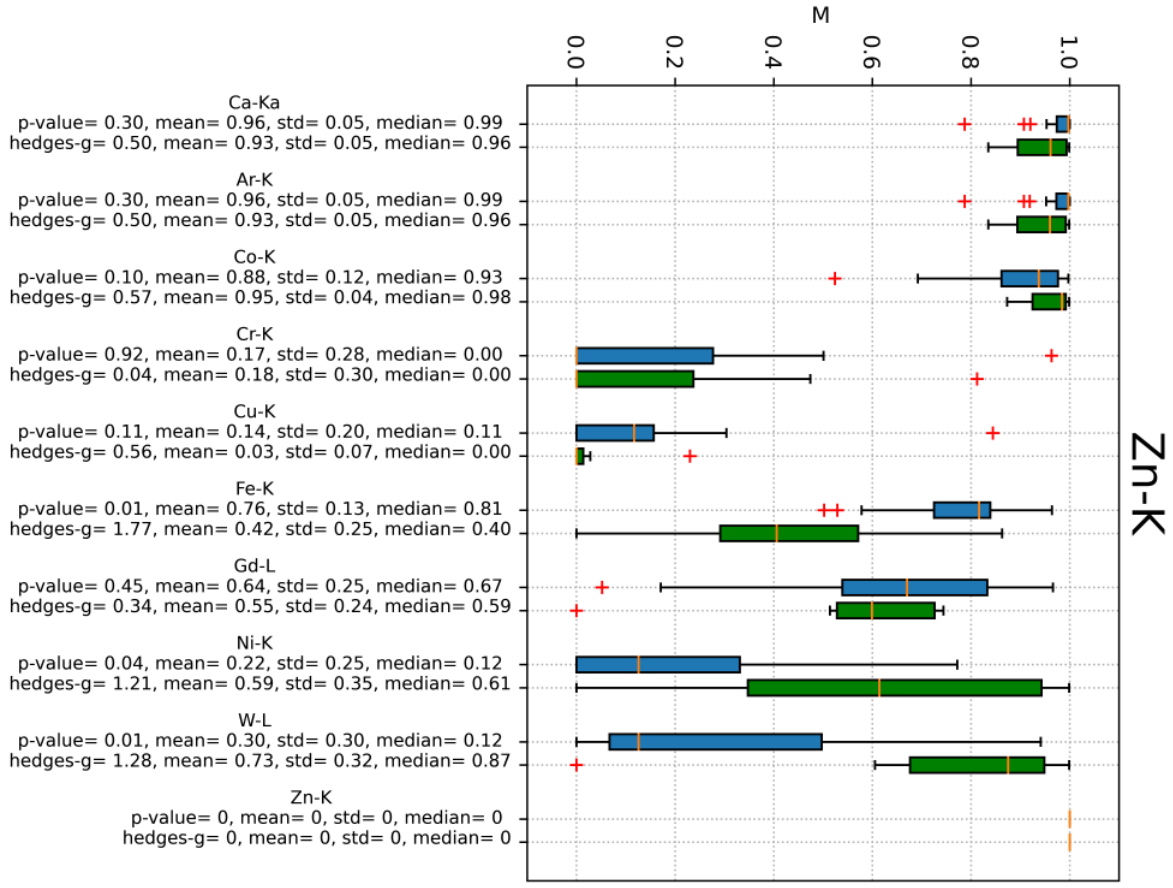

Figure 59: Overlap coefficient (M) between Zn and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The element combination Ni-Ca in Figure 60 shows a significant difference between GD1 and GD5 (p-value= 0.01) where GD1 shows a significant higher monotonic correlation compared to GD5 (the same in Figure 56). For Ni and Zn, GD1 shows also a higher monotonic correlation (p-value= 0.03) compared to GD5. For all other elements, no significant differences were found.

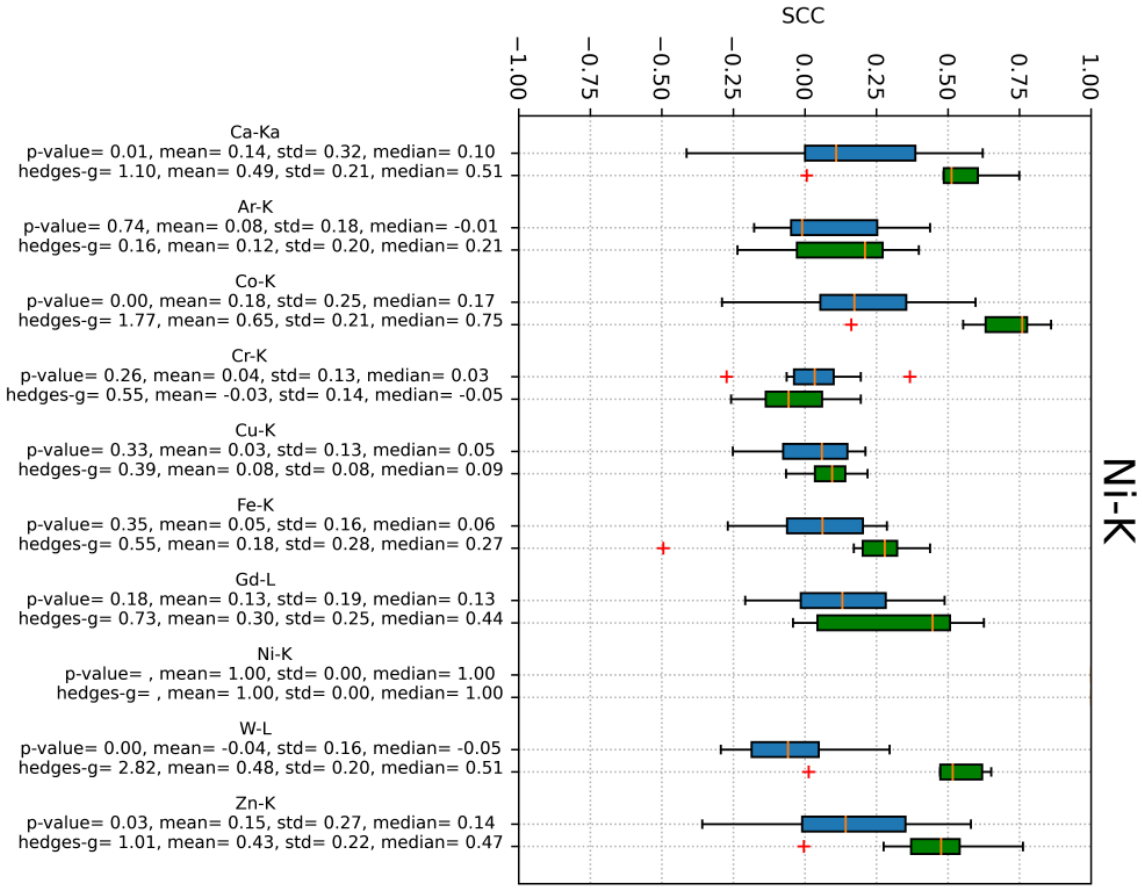

Figure 60: Monotonic correlation (SCC) between Ni and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

GD1 shows a significant difference (p-value= 0.04) to GD5 in Figure 61, for the element combination Ni and Zn. GD1 shows a higher overlap compared to GD5. For the element ensemble Ni-Gd, a high leaning overlap for GD1 compared to GD5 was observed. The Ni-Ca interaction shows a higher overlap for GD1 compared to GD5 (p-value = 0.05). For all other elements, no significant differences between GD1 and GD5 were found.

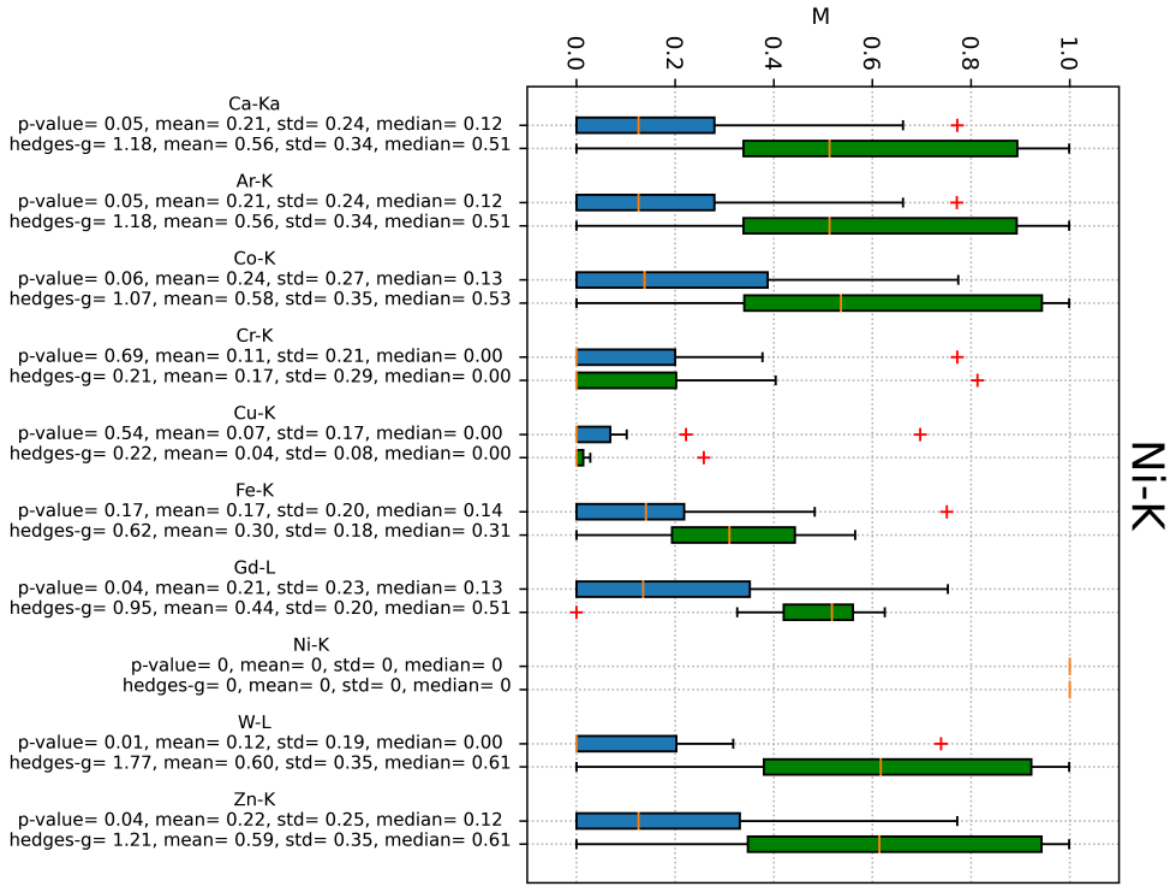

Figure 61: Overlap coefficient (M) between Ni and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The element combination Fe-Zn in Figure 62 shows a suspected difference between GD1 and GD5 (p-value= 0.08) where GD5 shows a higher monotonic correlation in compare to GD1. No significant differences were found for the other elements.

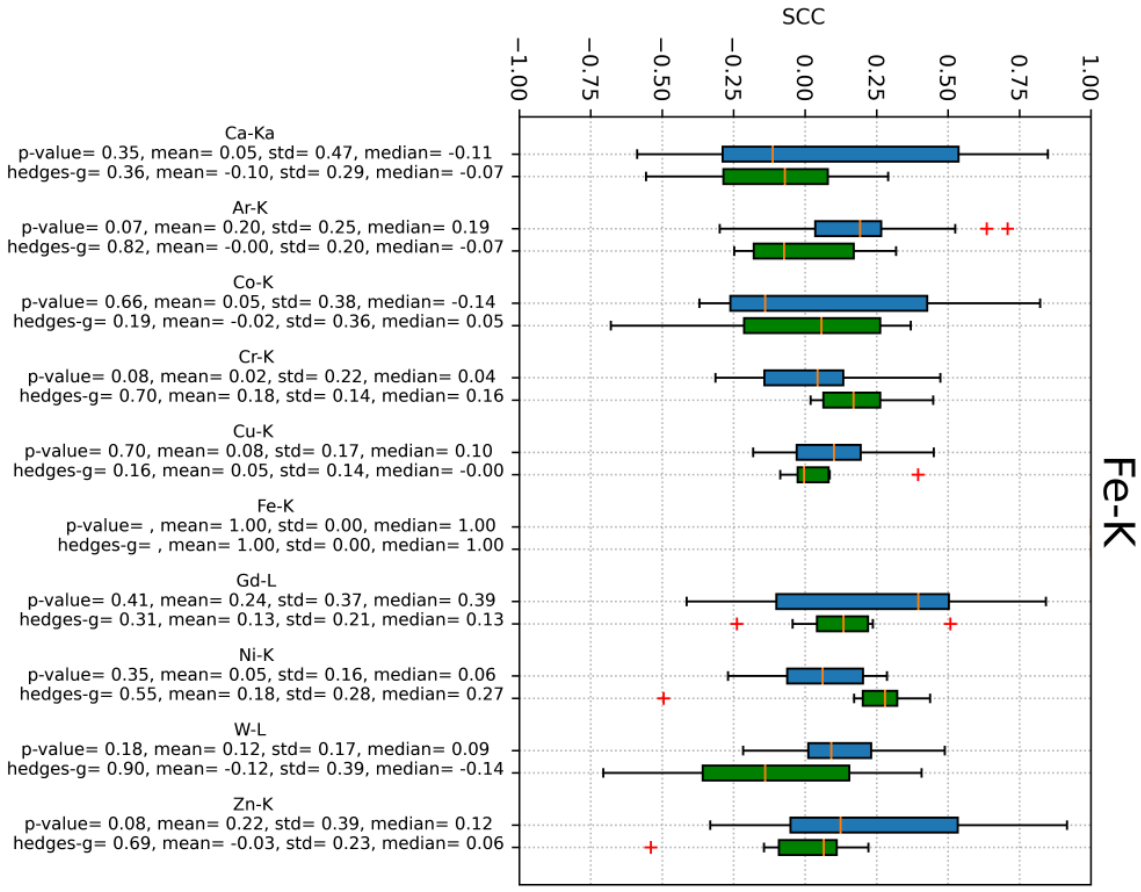

Figure 62: Monotonic correlation (SCC) between Fe and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

In Figure 63 GD1 shows a significant difference (p-value= 0.01) to GD5, for the element combination Fe and Ca (same in Figure 57), where GD5 shows a higher overlap than GD1. The Fe-Gd combination shows for GD5 a higher overlap than for GD1 (p-value= 0.02). GD5 has a higher overlap compared to GD1, for the element ensemble Fe and Zn (p-value= 0.01). For the elements Cr and Cu no significant differences between GD1 and GD5 were observed (Ar and Co are not investigated). The same behaviour for Fe is shown in the appendix (section 7.3) for the Fe M2-coefficient.

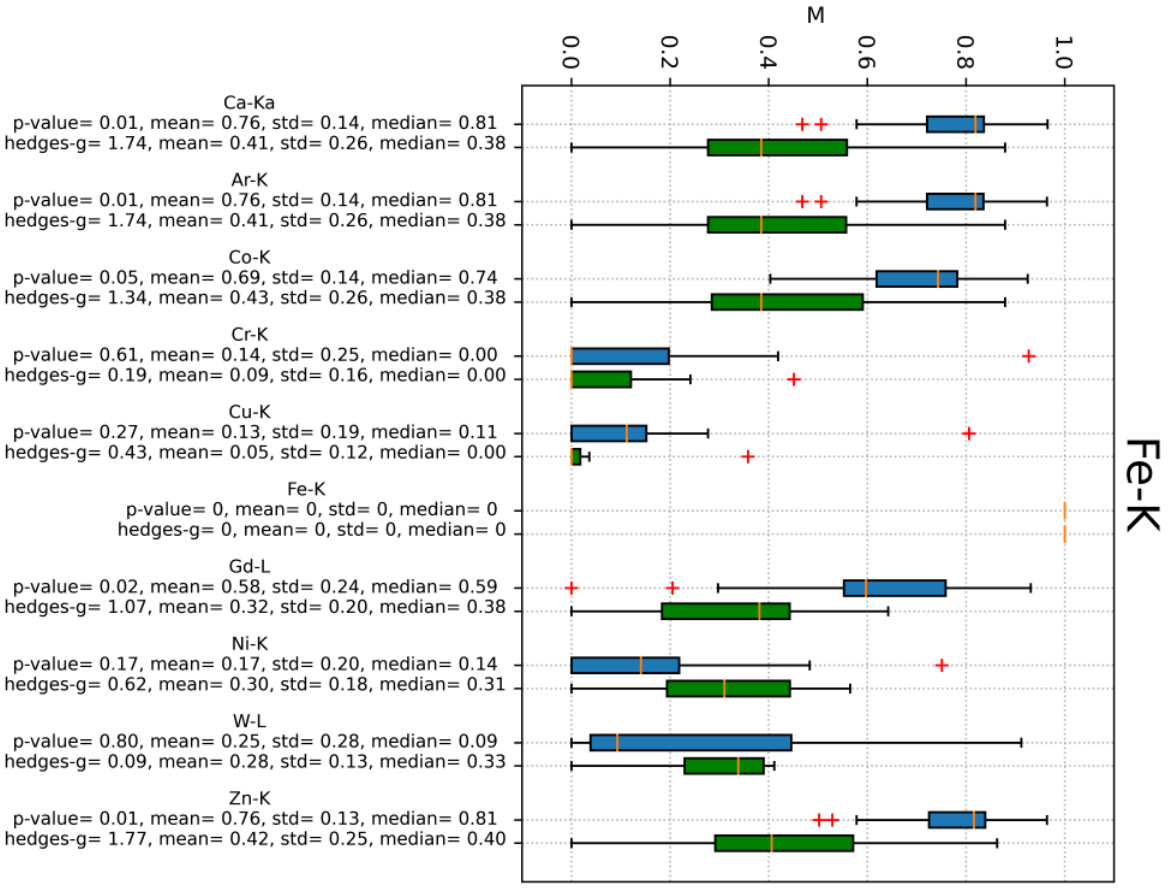

Figure 63: Overlap coefficient (M) between Fe and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The ensemble in Figure 64 for Gd and Cu shows a higher overlap for GD1 in compare to GD5 (p-value = 0.04). For all other elements, no significant differences could be shown.

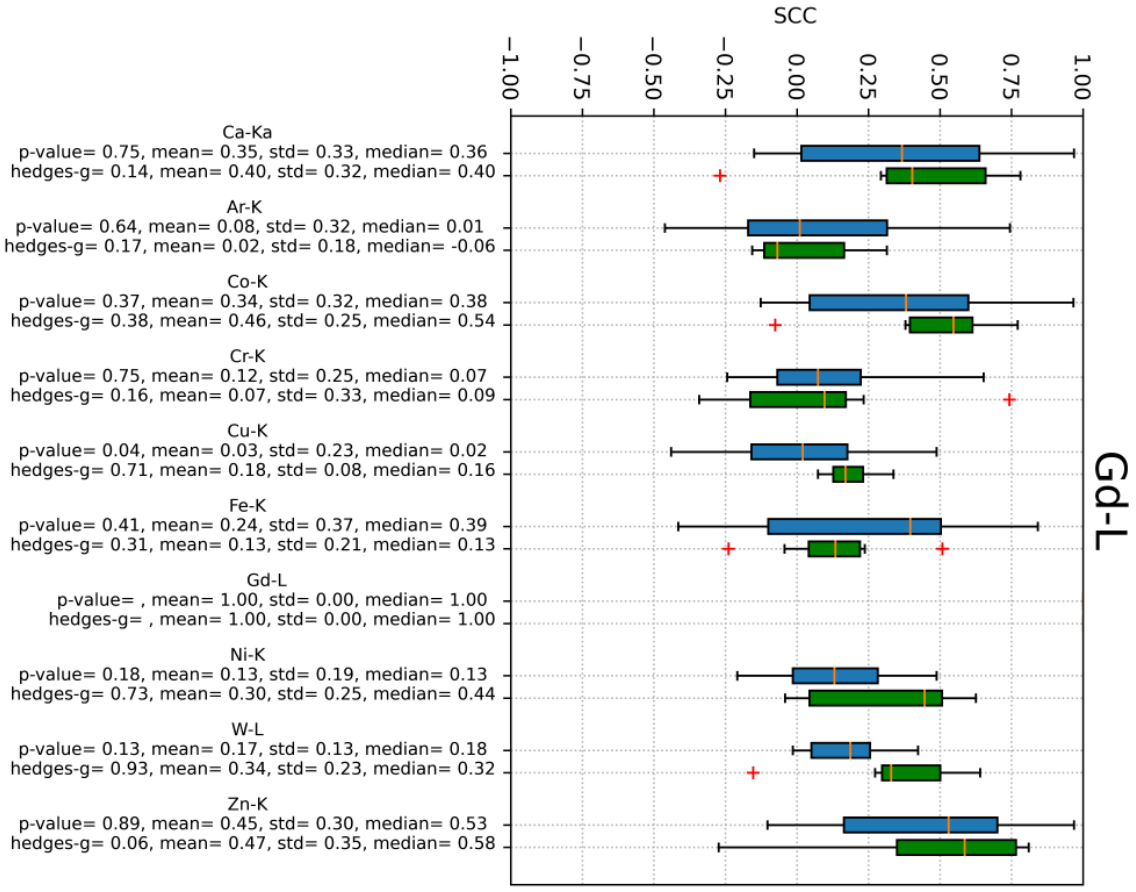

Figure 64: Monotonic correlation (SCC) between Gd and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The Gd-Fe combination in Figure 65 shows a higher overlap for GD5 compared to GD1 (p-value= 0.02) same in Figure 63. For the element ensemble Gd-Ni, a higher overlap for GD1 compared to GD5 could be shown (same for Figure 61). For all other elements, no significant differences between GD1 and GD5 could be shown.

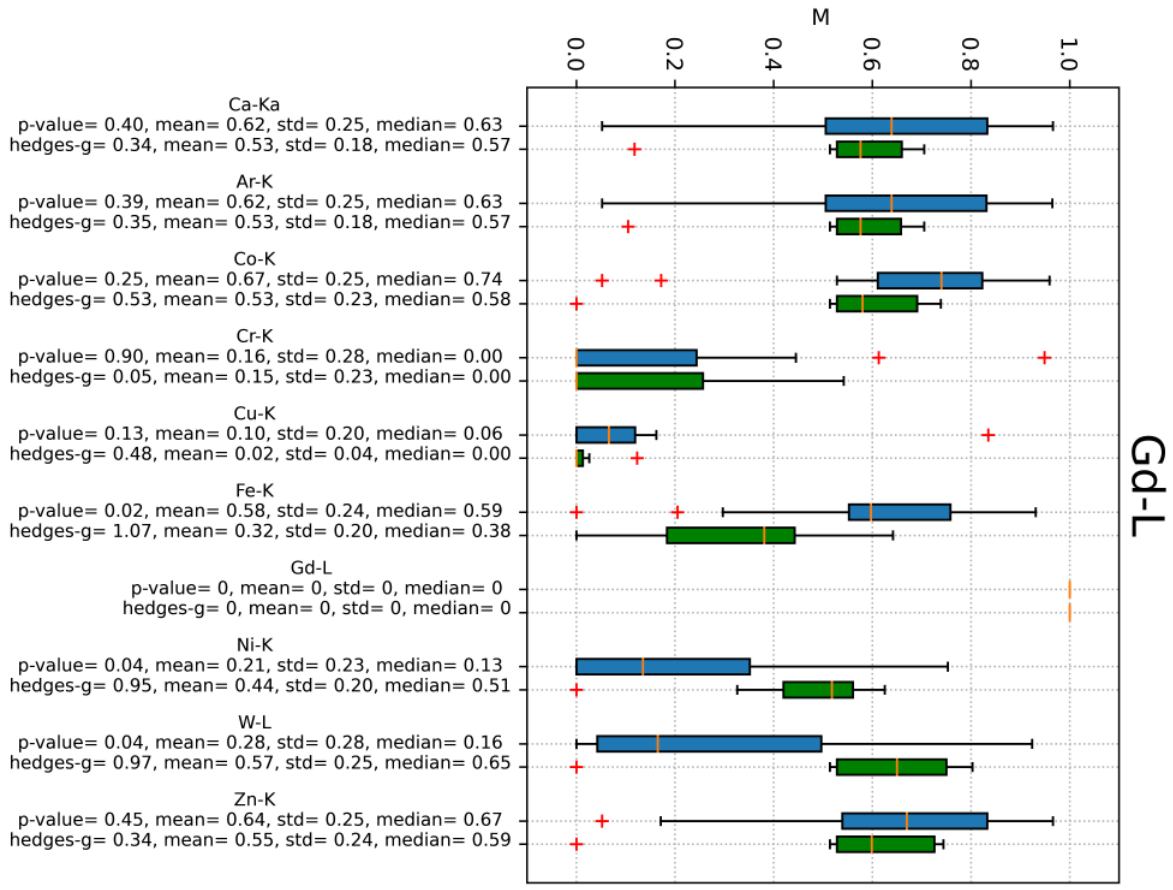

Figure 65: Overlap coefficient (M) between Gd and other elements, for the groups GD1 and GD5 in comparison. The blue bar describes GD5 and the green bar describes GD1.

The following graphs depict the overlap between element combinations by using the median values of the box plots. The graphs show the correlation values and the overlap coefficients corresponding to the median value. The presented images illustrate only the overlap coefficients. It should serve as a general overview to see the approximated overlap behaviour in combination to all other coefficients. Only the elements of interest Ca, Zn, Ni, Fe and Gd are shown. The red marked entries refer to a significant difference between GD1 and GD5.
Figure 66 presents the overlap behaviour between Ca and other elements of interest. Ca is represented by the color blue and the compared elements (Fe, Gd, Ni and Zn) are represented by the color red. The overlap area between Ca and the compared elements is described through the color magenta.

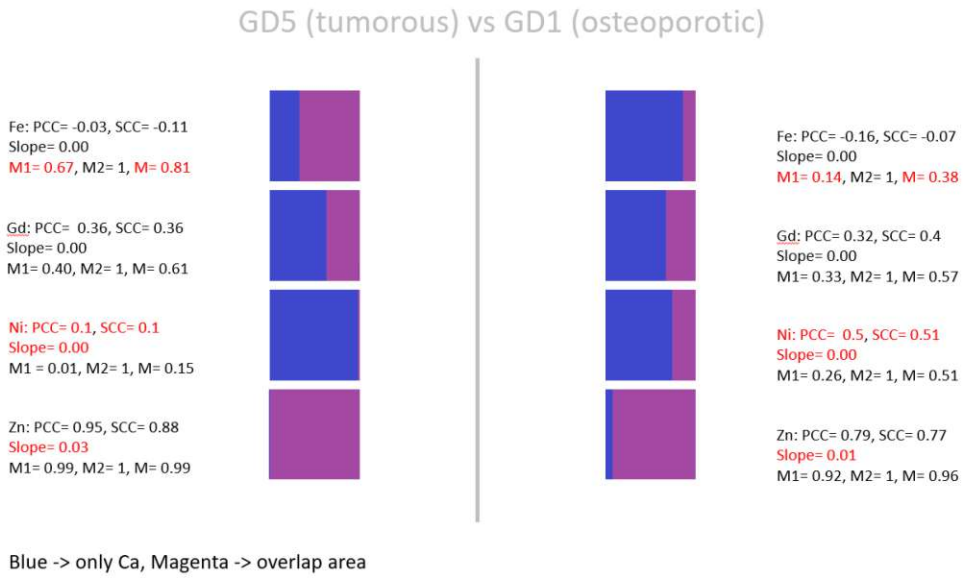

Figure 66: Visualization of the overlap, to compare GD1 (right) and GD5 (left) by using the median value. The red marked coefficients implement a significant difference between GD1 and GD5. Blue: pure Ca, red: pure Fe, Gd, Ni or Zn (up to down), magenta: overlap region.

Figure 67 presents the overlap behaviour between Gd and other elements of interest. Gd is represented by the color blue and the compared elements (Fe, Ni and Zn) are represented by the color red. The overlap area between Gd and the compared elements is described through the color magenta.

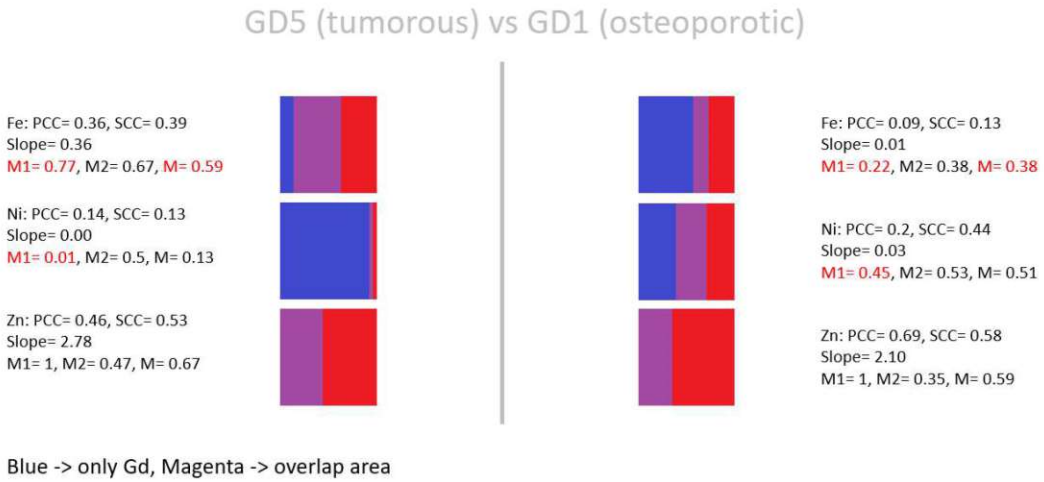

Figure 67: Visualization of the overlap, to compare GD1 (right) and GD5 (left) by using the median value. The red marked coefficients implement a significant difference between GD1 and GD5. Blue: pure Gd, red: pure Fe, Ni or Zn (up to down), magenta: overlap region.

Figure 68 presents the overlap behaviour between Fe and other elements of interest. Fe is represented by the color blue and the compared elements (Ni and Zn) are represented by the color red. The overlap area between Fe and the compared elements is described through the color magenta.

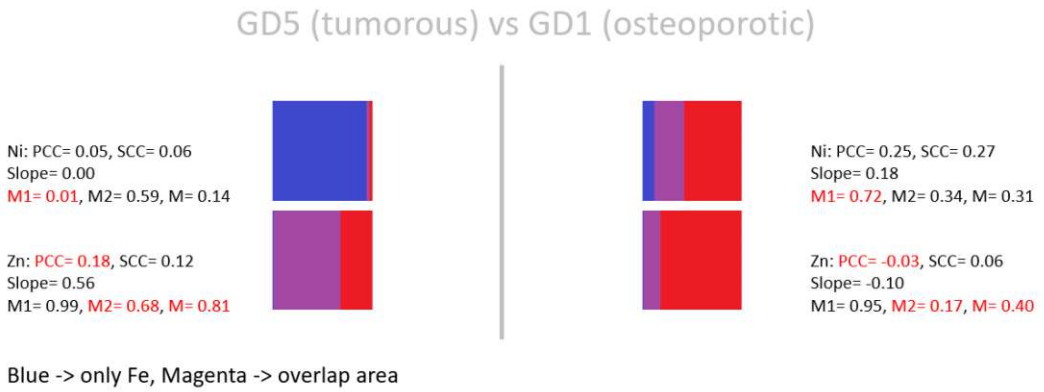

Figure 68: Visualization of the overlap, to compare GD1 (right) and GD5 (left) by using the median value. The red marked coefficients implement a significant difference between GD1 and GD5. Blue: pure Fe, red: pure Ni or Zn (up to down), magenta: overlap region.

Figure 69 presents the overlap behaviour between Ni and Zn. Ni is laid out by the color blue and Zn is represented by the color red. The overlap area between Ni and Zn is described through the color magenta.

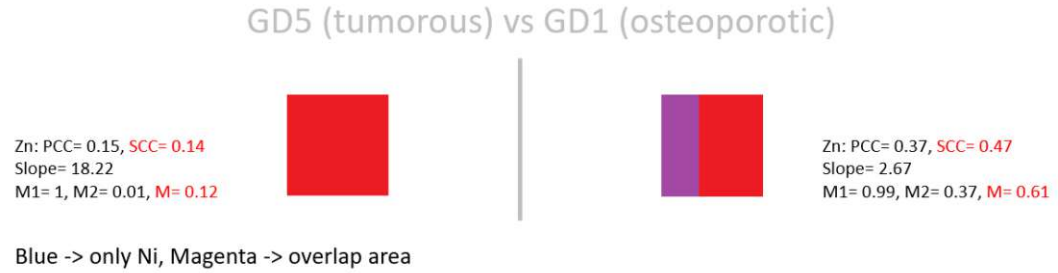

Figure 69: Visualization of the overlap, to compare GD1 (right) and GD5 (left) by using the median value. The red marked coefficients implement a significant difference between GD1 and GD5. Blue: pure Ca, red: pure Zn, magenta: overlap region.

5 Discussion

Interpretation

We only found one overlap image where Gd is located without Zn (Figure 45). It can be seen as abnormal behaviour, because the statistic shows Gd overlaps Zn to nearly 100%.

For the PCC and the SCC the following statements can be made:

-) A high linear correlation, in average, between Ca and Zn (approximated 0.8) was found.
-) A small linear correlation, in average, between Gd and Zn (approximated 0.5) could be found.
-) A monotonic correlation, which is stronger than the linear correlation, could not be observed.

For the overlap coefficients M1, M2 and M the following conclusions could be found:

-) A high co-localization between the elements Ca and Zn could be found.
-) Ca overlaps Fe to nearly 60% but the deviations is also very high.
-) Fe, Gd, Ni and Zn overlap Ca to nearly 100%. Cr and Cu do not overlap Ca (perhaps noise).
-) Ca, Fe, Gd and Ni overlap Zn to nearly 100% in average. Cr and Cu do not overlap Zn (perhaps noise).
-) It could not be shown if Cr or Cu are really present in the samples. Noise can cause this behaviour.

For the PCC, SCC and slope results, following interpretations could be found between patient GD1 and GD5:

-) Ca and Ni show a significant difference in the PCC ($p = 0.05$). GD1 has higher correlation compared to GD5 between Ca and Ni.
-) Ca and Ni show a significant difference in the SCC ($p = 0.01$). GD1 has higher correlation compared to GD5 between Ca and Ni.
-) Cr and Ni show a significant difference in the PCC ($p = 0.05$). GD5 has higher correlation compared to GD1 between Cr and Ni.
-) Ca and Ni shows for the Slope a p-value of 0.04. The Ni increase with Ca is higher for GD1 compared to GD5.
-) Ca and Cu show a significant difference in the SCC ($p= 0.03$). GD1 has higher correlation compared to GD5 between Ca and Cu.
-) Cu and Zn show a significant difference in the PCC ($p = 0.04$). GD1 has higher correlation compared to GD5 between Cu and Zn.
-) Cu and Zn show a significant difference in the SCC ($p = 0.01$). GD1 has higher correlation compared to GD5 between Cu and Zn.
-) Cu and Gd show a significant difference in the PCC ($p = 0.03$). GD1 has higher correlation compared to GD5 between Cu and Gd.
-) Cu and Gd show a significant difference in the SCC ($p = 0.04$). GD1 has higher correlation compared to GD5 between Cu and Gd.
-) Cu and Ni show a significant difference in the Slope ($p = 0.05$). The Ni increase in combination to Cu is higher for GD5 in compare to GD1.
-) Fe and Zn show a significant difference in the PCC ($p= 0.03$). GD5 has higher correlation compared to GD1 between Fe and Zn.
-) Ni and Zn show a significant difference in the SCC ($p= 0.03$). GD1 has higher correlation compared to GD5 between Ni and Zn.
-) Ni and Zn shows for the Slope a p-value of 0.01. The Zn increase with Ni is higher for GD5 compared to GD1.
-) Ca and Zn shows for the Slope a p-value of 0.00. The Zn increase with Ca is higher for GD5 compared to GD1.

For the overlap coefficient M, the following element combinations are significant different between patient GD1 and GD5:

-) Ca and Fe: For GD5, the bilateral overlap M is 43% higher in compare to GD1.
-) Ca and Ni: For GD5, the bilateral overlap M is 39% higher in compare to GD1.
-) Fe and Gd: For GD5, the bilateral overlap M is 21% higher in compare to GD1.
-) Ni and Gd: For GD5, the bilateral overlap M is 38% higher in compare to GD1.
-) Fe and Zn: For GD5, the bilateral overlap M is 41% higher in compare to GD1.
-) Ni and Zn: For GD5, the bilateral overlap M is 49% lower in compare to GD1. The co-localization is significant higher for GD1 in compare to GD5.

For the overlap coefficients M1 and M2, the following element combinations are significant different between patient GD1 and GD5:

-) Ca overlaps Fe: For GD5, Ca overlaps Fe to 53% more (median), in compare to GD1. Fe overlaps Ca to 100% for GD1 and GD5. The co-localization is significant higher for GD5 in compare to GD1.
-) Fe overlaps Ni: For GD5, Fe overlaps Ni to 71% less (median), in compare to GD1. The co-localization is significant higher for GD1 in compare to GD5.
-) Gd overlaps Fe: For GD5, Gd overlaps Fe to 55% more (median), in compare to GD1. The co-localization is significant higher for GD5 in compare to GD1.
-) Gd overlaps Ni: For GD5, Gd overlaps Ni to 44% less (median), in compare to GD1. The co-localization is significant higher for GD1 in compare to GD5.
-) Zn overlaps Fe: For GD5, Zn overlaps Fe to 51% more (median), in compare to GD1. The co-localization is significant higher for GD5 in compare to GD1.

Implications

The abnormal occurrence of one single Gd spot, where no Zn is located could be found. The elements Fe, Gd, Ni, Zn could not be measured without Ca, for the whole data set. The measurement for Cr and Cu gives insufficient overlap results which would clarify the occurrences of those materials in general.
GD5 (tumorous) shows a significant higher co-localization for Ca-Fe in compare to GD1 (osteoporotic). Which suggests less Fe occurrences for GD1 than for GD5, relative to the Ca occurrences, the relativity of Ca occurrences can be ignored. Due to the special status of Ca compared to all other elements, which is defined by the gapless existence of Ca in each measured pixel for all samples, a reversed interpretation (Ca surfaces for GD1 are bigger than for GD5 and the Fe surfaces stays the same) can be discarded.
The same behaviour exists for the elements Zn and Gd compared to Fe, which supports the Fe absence in GD1.
Furthermore, it has to be noted that no statements about the absolute element occurrences can be made. This means that if an element is located somewhere, it can not determined if the amount of one element is low or high in general. Recording the correlation comparison between GD1 and GD5, we are only able to make difference between the amount of one element relative to other elements. In the case of Ca and Fe, the correlation is not significant different between GD1 and GD5. In general, no correlation is measured between Ca and Fe from a statistical point of view.

Limitations

Cr and Cu to Ca show a contradicting behaviour to that observed in the overlap, where Ca overlaps Cr and Cu to 0% in the median, which represents a problem of Cr and Cu measurements. The problem could be answered with the small count rates of Cr and Cu and the chosen threshold level (10 counts/second), which could delete an important amount of informations. It has to be clarified that there are threshold algorithms which are able to mange huge and small count rates in a effective way to minimize the information loss.
To lend more expressive for the statistics results, the number for single scan locations per patient has to be increased. For this reason, GD2, GD3 and GD4 could not be taken for statistics investigations. Furthermore the localization for the single scans are not considered (type of bone tissue) which means, for statistical evaluations the samples origin could be cortical bone as well as trabecular bone for example.
To make the characteristics between different groups more meaningful, a higher number of different patients is necessary. In our study, we only used one patient for each group to represent a disease (GD1 represents osteoporotic tissue and GD5 represents tumorous tissue). Beyond diseased patient groups, health tissue could serve as a control group for comparison which would improve the results. A further important point is the incomplete data information about the patients (age, sex or how many magnetic resonance imaging investigations are done by Gd based contrast agents) where only the diseases are known.[23]

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6.1 Contact

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7 Appendix

How To

The folder structure has to be adapted. The working folder is MA_ LW_ Gd data selection and the original folder is labeled as MA_ LW_ Gd data selection_ orig. For data generation, execute the following programs in the right order.

1. set_ folders_ v1_ final.py
2. linear_ correlation_ plot_ master_ v5_ final.py
3. box_ plot_ PCC_ SCC_ Slope_ master_ v1_ final.py
4. box_ plot_ PCC_ SCC_ Slope_ GD1_ GD5_ compare_ master_ v1_ final.py
5. Overlap_ macro_ multi_ intensity_ normal_ master_ v4_ final.ijm
6. overlap_ data_ collector_ master_ M_ M1_ M2_ v1_ final.py
7. box_ plot_ MOC_ master_ M_ M1_ M2_ v1_ final.py
8. box_ plot_ MOC_ master_ M_ M1_ M2_ GD1_ GD5_ compare_ v1_ final.py
9. multi_ plot_ all_ data_ v4_ final.py

The whole program code can be considered in the following chapter section 7.1.

7.1 Python Code

```
0
1  import os
2
3
4  os.mkdir("box_plot_MOC_M")
5
6  os.mkdir("box_plot_PCC_SCC")
7
8  os.mkdir("box_plot_MOC_M_GD1_GD5")
9
10 os.mkdir("box_plot_MOC_is_overlaped_M2_GD1_GD5")
11
12 os.mkdir("box_plot_MOC_overlaps_M1_GD1_GD5")
13
14 os.mkdir("box_plot_PCC_SCC_GD1_GD5")
15
16 os.mkdir("PCC_SCC")
17
18 os.mkdir("plots")
19
20 os.mkdir("plots_high_correlation")
21
22 os.mkdir("multi_plot")
23
24 os.mkdir("overlap_log")
25
26 os.mkdir("overlap_all_data_M2")
27
28 os.mkdir("box_plot_MOC_is_overlaped_M2")
29
30 os.mkdir("box_plot_MOC_overlaps_M1")
31
32 os.mkdir("overlap_all_data_M")
33
34 os.mkdir("overlap_all_data_M1")
35
36
37 \begin{lstlisting}[firstnumber=0]
```

```
0  #linear_correlation_plot_maaster_v5_final.py
1  #Lukas Warnung
2  import numpy as np
3  import matplotlib as mpl
4  import matplotlib.pyplot as plt
5  from scipy import stats
6  from scipy.stats import pearsonr
7  from scipy.stats import spearmanr
8  import seaborn as sns
9
10 d={}
11 x=0
```

```
i=0
r=0
k=0
counter=0

synchrotronS = ["Diamond","ESRF"]

for synchrotron in synchrotronS:


        if synchrotron == "Diamond":
            specimens = ["GD1a","GD1b","GD1bsecond","GD2a","GD2b","GD3","GD4","GD5"]
            elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]

        if synchrotron == "ESRF":
            specimens = ["GD1b2","GD4","GD5"]
            elements =["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-Ka","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]

        for specimen in specimens:
            if specimen == "GD1a":
                spots = ["183278","183279","183280"]
            if specimen == "GD1b":
                spots = ["183242"]
            if specimen == "GD1bsecond":
                spots = ["183309"]
            if specimen == "GD2a":
                spots = ["183287","183288"]
            if specimen == "GD2b":
                spots = ["183247","183248"]
            if specimen == "GD3":
                spots = ["183270"]
            if specimen == "GD4" and synchrotron == "Diamond":
                spots = ["183258"]
            if specimen == "GD5" and synchrotron == "Diamond":
                spots = ["183293","183294","183297","183298","183300","183302","183303"]
            if specimen == "GD1b2":
                spots = ["scan10","scan22"]
            if specimen == "GD4" and synchrotron == "ESRF":
                spots = ["18","scan17","scan35"]
            if specimen == "GD5" and synchrotron == "ESRF":
                spots = ["scan18","scan21","scan27","scan28","scan32","scan34","scan37","scan38"]


            for spot in spots:

                if spot == "scan10" or spot == "scan18" or spot == "scan21" or spot == "scan32":
                    maps = "maps"
                elif spot == "scan17" or spot == "scan37" or spot == "scan38":
                    maps = "maps_20s"

                else:
                    maps = "maps_1s"

                    print(spot)

                counter = counter+1
                x=0
                counter_1=0

                for element1 in elements:

                    counter_1=counter_1+1
                    counter_2=0

                    for element2 in elements:

                                counter_2=counter_2+1

                                collectn_1x = np.loadtxt("MA_LW_Gd data
                                     selection/"+synchrotron+"/"+specimen+"/"+spot+'/'+maps+'/'+element1+".txt")
                                collectn_1x = collectn_1x.flatten()

                                collectn_1y = np.loadtxt("MA_LW_Gd data
                                     selection/"+synchrotron+"/"+specimen+"/"+spot+'/'+maps+'/'+element2+".txt")
                                collectn_1y = collectn_1y.flatten()

                                corr,_ = pearsonr(collectn_1x, collectn_1y)
                                corr2, _ = spearmanr(collectn_1x, collectn_1y)

                                slope, intercept, r_value, p_value, std_err = stats.linregress(collectn_1x,collectn_1y)

                                plt.plot(collectn_1x, collectn_1y, 'o', color="green", markersize = "1")

                                plt.plot(collectn_1x, slope*collectn_1x + intercept, color='red')

```

```
                        plt.title(synchrotron+', '+specimen+', '+spot+', '+maps+"\n"+'PCC=%.3f ' % corr+'SCC=%.3f' %
                              corr2+"\n" +'Slope=%.3f' % slope ,size=18)
                        plt.tight_layout()

                        plt.xlabel(element1+' in counts [1/s]',size=22)

                        plt.tight_layout()

                        plt.ylabel(element2+' in counts [1/s]',size=22)

                        plt.tight_layout()

                        plt.savefig('plots/'+synchrotron+'_'+specimen+'_'+spot+'_'+element1+'_vs_'+element2+'_'+maps+'.png')

                        if element1 == "Cu-Ka":
                            element1 = "Cu-K"

                        if element2 == "Cu-Ka":
                            element2 = "Cu-K"

                        file_PCC = open("PCC_SCC/PCC_all_data_"+element1+"_"+element2+".txt","a")
                        file_PCC.writelines(synchrotron+" "+specimen+" "+spot+" ")
                        file_PCC.writelines('Pearsons correlation: %.3f' % corr)
                        file_PCC.writelines("\n")
                        file_PCC.close()

                        file_PCC = open("PCC_SCC/Slope_all_data_"+element1+"_"+element2+".txt","a")
                        file_PCC.writelines(synchrotron+" "+specimen+" "+spot+" ")
                        file_PCC.writelines('Slope: %.3f' % slope)
                        file_PCC.writelines("\n")
                        file_PCC.close()

                        file_SCC = open("PCC_SCC/SCC_all_data_"+element1+"_"+element2+".txt","a")
                        file_SCC.writelines(synchrotron+" "+specimen+" "+spot+" ")
                        file_SCC.writelines('Spearmans correlation: %.3f' % corr2)
                        file_SCC.writelines("\n")
                        file_SCC.close()

                        if element1 == "Cu-K" and synchrotron == "ESRF":
                            element1 = "Cu-Ka"

                        if element2 == "Cu-K" and synchrotron == "ESRF":
                            element2 = "Cu-Ka"

                        if corr > 0.65 or corr2 > 0.65 or corr < -0.65 or corr2 < -0.65:

                            if element1 != element2 and counter_1<counter_2:

                                plt.savefig('plots_high_correlation/'+synchrotron+
                                '_'+specimen+'_'+spot+'_'+element1+'_vs_'+element2+'_'+maps+'.png')

                                if element1 == "Cu-Ka":
                                    element1 = "Cu-K"

                                if element2 == "Cu-Ka":
                                    element2 = "Cu-K"

                                file_PCC_HIGH = open("PCC_SCC/PCC_all_data_high"+element1+"_"+element2+".txt","a")
                                file_PCC_HIGH.writelines(synchrotron+" "+specimen+" "+spot+" ")
                                file_PCC_HIGH.writelines('Pearsons correlation: %.3f' % corr)
                                file_PCC_HIGH.writelines("\n")
                                file_PCC_HIGH.close()

                                file_PCC = open("PCC_SCC/Slope_all_data_high"+element1+"_"+element2+".txt","a")
                                file_PCC.writelines(synchrotron+" "+specimen+" "+spot+" ")
                                file_PCC.writelines('Slope: %.3f' % slope)
                                file_PCC.writelines("\n")
                                file_PCC.close()

                                file_SCC_HIGH = open("PCC_SCC/SCC_all_data_high"+element1+"_"+element2+".txt","a")
                                file_SCC_HIGH.writelines(synchrotron+" "+specimen+" "+spot+" ")
                                file_SCC_HIGH.writelines('Spearmans correlation: %.3f' % corr2)
                                file_SCC_HIGH.writelines("\n")
                                file_SCC_HIGH.close()

                                if element1 == "Cu-K" and synchrotron == "ESRF":
                                    element1 = "Cu-Ka"

                                if element2 == "Cu-K" and synchrotron == "ESRF":
                                    element2 = "Cu-Ka"

                        plt.cla()
                        plt.clf()

        print("------")
```

```
            print(counter)
            print("------")

            if counter == "31":
                print("Done!")
```

```

#box_plot_PCC_SCC_Slope_master_v1_final.py
#Lukas Warnung
import numpy as np
import matplotlib as mpl
import matplotlib.pyplot as plt
from scipy import stats
from scipy.stats import pearsonr
from scipy.stats import spearmanr

d={}
x=0
i=0
r=0
k=0
collectn_1={}
mean_all = {}
std_all = {}
median_all = {}
parameters = ["PCC","SCC","Slope"]

for parameter in parameters:

        elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]
        x=0

        for element1 in elements:

                        y=0

                        for element2 in elements:

                            y=y+1

                            print("-------------")
                            print(element1)
                            print(element2)
                            print("-------------")

                            i=0
                            DATA_array = []
                            DATA_float = 0

                            for i in range(31):
                                with open("PCC_SCC/"+parameter+"_all_data_"+element1+"_"+element2+".txt") as f:

                                    DATA = f.readlines()[i]
                                    DATA = DATA[-7:]
                                    DATA_float = float(DATA)
                                    DATA_array.append(DATA_float)

                            collectn_1[y] = DATA_array
                            mean_all[y] = np.mean(collectn_1[y])
                            std_all[y] = np.std(collectn_1[y])
                            median_all[y] = np.median(collectn_1[y])

                            mean_all[y] = (format(mean_all[y], 'f'))[:-4]
                            mean_all[y] = str(mean_all[y])

                            std_all[y] = (format(std_all[y], 'f'))[:-4]
                            std_all[y] = str(std_all[y])

                            median_all[y] = (format(median_all[y], 'f'))[:-4]
                            median_all[y] = str(median_all[y])

                        data_to_plot = collectn_1[1],collectn_1[2],collectn_1[3],collectn_1[4],
                        collectn_1[5],collectn_1[6],collectn_1[7],collectn_1[8],collectn_1[9],collectn_1[10]

                        fig = plt.figure(1, figsize=(12, 10))

                        ax = fig.add_subplot(111)

                        bp = ax.boxplot(data_to_plot)

                        bp = ax.boxplot(data_to_plot, patch_artist=True)

                        for box in bp['boxes']:
```

```
                            box.set( color='#7570b3', linewidth=2)
                            box.set( facecolor = '#1b9e77' )

                        for whisker in bp['whiskers']:
                            whisker.set(color='#7570b3', linewidth=2)

                        for cap in bp['caps']:
                            cap.set(color='#7570b3', linewidth=2)

                        for median in bp['medians']:
                            median.set(color='#b2df8a', linewidth=2)

                        for flier in bp['fliers']:
                            flier.set(marker='o', color='#e7298a', alpha=0.5)

                        plt.title(element1,size=28)

                        labels_list = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K",
                        "Ca-Ka \n mean= "+mean_all[1]+" std= "+std_all[1]+" median= "+median_all[1],
                        "Ar-K  \n mean= "+mean_all[2]+" std= "+std_all[2]+" median= "+median_all[2],
                        "Co-K \n mean= "+mean_all[3]+" std= "+std_all[3]+" median= "+median_all[3],
                        "Cr-K \n mean= "+mean_all[4]+" std= "+std_all[4]+" median= "+median_all[4],
                        "Cu-K \n mean= "+mean_all[5]+" std= "+std_all[5]+" median= "+median_all[5],
                        "Fe-K \n mean= "+mean_all[6]+" std= "+std_all[6]+" median= "+median_all[6],
                        "Gd-L \n mean= "+mean_all[7]+" std= "+std_all[7]+" median= "+median_all[7],
                        "Ni-K \n mean= "+mean_all[8]+" std= "+std_all[8]+" median= "+median_all[8],
                        "W-L \n mean= "+mean_all[9]+" std= "+std_all[9]+" median= "+median_all[9],
                        "Zn-K \n mean= "+mean_all[10]+" std= "+std_all[10]+" median= "+median_all[10],
                        ]

                        ax.set_xticklabels(labels_list, rotation=90, fontsize=15 )

                        plt.ylabel(parameter,size=22)
                        plt.yticks(size=22)

                        ax.get_xaxis().tick_bottom()
                        ax.get_yaxis().tick_left()

                        fig.savefig('box_plot_PCC_SCC/'+element1+'_'+parameter+'.png', bbox_inches='tight')
                        plt.cla()

                        plt.clf()
```

```
#box_plot_PCC_SCC_Slope_GD1_GD5_compare_master_v1_final.py
#Lukas Warnung
import numpy as np
import matplotlib as mpl
import matplotlib.pyplot as plt
from scipy import stats
from scipy.stats import pearsonr
from scipy.stats import spearmanr
from scipy import *

import pandas as pd
import researchpy
import math

d={}
x=0
i=0
r=0
k=0
collectn_1={}
collectn_2={}
pvalue = {}
hedgesg = {}
p_value = {}
hedges_g = {}
result = {}
stats_11 = {}
stats_12 = {}
stats_21 = {}
stats_22 = {}
stats_13 = {}
stats_23 = {}

parameters = ["PCC","SCC","Slope"]

for parameter in parameters:

            elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]
```

```
x=0

for element1 in elements:

                    y=0

                    for element2 in elements:

                        y=y+1
                        i=0
                        DATA_array = []
                        DATA_float = 0
                        DATA_array_2 = []
                        DATA_float_2 = 0

                        for i in range(31):
                            with open("PCC_SCC/"+parameter+"_all_data_"+element1+"_"+element2+".txt") as f:

                                DATA = f.readlines()[i]
                                patient = DATA.split(" ")
                                DATA = DATA[-7:]
                                DATA_float = float(DATA)
                                patient_x = str(patient[1])

                                if patient_x == " GD1a" or patient_x == " GD1b" or patient_x == " GD1bsecond" or
                                     patient_x == " GD1b2":

                                    DATA_array.append(DATA_float)

                                if patient_x == " GD5":
                                    DATA_array_2.append(DATA_float)

                        collectn_1[y] = DATA_array

                        collectn_2[y] = DATA_array_2

                        mean_patient_1 = np.mean(collectn_1[y])
                        std_patient_1 = np.std(collectn_1[y])
                        n_patient_1 = len(collectn_1[y])
                        median_patient_1 = np.median(collectn_1[y])

                        mean_patient_2 = np.mean(collectn_2[y])
                        std_patient_2 = np.std(collectn_2[y])
                        n_patient_2 = len(collectn_2[y])
                        median_patient_2 = np.median(collectn_2[y])

                        Diff_mean = mean_patient_1-mean_patient_2

                        Z =
                              ((n_patient_1-1)*std_patient_1*std_patient_1)+((n_patient_2-1)*std_patient_2*std_patient_2)
                        N = n_patient_1+n_patient_2-2

                        root = math.sqrt(Z/N)

                        Cohens_d = Diff_mean / root

                        Cohens_abs = abs(Cohens_d)

                        hedges_g = Cohens_abs * (1-(3/(4*(n_patient_1+n_patient_2-9))))

                        result, p_value = stats.ttest_ind(collectn_1[y], collectn_2[y], equal_var = False)

                        print("--------------")
                        print(collectn_1[y])
                        print(collectn_2[y])
                        print("--------------")
                        print("Welch-T-Test:")
                        print(p_value)
                        print("--------------")
                        print("hedges_g:")
                        print(hedges_g)
                        print("--------------")
                        print("--------------")
                        print(y)

                        pvalue[y] = (format(p_value, 'f'))[:-4]
                        pvalue[y] = str(pvalue[y])

                        hedgesg[y] = (format(hedges_g, 'f'))[:-4]
                        hedgesg[y] = str(hedgesg[y])

                        stats_11[y] = (format(mean_patient_2, 'f'))[:-4]
                        stats_11[y] = str(stats_11[y])

                        stats_12[y] = (format(std_patient_2, 'f'))[:-4]
```

```
122                     stats_12[y] = str(stats_12[y])
123
124                     stats_13[y] = (format(median_patient_2, 'f'))[:-4]
125                     stats_13[y] = str(stats_13[y])
126
127                     stats_21[y] = (format(mean_patient_1, 'f'))[:-4]
128                     stats_21[y] = str(stats_21[y])
129
130                     stats_22[y] = (format(std_patient_1, 'f'))[:-4]
131                     stats_22[y] = str(stats_22[y])
132
133
134                     stats_23[y] = (format(median_patient_1, 'f'))[:-4]
135                     stats_23[y] = str(stats_23[y])
136
137                 data_group1 = collectn_1[1],collectn_1[2],collectn_1[3],collectn_1[4],
138                 collectn_1[5],collectn_1[6],collectn_1[7],collectn_1[8],collectn_1[9],collectn_1[10]
139                 data_group2 = collectn_2[1],collectn_2[2],collectn_2[3],collectn_2[4],
140                 collectn_2[5],collectn_2[6],collectn_2[7],collectn_2[8],collectn_2[9],collectn_2[10]
141
142                 labels_list = [
143                 "Ca-Ka \n p-value= "+pvalue[1]+", mean= "+stats_11[1]+", std= "+stats_12[1]+", median=
                         "+stats_13[1]+"\n hedges-g= "+hedgesg[1]+", mean= "+stats_21[1]+", std=
                         "+stats_22[1]+", median= "+stats_23[1]
144                 ,"Ar-K \n p-value= "+pvalue[2]+", mean= "+stats_11[2]+", std= "+stats_12[2]+", median=
                         "+stats_13[2]+"\n hedges-g= "+hedgesg[2]+", mean= "+stats_21[2]+", std=
                         "+stats_22[2]+", median= "+stats_23[2]
145                 ,"Co-K \n p-value= "+pvalue[3]+", mean= "+stats_11[3]+", std= "+stats_12[3]+", median=
                         "+stats_13[3]+"\n hedges-g= "+hedgesg[3]+", mean= "+stats_21[3]+", std=
                         "+stats_22[3]+", median= "+stats_23[3]
146                 ,"Cr-K \n p-value= "+pvalue[4]+", mean= "+stats_11[4]+", std= "+stats_12[4]+", median=
                         "+stats_13[4]+"\n hedges-g= "+hedgesg[4]+", mean= "+stats_21[4]+", std=
                         "+stats_22[4]+", median= "+stats_23[4]
147                 ,"Cu-K \n p-value= "+pvalue[5]+", mean= "+stats_11[5]+", std= "+stats_12[5]+", median=
                         "+stats_13[5]+"\n hedges-g= "+hedgesg[5]+", mean= "+stats_21[5]+", std=
                         "+stats_22[5]+", median= "+stats_23[5]
148                 ,"Fe-K \n p-value= "+pvalue[6]+", mean= "+stats_11[6]+", std= "+stats_12[6]+", median=
                         "+stats_13[6]+"\n hedges-g= "+hedgesg[6]+", mean= "+stats_21[6]+", std=
                         "+stats_22[6]+", median= "+stats_23[6]
149                 ,"Gd-L \n p-value= "+pvalue[7]+", mean= "+stats_11[7]+", std= "+stats_12[7]+", median=
                         "+stats_13[7]+"\n hedges-g= "+hedgesg[7]+", mean= "+stats_21[7]+", std=
                         "+stats_22[7]+", median= "+stats_23[7]
150                 ,"Ni-K \n p-value= "+pvalue[8]+", mean= "+stats_11[8]+", std= "+stats_12[8]+", median=
                         "+stats_13[8]+"\n hedges-g= "+hedgesg[8]+", mean= "+stats_21[8]+", std=
                         "+stats_22[8]+", median= "+stats_23[8]
151                 ,"W-L \n p-value= "+pvalue[9]+", mean= "+stats_11[9]+", std= "+stats_12[9]+", median=
                         "+stats_13[9]+"\n hedges-g= "+hedgesg[9]+", mean= "+stats_21[9]+", std=
                         "+stats_22[9]+", median= "+stats_23[9]
152                 ,"Zn-K \n p-value= "+pvalue[10]+", mean= "+stats_11[10]+", std= "+stats_12[10]+", median=
                         "+stats_13[10]+"\n hedges-g= "+hedgesg[10]+", mean= "+stats_21[10]+", std=
                         "+stats_22[10]+", median= "+stats_23[10]
153                 ]
154                 xlocations = range(len(data_group1))
155                 width      = 0.2
156                 symbol     = 'r+'
157
158                 ax = plt.gca()
159                 if parameter != "Slope":
160
161                     ymin       = -1
162                     ymax       =  1
163                     ax.set_ylim(ymin,ymax)
164
165                 ax.set_xticklabels( labels_list, rotation=90, fontsize=8 )
166
167                 ax.grid(True, linestyle='dotted')
168                 ax.set_axisbelow(True)
169                 ax.set_xticks(xlocations)
170                 plt.xlabel('')
171                 plt.ylabel(parameter)
172                 plt.title(element1, fontsize=20)
173
174                 positions_group1 = [x-(width-0.5) for x in xlocations]
175                 positions_group2 = xlocations
176
177                 plt.boxplot(data_group1,
178                           sym=symbol,
179                           labels=['']*len(labels_list),
180                           positions=positions_group1,
181                           widths=width,
182
183                          patch_artist=True,
184                          boxprops=dict(facecolor='g'),
185                           )
186
```

```
                                plt.boxplot(data_group2,
                                            labels=labels_list,
                                            sym=symbol,
                                            positions=positions_group2,
                                            widths=width,

                                           patch_artist=True,
                                            )

                                plt.savefig('box_plot_PCC_SCC_GD1_GD5/'+element1+'_'+parameter+'.png', bbox_inches='tight',
                                     dpi=800)
                                plt.cla()
                                plt.clf()
```

```
#overlap_data_collector_master_M_M1_M2_v1_final.py
#Lukas Warnung
import numpy as np
import matplotlib as mpl
import matplotlib.pyplot as plt
from scipy import stats
from scipy.stats import pearsonr
from scipy.stats import spearmanr

d={}
x=0
i=0
r=0
k=0
parameters = ["M","M1","M2"]

for parameter in parameters:

               if parameter == "M":
                   line = 8
               if parameter == "M1":
                   line = 25
               if parameter == "M2":
                   line = 26

               synchrotronS = ["Diamond","ESRF"]

               for synchrotron in synchrotronS:

                      if synchrotron == "Diamond":
                          specimens = ["GD1a","GD1b","GD1bsecond","GD2a","GD2b","GD3","GD4","GD5"]
                          elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]

                      if synchrotron == "ESRF":
                          specimens = ["GD1b2","GD4","GD5"]
                          elements =["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-Ka","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]

                      for specimen in specimens:
                          if specimen == "GD1a":
                              spots = ["183278","183279","183280"]
                          if specimen == "GD1b":
                              spots = ["183242"]
                          if specimen == "GD1bsecond":
                              spots = ["183309"]
                          if specimen == "GD2a":
                              spots = ["183287","183288"]
                          if specimen == "GD2b":
                              spots = ["183247","183248"]
                          if specimen == "GD3":
                              spots = ["183270"]
                          if specimen == "GD4" and synchrotron == "Diamond":
                              spots = ["183258"]
                          if specimen == "GD5" and synchrotron == "Diamond":
                              spots = ["183293","183294","183297","183298","183300","183302","183303"]
                          if specimen == "GD1b2":
                              spots = ["scan10","scan22"]
                          if specimen == "GD4" and synchrotron == "ESRF":
                              spots = ["18","scan17","scan35"]
                          if specimen == "GD5" and synchrotron == "ESRF":
                              spots = ["scan18","scan21","scan27","scan28","scan32","scan34","scan37","scan38"]

                          for spot in spots:

                              if spot == "scan10" or spot == "scan18" or spot == "scan21" or spot == "scan32":
                                  maps = "maps"
                              elif spot == "scan17" or spot == "scan37" or spot == "scan38":
                                  maps = "maps_20s"
                              else:
```

```
                                maps = "maps_1s"

                            print(spot)

                            x=0
                            for element1 in elements:

                                    for element2 in elements:

                                        if element1 != element2:

                                                with open('overlap_log/'+synchrotron+'_'+specimen+'_'+
                                                spot+'_'+element1+'_vs_'+element2+'_'+maps+'_Log.txt') as f:

                                                    manders_data = f.readlines()[line]

                                                if element1 == "Cu-Ka":
                                                    element1 = "Cu-K"

                                                if element2 == "Cu-Ka":
                                                    element2 = "Cu-K"

                                                file_PCC = open("overlap_all_data_"+parameter+
                                                "/overlap_all_data_"+element1+"_"+element2+".txt","a")
                                                file_PCC.writelines(synchrotron+" "+specimen+" "+spot+" ")
                                                file_PCC.writelines(manders_data)
                                                file_PCC.close()

                                                if element1 == "Cu-K" and synchrotron == "ESRF":
                                                    element1 = "Cu-Ka"

                                                if element2 == "Cu-K" and synchrotron == "ESRF":
                                                    element2 = "Cu-Ka"
```

```
#box_plot_MOC_master_M_M1_M2_v1_final.py
#Lukas Warnung
import numpy as np
import matplotlib as mpl
import matplotlib.pyplot as plt
from scipy import stats
from scipy.stats import pearsonr
from scipy.stats import spearmanr

import re

d={}
x=0
i=0
r=0
k=0
collectn_1={}
collectn_1={}
mean_all = {}
std_all = {}
median_all = {}
parameters = ["M","M1","M2"]

for parameter in parameters:

            elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]

            x=0

            for element1 in elements:

                                x=x+1
                                y=1
                                x_label = []
                                r = [1]

                                for element2 in elements:

                                    DATA_array = []
                                    x_label.append(element2)

                                    if element1 == element2:
                                        collectn_1[y] = r

                                        print(collectn_1[y])

                                    if element1 != element2:
```

```
                    i=0

                    head_label = []

                    for i in range(31):
                        with
                              open("overlap_all_data_"+parameter+"/overlap_all_data_"+element1+"_"+element2+".txt")
                              as f:

                            DATA = f.readlines()[i]
                            DATA = DATA[-38:]
                            new_DATA = DATA.split('=')
                            new_DATA_2 = new_DATA[1]
                            new_DATA = new_DATA_2.split(' ')
                            DATA_float = float(new_DATA[0])
                            DATA_array.append(DATA_float)

        collectn_1[y] = DATA_array
        mean_all[y] = np.mean(collectn_1[y])
        std_all[y] = np.std(collectn_1[y])
        median_all[y] = np.median(collectn_1[y])

        mean_all[y] = (format(mean_all[y], 'f'))[:-4]
        mean_all[y] = str(mean_all[y])

        std_all[y] = (format(std_all[y], 'f'))[:-4]
        std_all[y] = str(std_all[y])

        median_all[y] = (format(median_all[y], 'f'))[:-4]
        median_all[y] = str(median_all[y])

        print(collectn_1[y])
        y=y+1

print("--------------------")

data_to_plot = collectn_1[1],collectn_1[2],collectn_1[3],collectn_1[4],
collectn_1[5],collectn_1[6],collectn_1[7],collectn_1[8],collectn_1[9],collectn_1[10]

fig = plt.figure(1, figsize=(12, 10))

ax = fig.add_subplot(111)

bp = ax.boxplot(data_to_plot)

bp = ax.boxplot(data_to_plot, patch_artist=True)

for box in bp['boxes']:

    box.set( color='#7570b3', linewidth=2)
    box.set( facecolor = '#1b9e77' )

for whisker in bp['whiskers']:
    whisker.set(color='#7570b3', linewidth=2)

for cap in bp['caps']:
    cap.set(color='#7570b3', linewidth=2)

for median in bp['medians']:
    median.set(color='#b2df8a', linewidth=2)

for flier in bp['fliers']:
    flier.set(marker='o', color='#e7298a', alpha=0.5)

plt.title(element1,size=28)

labels_list = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K",
"Ca-Ka \n mean= "+mean_all[1]+" std= "+std_all[1]+" median= "+median_all[1],
"Ar-K \n mean= "+mean_all[2]+" std= "+std_all[2]+" median= "+median_all[2],
"Co-K \n mean= "+mean_all[3]+" std= "+std_all[3]+" median= "+median_all[3],
"Cr-K \n mean= "+mean_all[4]+" std= "+std_all[4]+" median= "+median_all[4],
"Cu-K \n mean= "+mean_all[5]+" std= "+std_all[5]+" median= "+median_all[5],
"Fe-K \n mean= "+mean_all[6]+" std= "+std_all[6]+" median= "+median_all[6],
"Gd-L \n mean= "+mean_all[7]+" std= "+std_all[7]+" median= "+median_all[7],
"Ni-K \n mean= "+mean_all[8]+" std= "+std_all[8]+" median= "+median_all[8],
"W-L \n mean= "+mean_all[9]+" std= "+std_all[9]+" median= "+median_all[9],
"Zn-K \n mean= "+mean_all[10]+" std= "+std_all[10]+" median= "+median_all[10],
]

ax.set_xticklabels(labels_list, rotation=90, fontsize=15 )

print(x_label)

plt.ylabel(parameter,size=22)
plt.yticks(size=22)
```

```
                            ax.get_xaxis().tick_bottom()
                            ax.get_yaxis().tick_left()

                            fig.savefig('box_plot_MOC_'+parameter+'/'+element1+'_MOC.png', bbox_inches='tight')
                            plt.cla()
                            plt.clf()
```

```
#box_plot_MOC_master_M_M1_M2_GD1_GD5_compare_v1_final.py
#Lukas Warnung
import numpy as np
import matplotlib as mpl
import matplotlib.pyplot as plt
from scipy import stats
from scipy.stats import pearsonr
from scipy.stats import spearmanr
from scipy import *

import pandas as pd
import researchpy
import math

d={}
x=0
i=0
r=0
k=0
collectn_1={}
collectn_2={}
pvalue = {}
hedgesg = {}
p_value = {}
hedges_g = {}
result = {}
stats_11 = {}
stats_12 = {}
stats_21 = {}
stats_22 = {}
stats_13 = {}
stats_23 = {}
parameters = ["M","M1","M2"]

for parameter in parameters:

        elements = ["Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K"]
        x=0

        for element1 in elements:

                        y=0
                        r = [1,1]

                        for element2 in elements:

                            y=y+1
                            print(y)
                            if element1 == element2:
                                collectn_1[y] = r
                                collectn_2[y] = r
                                pvalue[y] = "0"
                                hedgesg[y] = "0"
                                stats_11[y] = "0"
                                stats_12[y] = "0"
                                stats_13[y] = "0"
                                stats_21[y] = "0"
                                stats_22[y] = "0"
                                stats_23[y] = "0"

                            if element1 != element2:

                                i=0
                                DATA_array = []
                                DATA_float = 0
                                DATA_array_2 = []
                                DATA_float_2 = 0

                                for i in range(31):
                                    with
                                        open("overlap_all_data_"+parameter+"/overlap_all_data_"+element1+"_"+element2+".txt")
                                        as f:

                                        DATA = f.readlines()[i]
```

```
                DATA_2 = DATA[-38:]
                new_DATA = DATA_2.split('=')
                new_DATA_2 = new_DATA[1]
                new_DATA = new_DATA_2.split(' ')
                DATA_float = float(new_DATA[0])
                patient = DATA.split(" ")
                patient_x = patient[3]

                if patient_x == "GD1a" or patient_x == "GD1b" or patient_x == "GD1bsecond"
                     or patient_x == "GD1b2":

                    DATA_array.append(DATA_float)

                if patient_x == "GD5":
                    DATA_array_2.append(DATA_float)

            collectn_1[y] = DATA_array
            collectn_2[y] = DATA_array_2

            mean_patient_1 = np.mean(collectn_1[y])
            std_patient_1 = np.std(collectn_1[y])
            n_patient_1 = len(collectn_1[y])
            median_patient_1 = np.median(collectn_1[y])

            mean_patient_2 = np.mean(collectn_2[y])
            std_patient_2 = np.std(collectn_2[y])
            n_patient_2 = len(collectn_2[y])
            median_patient_2 = np.median(collectn_2[y])

            Diff_mean = mean_patient_1-mean_patient_2

            Z =
                 ((n_patient_1-1)*std_patient_1*std_patient_1)+((n_patient_2-1)*std_patient_2*std_patient_2)
            N = n_patient_1+n_patient_2-2

            root = math.sqrt(Z/N)
            Cohens_d = Diff_mean / root
            Cohens_abs = abs(Cohens_d)
            hedges_g = Cohens_abs * (1-(3/(4*(n_patient_1+n_patient_2-9))))
            result, p_value = stats.ttest_ind(collectn_1[y], collectn_2[y], equal_var = False)

            pvalue[y] = (format(p_value, 'f'))[:-4]
            pvalue[y] = str(pvalue[y])

            hedgesg[y] = (format(hedges_g, 'f'))[:-4]
            hedgesg[y] = str(hedgesg[y])

            stats_11[y] = (format(mean_patient_2, 'f'))[:-4]
            stats_11[y] = str(stats_11[y])

            stats_12[y] = (format(std_patient_2, 'f'))[:-4]
            stats_12[y] = str(stats_12[y])

            stats_13[y] = (format(median_patient_2, 'f'))[:-4]
            stats_13[y] = str(stats_13[y])

            stats_21[y] = (format(mean_patient_1, 'f'))[:-4]
            stats_21[y] = str(stats_21[y])

            stats_22[y] = (format(std_patient_1, 'f'))[:-4]
            stats_22[y] = str(stats_22[y])

            stats_23[y] = (format(median_patient_1, 'f'))[:-4]
            stats_23[y] = str(stats_23[y])


data_group1 = collectn_1[1],collectn_1[2],collectn_1[3],collectn_1[4],
collectn_1[5],collectn_1[6],collectn_1[7],collectn_1[8],collectn_1[9],collectn_1[10]
data_group2 = collectn_2[1],collectn_2[2],collectn_2[3],collectn_2[4],
collectn_2[5],collectn_2[6],collectn_2[7],collectn_2[8],collectn_2[9],collectn_2[10]

labels_list = [
"Ca-Ka \n p-value= "+pvalue[1]+", mean= "+stats_11[1]+", std= "+stats_12[1]+", median=
     "+stats_13[1]+
"\n hedges-g= "+hedgesg[1]+", mean= "+stats_21[1]+", std= "+stats_22[1]+", median=
     "+stats_23[1]
,"Ar-K \n p-value= "+pvalue[2]+", mean= "+stats_11[2]+", std= "+stats_12[2]+", median=
     "+stats_13[2]+
"\n hedges-g= "+hedgesg[2]+", mean= "+stats_21[2]+", std= "+stats_22[2]+", median=
     "+stats_23[2]
,"Co-K \n p-value= "+pvalue[3]+", mean= "+stats_11[3]+", std= "+stats_12[3]+", median=
     "+stats_13[3]+
"\n hedges-g= "+hedgesg[3]+", mean= "+stats_21[3]+", std= "+stats_22[3]+", median=
     "+stats_23[3]
```

```
                                ,"Cr-K \n p-value= "+pvalue[4]+", mean= "+stats_11[4]+", std= "+stats_12[4]+", median=
                                    "+stats_13[4]+
                                "\n hedges-g= "+hedgesg[4]+", mean= "+stats_21[4]+", std= "+stats_22[4]+", median=
                                    "+stats_23[4]
                                ,"Cu-K \n p-value= "+pvalue[5]+", mean= "+stats_11[5]+", std= "+stats_12[5]+", median=
                                    "+stats_13[5]+
                                "\n hedges-g= "+hedgesg[5]+", mean= "+stats_21[5]+", std= "+stats_22[5]+", median=
                                    "+stats_23[5]
                                ,"Fe-K \n p-value= "+pvalue[6]+", mean= "+stats_11[6]+", std= "+stats_12[6]+", median=
                                    "+stats_13[6]+
                                "\n hedges-g= "+hedgesg[6]+", mean= "+stats_21[6]+", std= "+stats_22[6]+", median=
                                    "+stats_23[6]
                                ,"Gd-L \n p-value= "+pvalue[7]+", mean= "+stats_11[7]+", std= "+stats_12[7]+", median=
                                    "+stats_13[7]+
                                "\n hedges-g= "+hedgesg[7]+", mean= "+stats_21[7]+", std= "+stats_22[7]+", median=
                                    "+stats_23[7]
                                ,"Ni-K \n p-value= "+pvalue[8]+", mean= "+stats_11[8]+", std= "+stats_12[8]+", median=
                                    "+stats_13[8]+
                                "\n hedges-g= "+hedgesg[8]+", mean= "+stats_21[8]+", std= "+stats_22[8]+", median=
                                    "+stats_23[8]
                                ,"W-L \n p-value= "+pvalue[9]+", mean= "+stats_11[9]+", std= "+stats_12[9]+", median=
                                    "+stats_13[9]+
                                "\n hedges-g= "+hedgesg[9]+", mean= "+stats_21[9]+", std= "+stats_22[9]+", median=
                                    "+stats_23[9]
                                ,"Zn-K \n p-value= "+pvalue[10]+", mean= "+stats_11[10]+", std= "+stats_12[10]+", median=
                                    "+stats_13[10]+
                                "\n hedges-g= "+hedgesg[10]+", mean= "+stats_21[10]+", std= "+stats_22[10]+", median=
                                    "+stats_23[10]
                                ]
                                xlocations = range(len(data_group1))
                                width      = 0.2
                                symbol     = 'r+'
                                ymin       = -0.1
                                ymax       =  1.1

                                ax = plt.gca()
                                ax.set_ylim(ymin,ymax)
                                ax.set_xticklabels( labels_list, rotation=90, fontsize=8 )

                                ax.grid(True, linestyle='dotted')
                                ax.set_axisbelow(True)
                                ax.set_xticks(xlocations)
                                plt.xlabel('')
                                plt.ylabel(parameter)
                                plt.title(element1, fontsize=20)

                                positions_group1 = [x-(width-0.5) for x in xlocations]
                                positions_group2 = xlocations

                                plt.boxplot(data_group1,
                                           sym=symbol,
                                           labels=['']*len(labels_list),
                                           positions=positions_group1,
                                           widths=width,

                                          patch_artist=True,
                                          boxprops=dict(facecolor='g'),
                                           )

                                plt.boxplot(data_group2,
                                          labels=labels_list,
                                          sym=symbol,
                                          positions=positions_group2,
                                          widths=width,

                                          patch_artist=True,
                                           )

                                plt.savefig('box_plot_MOC_'+parameter+'_GD1_GD5/'+element1+'_'+parameter+'.png',
                                     bbox_inches='tight', dpi=800)
                                plt.cla()
                                plt.clf()
```

```
0
1  #multi_plot_all_data_v4_final.py
2  #Lukas Warnung
3  import numpy as np
4  import matplotlib as mpl
5  import matplotlib.pyplot as plt
6  import matplotlib.image as mpimg
7  from scipy import stats
8  from scipy.stats import pearsonr
9  from scipy.stats import spearmanr
```

```
10
11  import matplotlib.pyplot as plt
12  import matplotlib.image as mpimg
13  import os
14
15  text_kwargs = dict(ha='center', va='center', fontsize=8, color='C2')
16
17  dirPath = "plots_high_correlation"
18
19  data_set = os.listdir(dirPath)
20  for png in data_set[:]:
21      if not(png.endswith(".png")):
22          data_set.remove(png)
23
24  i=0
25  for data in data_set:
26
27      img_correlation = mpimg.imread('plots_high_correlation/'+data_set[i]+'')
28
29      overlap_0_file = data_set[i]
30
31      overlap_0_file = overlap_0_file.replace(".png", "_overlap.png")
32
33      img_overlap_0_ = mpimg.imread('overlap_log/'+overlap_0_file+'')
34
35      overlap_file = data_set[i]
36
37      overlap_file = overlap_file.replace(".png", "_overlap_intensity.png")
38
39      img_overlap = mpimg.imread('overlap_log/'+overlap_file+'')
40
41      zerteilen = data_set[i].split("_",9)
42      print(zerteilen)
43
44      synchrotron = zerteilen[0]
45      specimen = zerteilen[1]
46      spot = zerteilen[2]
47      element_X = zerteilen[3]
48      element_Y = zerteilen[5]
49
50      if zerteilen[6] == "maps":
51          maps = zerteilen[6]+"_"+zerteilen[7]
52
53      else:
54          maps = zerteilen[6]
55
56      maps_2 = maps.split(".",2)
57      maps_2 = maps_2[0]
58
59      print(maps_2)
60
61      file_origin_X = mpimg.imread('MA_LW_Gd data
            selection/'+synchrotron+'/'+specimen+'/'+spot+'/'+maps_2+'/'+'PNG/'+element_X+'.png')
62
63      zerteilen = data_set[i].split("_",9)
64      print(zerteilen)
65
66      synchrotron = zerteilen[0]
67      specimen = zerteilen[1]
68      spot = zerteilen[2]
69      element_X = zerteilen[3]
70      element_Y = zerteilen[5]
71
72      if zerteilen[6] == "maps":
73          maps = zerteilen[6]+"_"+zerteilen[7]
74      else:
75          maps = zerteilen[6]
76
77      maps_2 = maps.split(".",2)
78      maps_2 = maps_2[0]
79
80      print(zerteilen[2])
81
82      file_origin_Y = mpimg.imread('MA_LW_Gd data
            selection/'+synchrotron+'/'+specimen+'/'+spot+'/'+maps_2+'/'+'PNG/'+element_Y+'.png')
83
84      text_file = data_set[i]
85
86      text_file = text_file.replace(".png", "_Log.txt")
87
88      with open('overlap_log/'+text_file+'') as f:
89          manders_data = f.readlines()[8]
90
91      with open('overlap_log/'+text_file+'') as f:
92          manders_data_pre_A = f.readlines()[1]
```

```
93
94      with open('overlap_log/'+text_file+'') as f:
95          manders_data_pre_B = f.readlines()[2]
96
97      with open('overlap_log/'+text_file+'') as f:
98          manders_data_M1 = f.readlines()[25]
99
100     with open('overlap_log/'+text_file+'') as f:
101         manders_data_M2 = f.readlines()[26]
102
103     plt.subplot(231)
104     plt.imshow(file_origin_Y)
105     plt.axis('off')
106
107     plt.subplot(232)
108     plt.imshow(img_correlation)
109     plt.axis('off')
110
111     plt.subplot(233)
112     plt.imshow(img_overlap)
113     plt.axis('off')
114
115     plt.subplot(234)
116     plt.text(0.25, 0.25, '(Red) '+manders_data_pre_A+'(Blue) '+manders_data_pre_B+
117     'M='+manders_data[2:]+''+manders_data_M1+''+manders_data_M2, **text_kwargs)
118     plt.axis('off')
119
120     plt.subplot(235)
121     plt.imshow(file_origin_X)
122     plt.axis('off')
123
124     plt.subplot(236)
125     plt.imshow(img_overlap_0_)
126     plt.axis('off')
127
128     print("-"+element_X+"-")
129
130     if element_X != "Co-K" and element_Y != "Co-K" and element_X != "W-L" and
131     element_Y != "W-L" and element_X != "Ar-K" and element_Y and "Ar-K":
132
133             plt.savefig('multi_plot/multi_plot_'+data_set[i], dpi=500)
134
135     plt.close()
136
137     print(i)
138     print("-------")
139     print(data_set[i])
140     print("-------")
141     i=i+1
```

7.2 ImageJ (Macro Language) Code

```
//Overlap_macro_multi_intensity_normal_master_v4_final.ijm
//Lukas Warnung

main_folder2 = getDirectory("main_folder");
save_folder = getDirectory("save_folder");

Threshold = "Percentile white";

synchrotronS = newArray("Diamond","ESRF");

synchrotronS_Length = synchrotronS.length;

for (i=0;i<=synchrotronS_Length-1;i++){

	if (synchrotronS[i] == "Diamond")
	{
	    specimens = newArray("GD1a","GD1b","GD1bsecond","GD2a","GD2b","GD3","GD4","GD5");
	    elements = newArray("Ca-Ka","Ar-K","Co-K","Cr-K","Cu-K","Fe-K","Gd-L","Ni-K","W-L","Zn-K");
	}

	if (synchrotronS[i] == "ESRF")
	{
	    specimens = newArray("GD1b2","GD4","GD5");
	    elements = newArray("Ca-Ka","Ar-K","Co-K","Cr-K","Cu-Ka","Fe-K","Gd-L","Ni-K","W-L","Zn-K");
	}

specimensLength = specimens.length;

for (h=0; h <= specimensLength-1;h++){
	    if (specimens[h] == "GD1a")
	    {
	        spots = newArray("183278","183279","183280");
	    }
	    if (specimens[h] == "GD1b")
	    {
	        spots = newArray("183242");
	    }
	    if (specimens[h] == "GD1bsecond")
	    {
	        spots = newArray("183309");
	    }
	    if (specimens[h] == "GD2a")
	    {
	        spots = newArray("183287","183288");
	    }
	    if (specimens[h] == "GD2b")
	    {
	        spots = newArray("183247","183248");
	    }
	    if (specimens[h] == "GD3")
	    {
	        spots = newArray("183270");
	    }
	    if (specimens[h] == "GD4" && synchrotronS[i] == "Diamond")
	    {
	        spots = newArray("183258");
	    }
	    if (specimens[h] == "GD5" && synchrotronS[i] == "Diamond")
	    {
	        spots = newArray("183293","183294","183297","183298","183300","183302","183303");
	    }
	    if (specimens[h] == "GD1b2")
	    {
	        spots = newArray("scan10","scan22");
	    }
	    if (specimens[h] == "GD4" && synchrotronS[i] == "ESRF")
	    {
	        spots = newArray("18","scan17","scan35");
	    }
	    if (specimens[h] == "GD5" && synchrotronS[i] == "ESRF")
	    {
	        spots = newArray("scan18","scan21","scan27","scan28","scan32","scan34","scan37","scan38");
	    }

spotsLength = spots.length;

	for (s=0; s<=spotsLength-1;s++){

	        if (spots[s] == "scan10" || spots[s] == "scan18" || spots[s] == "scan21" || spots[s] == "scan32")
	            {
	            maps = "maps";
	            }
```

```
                else if (spots[s] == "scan17" || spots[s] == "scan37" || spots[s] == "scan38")
                  {
                    maps = "maps_20s";
                  }
                else
                  {
                    maps = "maps_1s";
                  }

elementsLength = elements.length;

        for (k=0;k<=elementsLength-1;k++){

           Element_1=elements[k];

              for (kk=0;kk<=elementsLength-1;kk++){

                 Element_2=elements[kk];

if(Element_1 != Element_2){

if(maps == "maps_20s")
{
   min_threshold="200";
}
else
{
   min_threshold="10";
}

main_folder = main_folder2+"/"+synchrotronS[i]+"/"+specimens[h]+"/"+spots[s]+"/"+maps;
File.makeDirectory(main_folder+"/PNG/");
File.makeDirectory(main_folder+"/"+Element_1+"_"+Element_2+"_plot");

run("Text Image... ", "open=["+main_folder+"/"+Element_1+".txt]");

run("Text Image... ", "open=["+main_folder+"/"+Element_2+".txt]");

selectWindow(""+Element_1+".txt");
//run("Threshold...");
setThreshold(min_threshold, 10000000000000000000000000000000000000000000.0000);
setOption("BlackBackground", true);
run("Convert to Mask");

saveAs("PNG", ""+main_folder+"/PNG/"+Element_1+"mask.png");

selectWindow(""+Element_2+".txt");
//run("Threshold...");
setThreshold(min_threshold, 10000000000000000000000000000000000000000000.0000);
setOption("BlackBackground", true);
run("Convert to Mask");

saveAs("PNG", ""+main_folder+"/PNG/"+Element_2+"mask.png");

close();
selectWindow(""+Element_1+"mask.png");
close();

open(""+main_folder+"/PNG/"+Element_1+"mask.png");

//run("Auto Threshold", "method="+Threshold+"");

open(""+main_folder+"/PNG/"+Element_2+"mask.png");

//run("Auto Threshold", "method="+Threshold+"");

run("JACoP ", "imga="+Element_1+"mask.png imgb="+Element_2+"mask.png thra=15 thrb=15 pearson overlap mm");
saveAs("Text", main_folder+"/"+Element_1+"_"+Element_2+"_plot/Log.txt");

selectWindow("Log");

path = save_folder+"/"+synchrotronS[i]+"_"+specimens[h]+"_"+
spots[s]+"_"+Element_1+"_vs_"+Element_2+"_"+maps+"_Log.txt";

saveAs("Text", path);
close("Log");

   selectWindow(""+Element_1+"mask.png");
   run("Red");
   selectWindow(""+Element_2+"mask.png");
   run("Blue");

   selectWindow(""+Element_2+"mask.png");
   run("Add Image...", ""+Element_2+"mask.png x=0 y=0 opacity=50");
   //saveAs("PNG", main_folder+"/"+Element_1+"_"+Element_2+"_plot"+Element_1+"_"+Element_2+"_overlap.png");
```

```
167
168     saveAs("PNG", save_folder+"/"+synchrotronS[i]+"_"+specimens[h]+"_"+
169    spots[s]+"_"+Element_1+"_vs_"+Element_2+"_"+maps+"_overlap.png");
170
171     close();
172     close();
173
174     run("Text Image... ", "open=["+main_folder+"/"+Element_1+".txt]");
175
176     run("Text Image... ", "open=["+main_folder+"/"+Element_2+".txt]");
177
178     selectWindow(""+Element_1+".txt");
179     run("Red");
180
181     selectWindow(""+Element_2+".txt");
182     run("Blue");
183
184     selectWindow(""+Element_2+".txt");
185     run("Add Image...", ""+Element_2+".txt x=0 y=0 opacity=50");
186
187     saveAs("PNG", save_folder+"/"+synchrotronS[i]+"_"+specimens[h]+"_"+
188    spots[s]+"_"+Element_1+"_vs_"+Element_2+"_"+maps+"_overlap_intensity.png");
189
190     close();
191     close();
192
193     run("Text Image... ", "open=["+main_folder+"/"+Element_1+".txt]");
194     selectWindow(""+Element_1+".txt");
195     saveAs("PNG", ""+main_folder+"/PNG/"+Element_1+".png");
196     close();
197
198     run("Text Image... ", "open=["+main_folder+"/"+Element_2+".txt]");
199     selectWindow(""+Element_2+".txt");
200     saveAs("PNG", ""+main_folder+"/PNG/"+Element_2+".png");
201     close();
202
203 }
204
205 }
206           }
207         }
208       }
209     }
```

7.3 Data

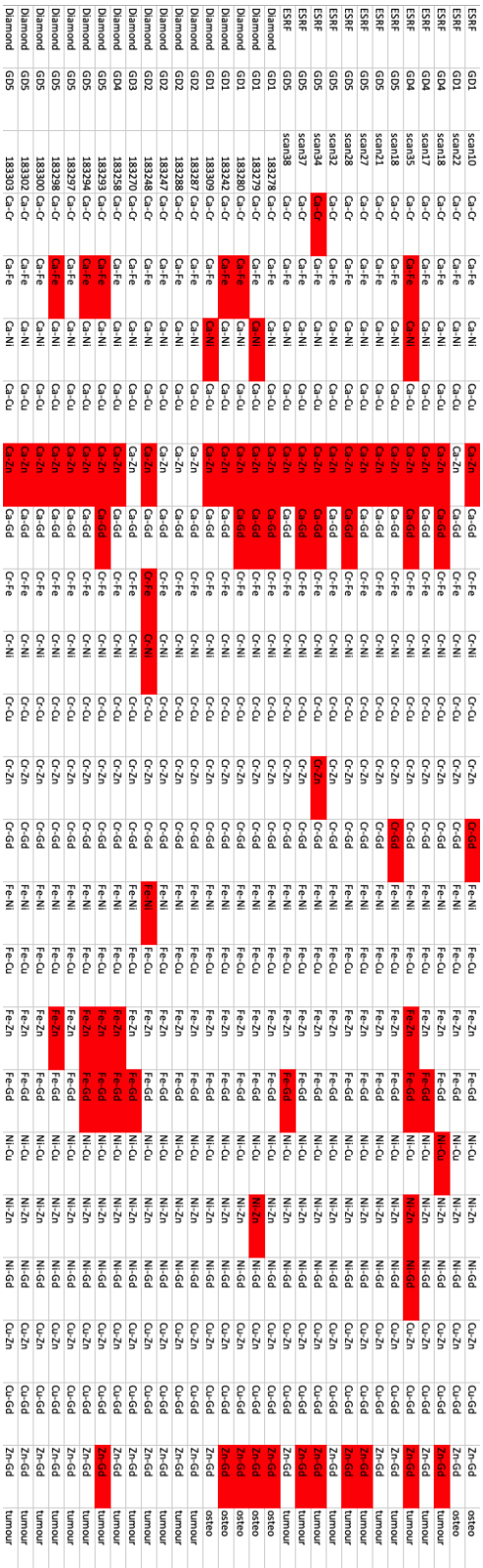


Figure 70: The table describes an overview for all possible element combinations about the 31 single scan spots. The first column describes the synchrotron, the second column shows the patient, the third column gives the scan spot number and from column four the element combinations are set.

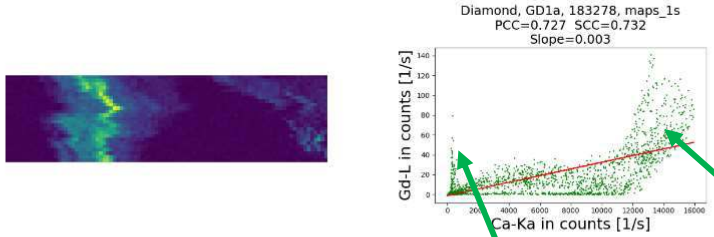
Data Interpretation

High Correlated Data
(PCC or SCC > 0,65)

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

GD1

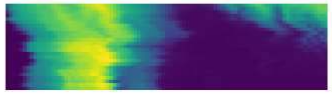
At the beginning of the correlation graph, a small Gd spot can be located and a small linear correlation part at the end. The overlap shows Gd overlaps Ca to 100%.



Linear correlation area: Gd is increasing relatively more than Ca in that region.

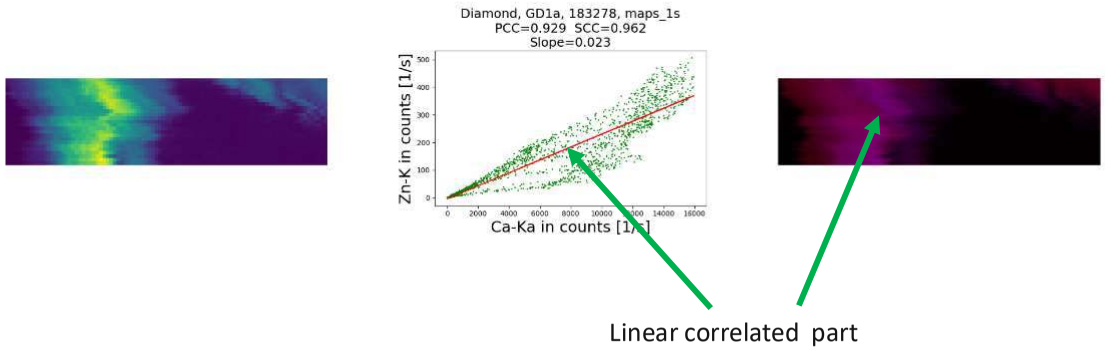
Gd spot without Ca

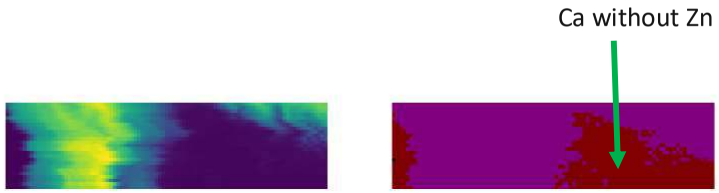
Ca without Zn



(Red) Image A: Ca-Kamask.png
(Blue) Image B: Gd-Lmask.png
M=0.649
M1=0.421 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

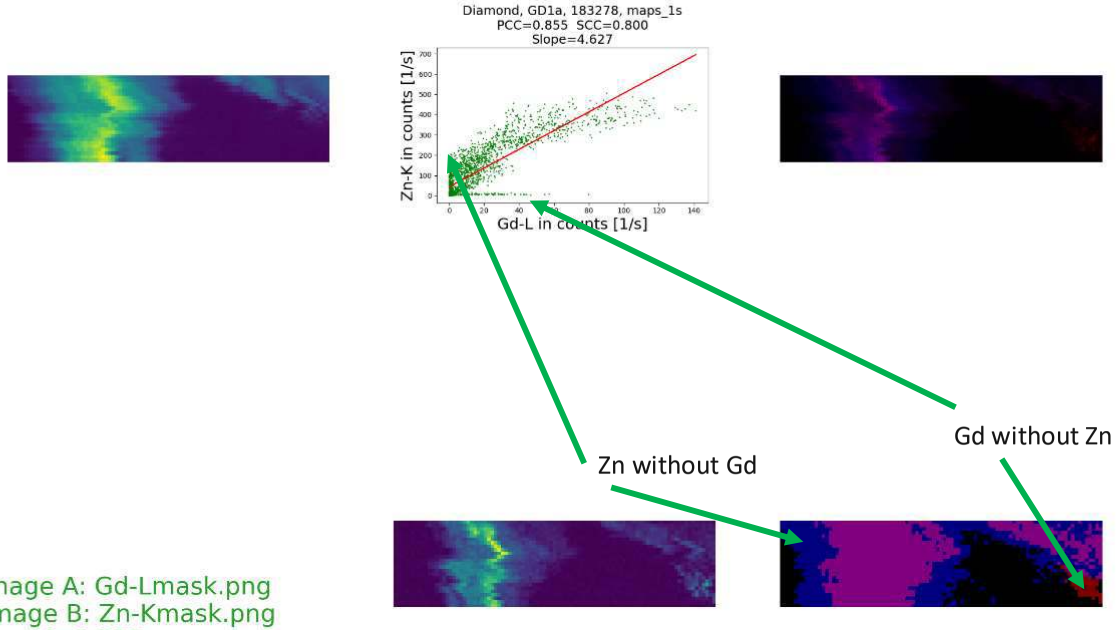
A high linear correlation can be observed. The Intensity of the signal is increasing from the outer to the inner region. Zn overlaps Ca to 100%.



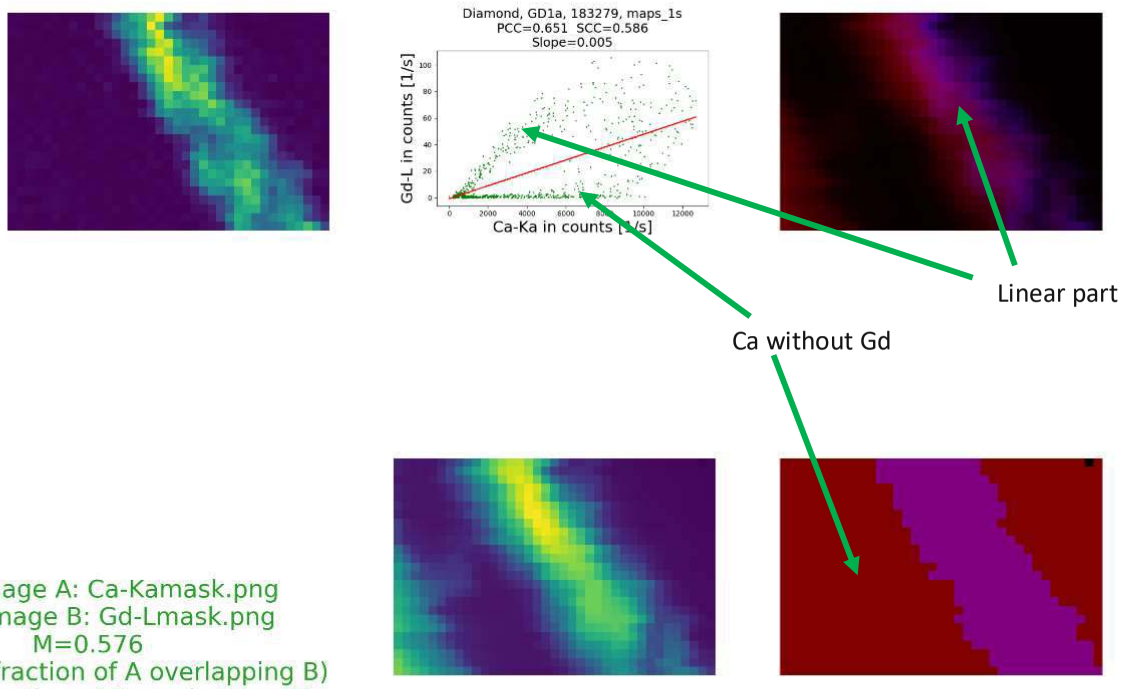


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.835
M1=0.697 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

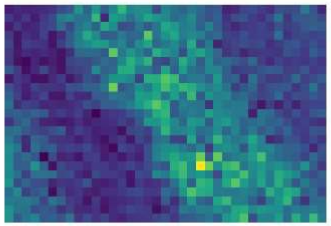
The correlation is high and from 0 to 80 Gd counts a small Gd spot without Zn can be shown. The overlap for Gd is near to 1.
Zn without Gd can be located at the edge regions.

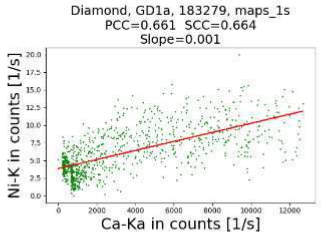


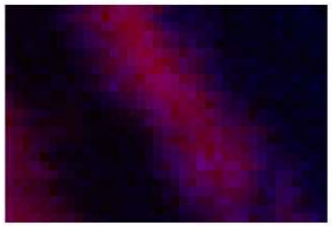
The linear correlation is shown in the middle area. A region (left lower corner) shows Ca without Gd. Gd overlaps Ca to 100%.



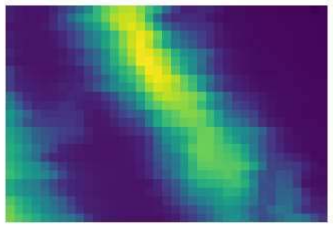
The linear correlation is low. Ni overlaps Ca to 100%.

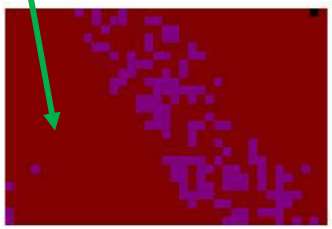






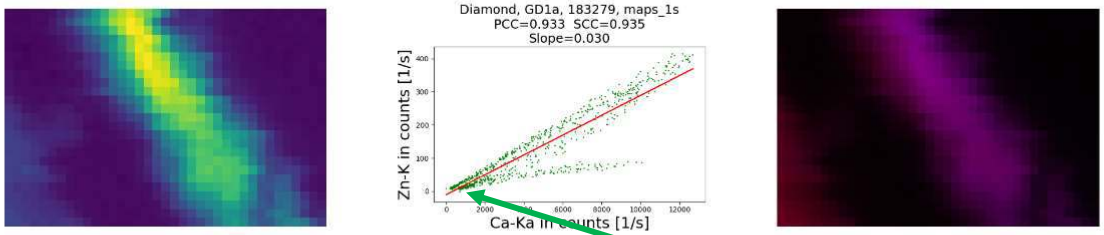
Ca without Ni



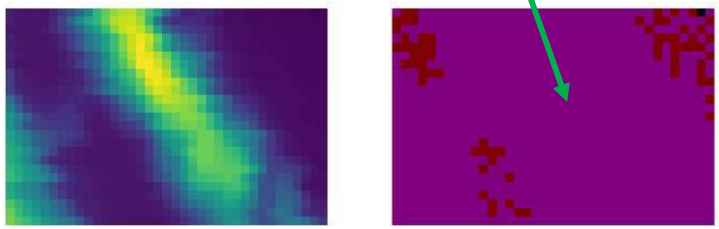


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Ni-Kmask.png
M=0.383
M1=0.147 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

Ca without Zn can be determined. Zn overlaps Ca to 100%.

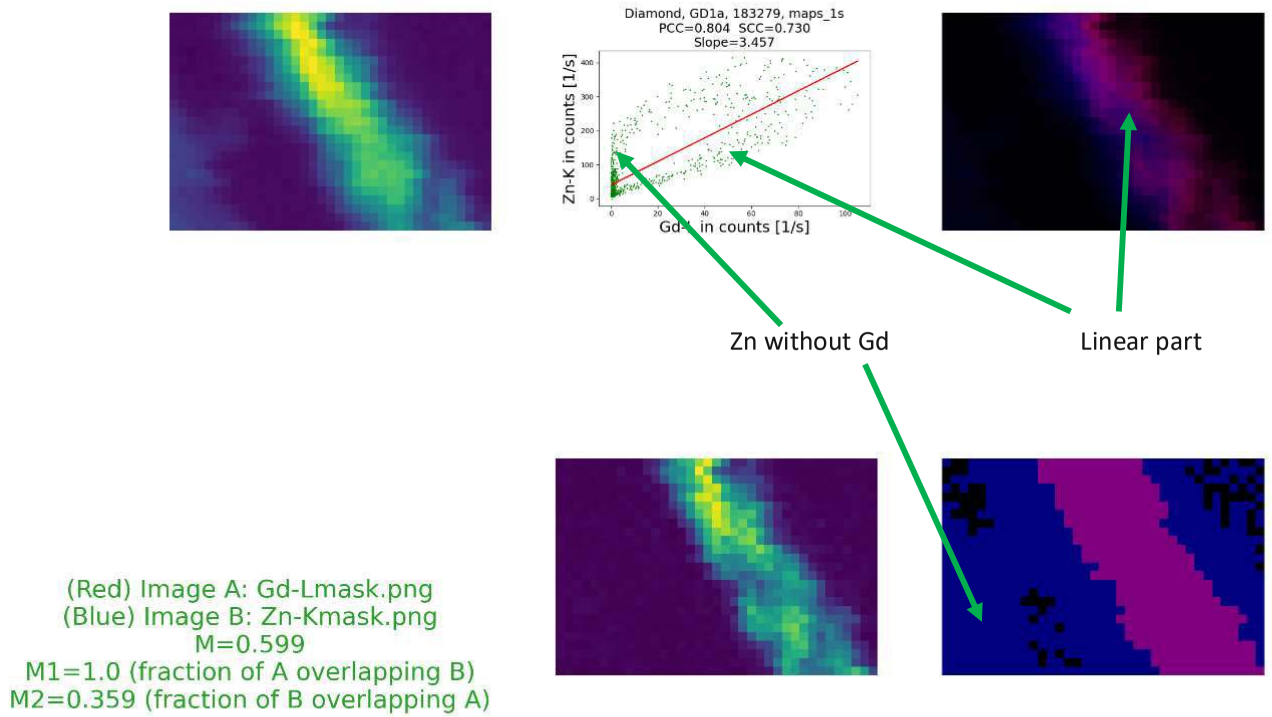


Ca without Zn

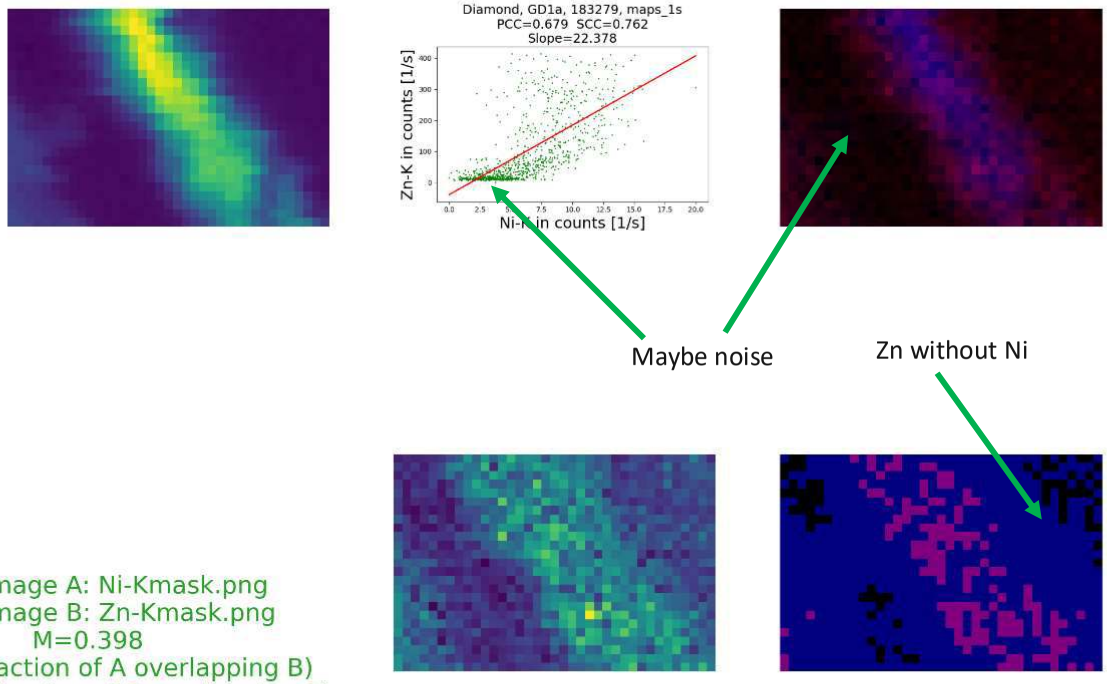


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.961
M1=0.925 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

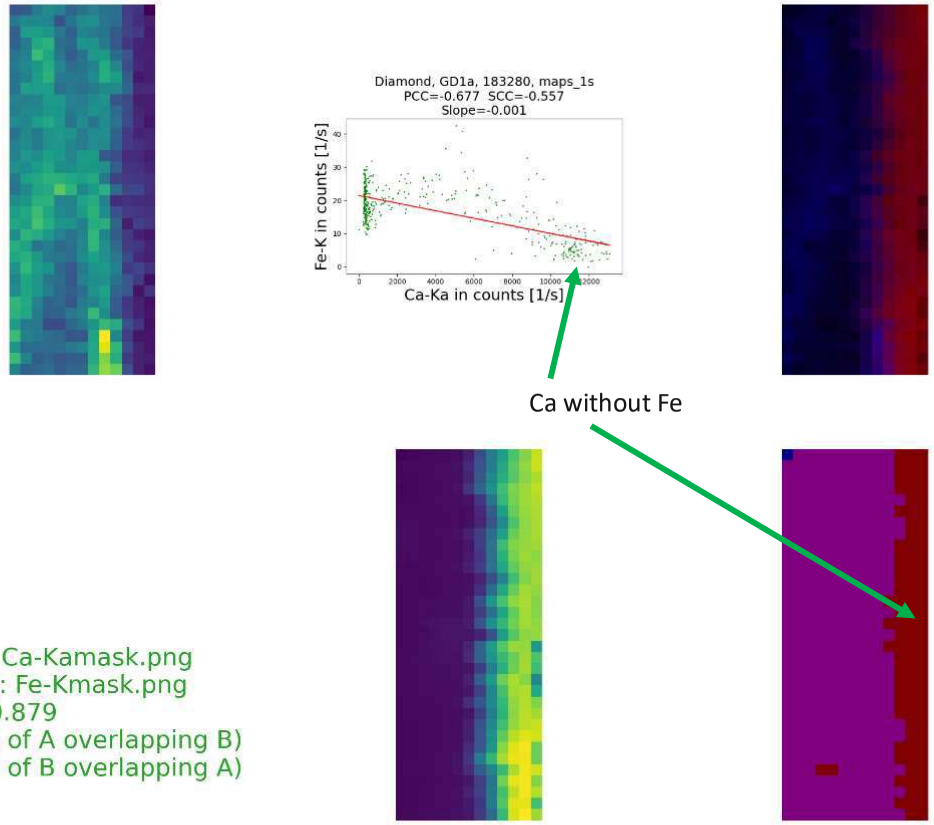
The two linear parts of the X-Y plots can be localized in the overlap image. Zn without Gd can be determined. Gd without Zn can not be detected in the X-Y plot. Gd overlaps Zn to 100%.



The linear correlation is nearly 0.7 and the non linear correlation is approx. 0.8. Zn without Ni can be determined. Ni overlaps Zn to 100%.

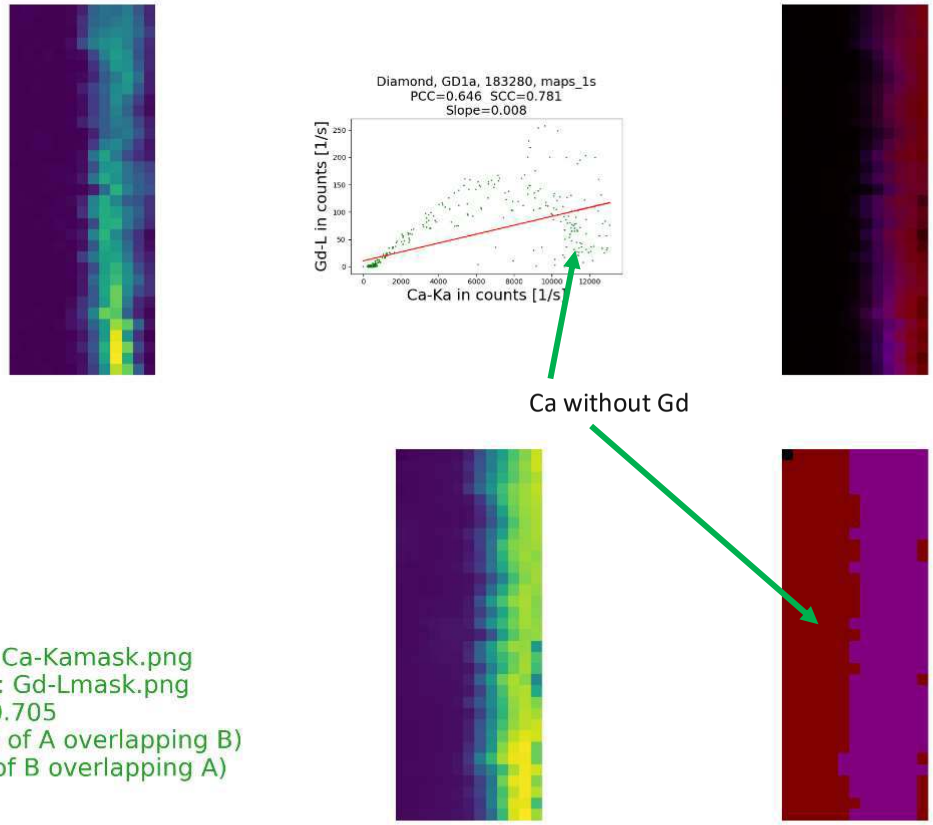


Fe overlaps Ca near to 100%.

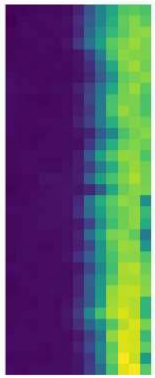


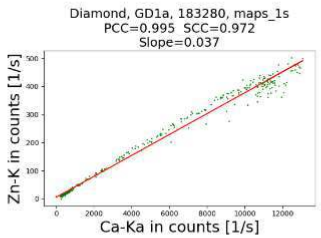
(Red) Image A: Ca-Kamask.png
(Blue) Image B: Fe-Kmask.png
M=0.879
M1=0.775 (fraction of A overlapping B)
M2=0.996 (fraction of B overlapping A)

The linear correlation is approx. 0.8. Gd overlaps Ca to 100%

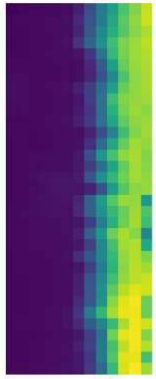


The linear correlation is approx. 1. Ca without can be determined. Zn overlaps Ca to 100%.



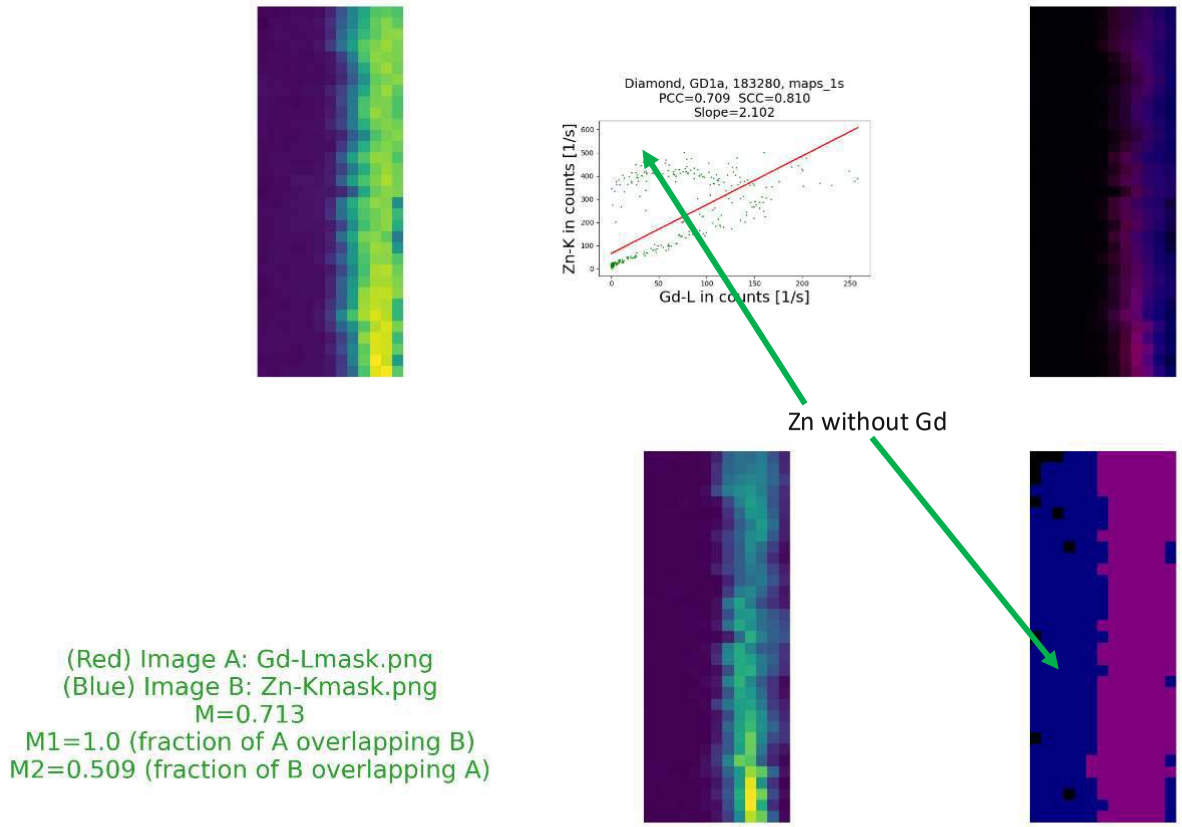


Ca without Zn

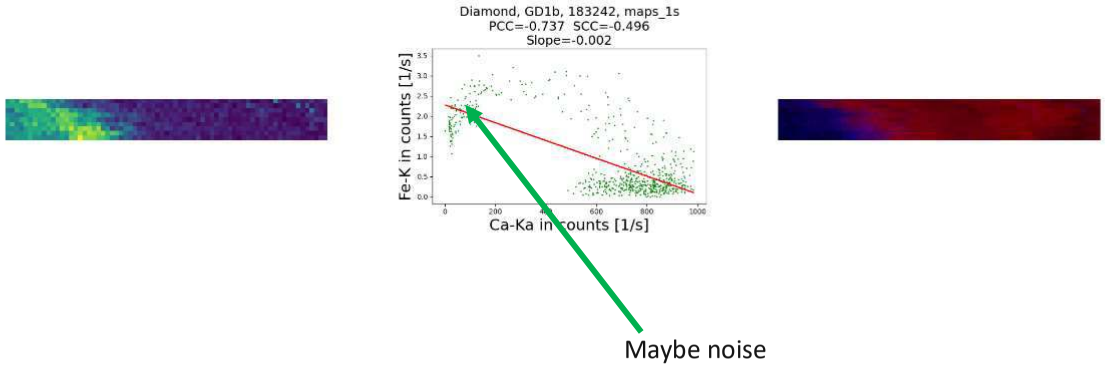


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.988
M1=0.976 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The non linear correlation is approx. 0.8. Gd without Zn can not be determined. Zn without Gd can be shown. Gd overlaps Zn to 100%.

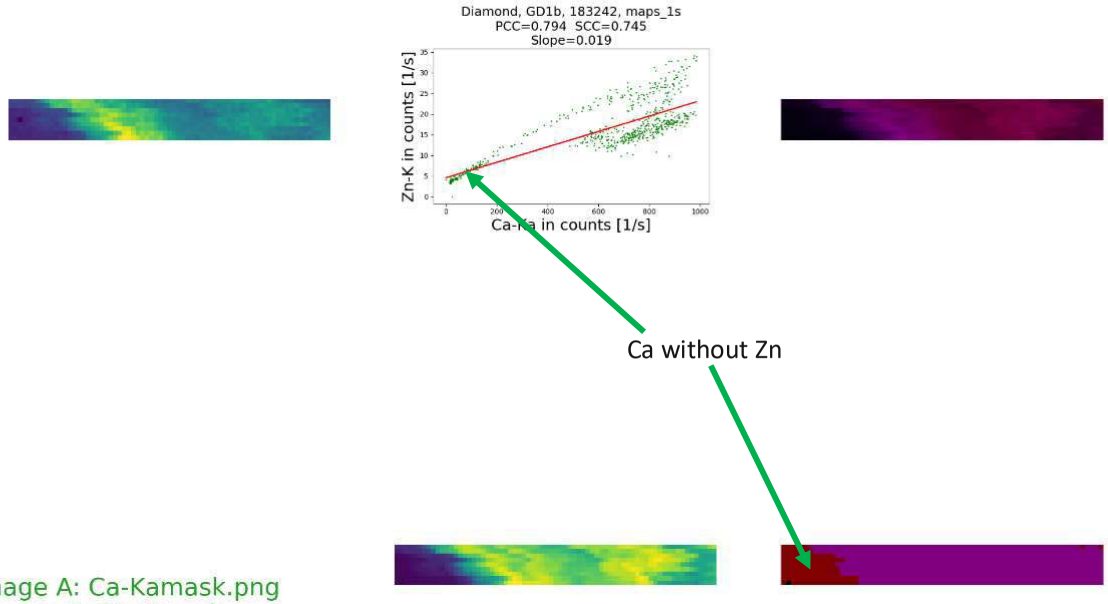


The non linear correlation is approx. 0.8, but Fe is under the threshold level and it can not be shown if Fe is present.

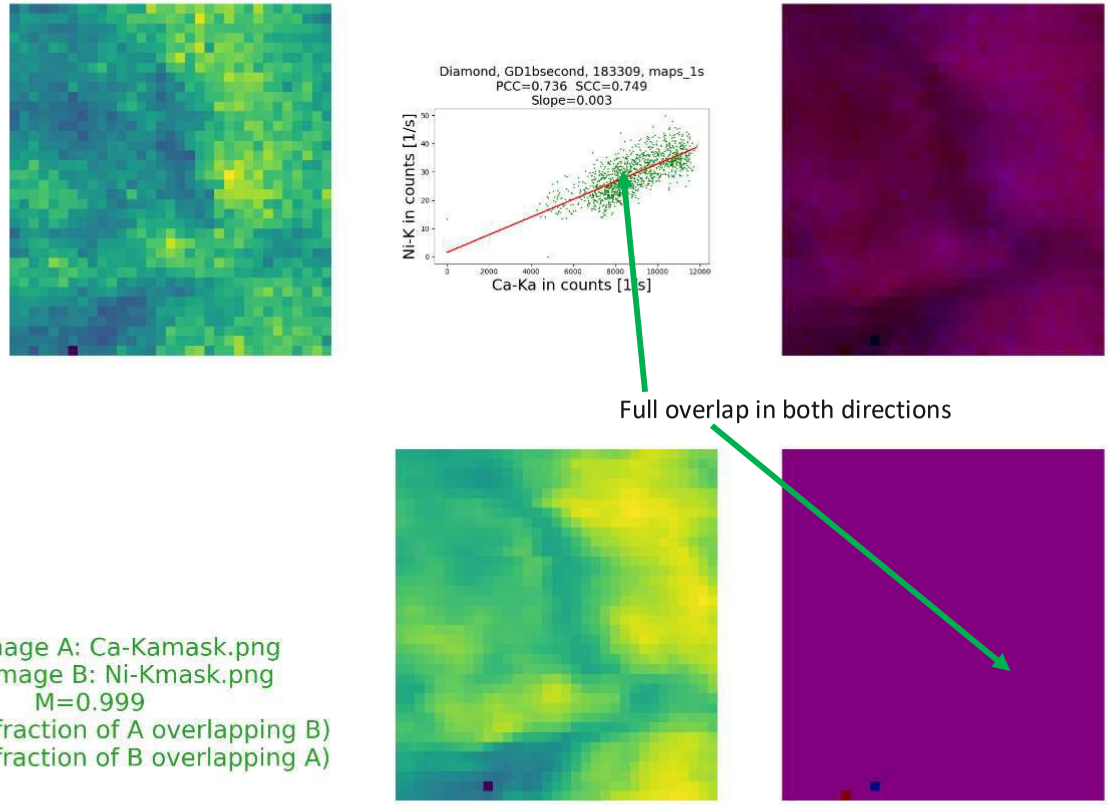


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Fe-Kmask.png
M=0.0
M1=0.0 (fraction of A overlapping B)
M2=0.0 (fraction of B overlapping A)

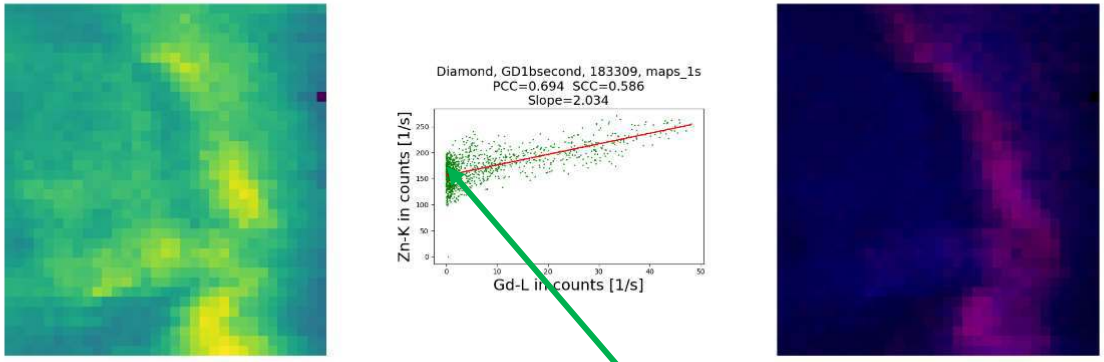
The linear correlation is approx. 0.8. Ca without Zn can be shown. Zn overlaps Ca to 100%.



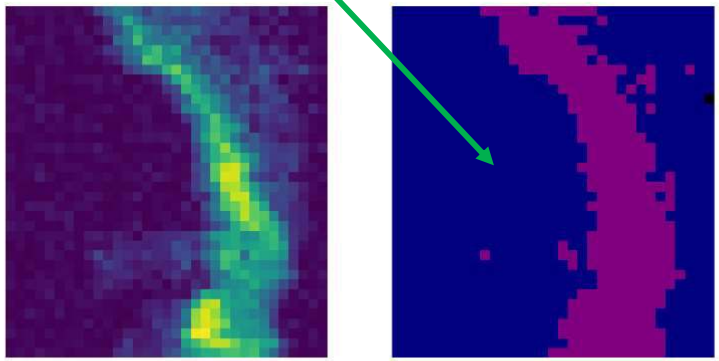
The non linear correlation is approx. 0.8. The full overlap can be shown in both directions.



The linear correlation is approx. 0.7. Zn without Gd can be determined. Gd overlaps Zn to 100%.



Zn without Gd

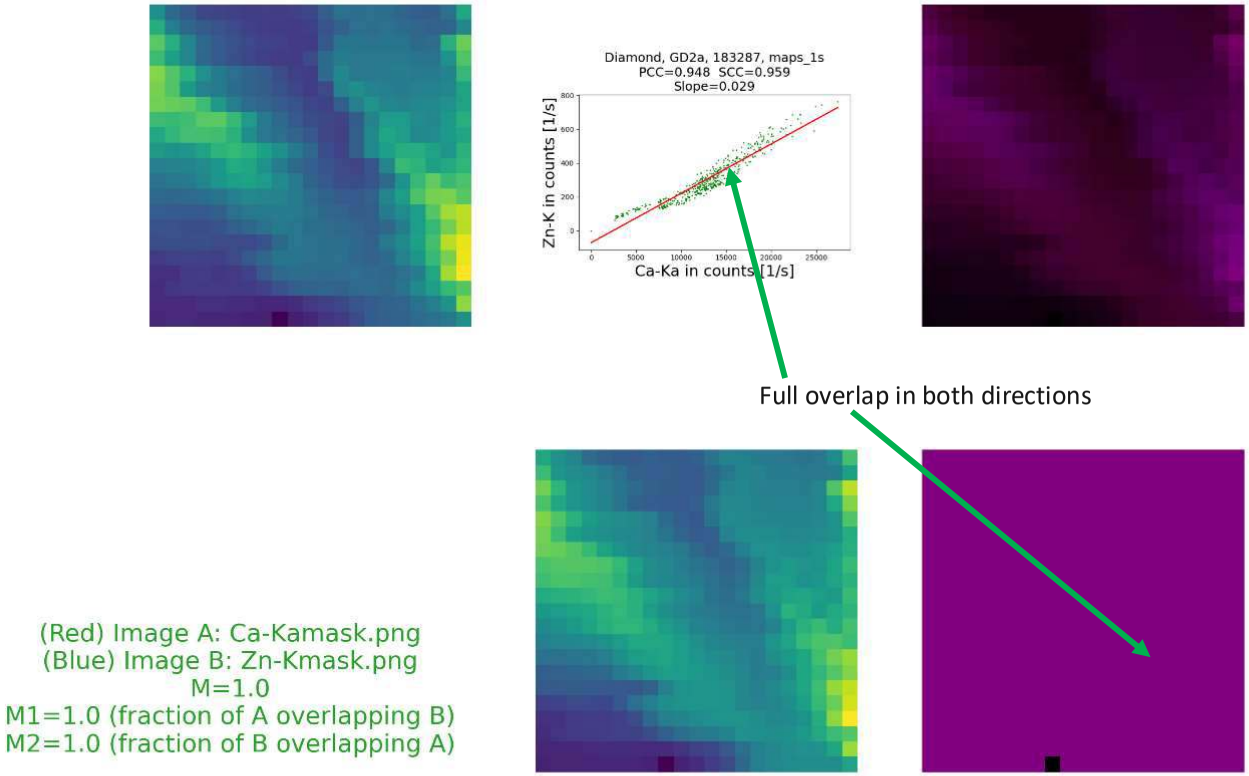


(Red) Image A: Gd-Lmask.png
(Blue) Image B: Zn-Kmask.png
M=0.514
M1=1.0 (fraction of A overlapping B)
M2=0.264 (fraction of B overlapping A)

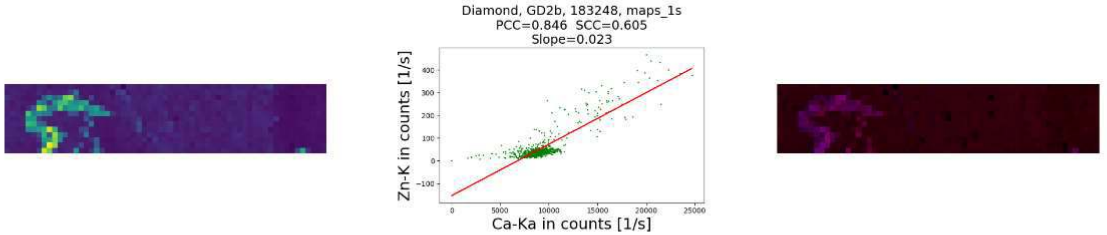
GD2

183287

The non linear correlation is approx. 1. The full overlap can be shown in both directions.



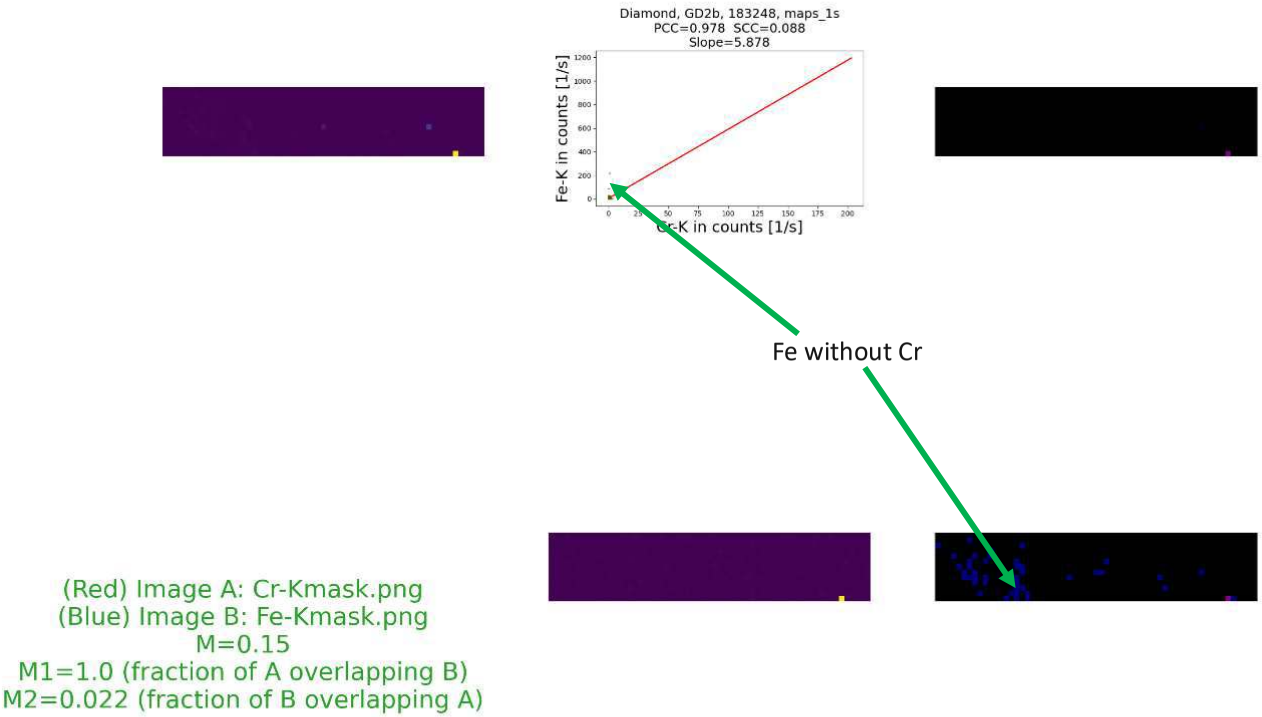
The linear correlation is approx. 0.9. The full overlap can be shown in both directions.



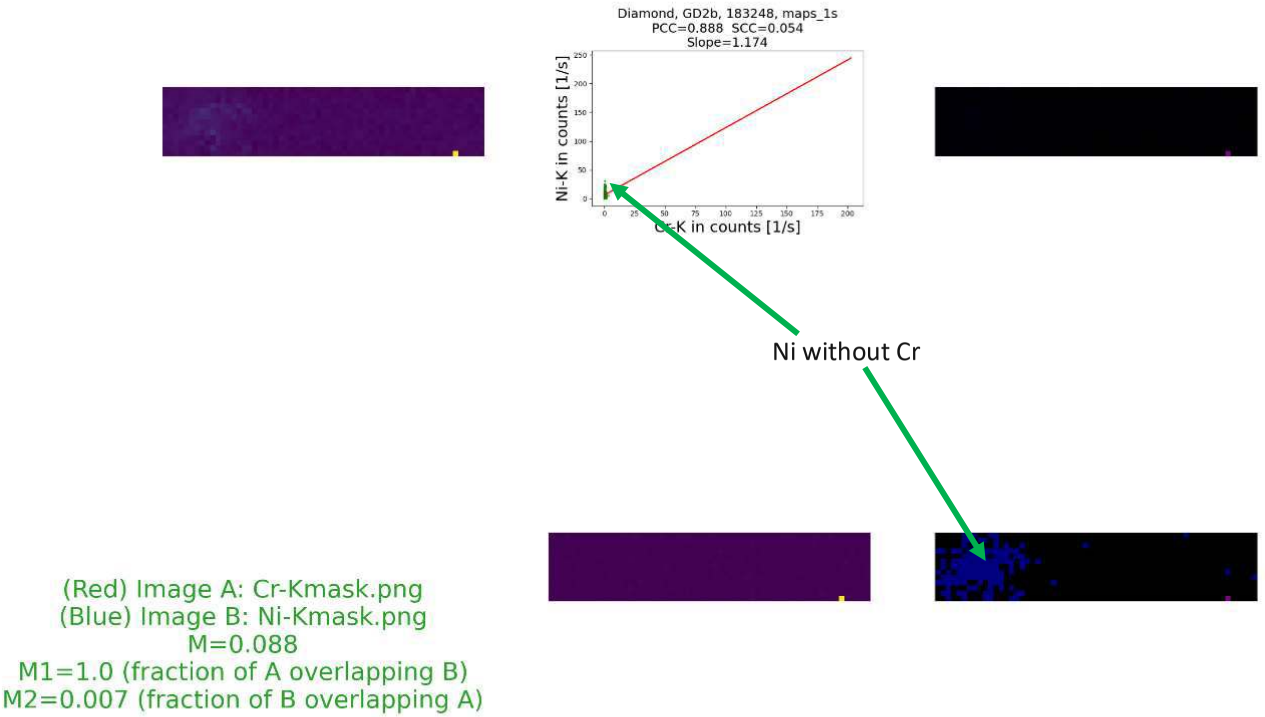
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.999
M1=0.998 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

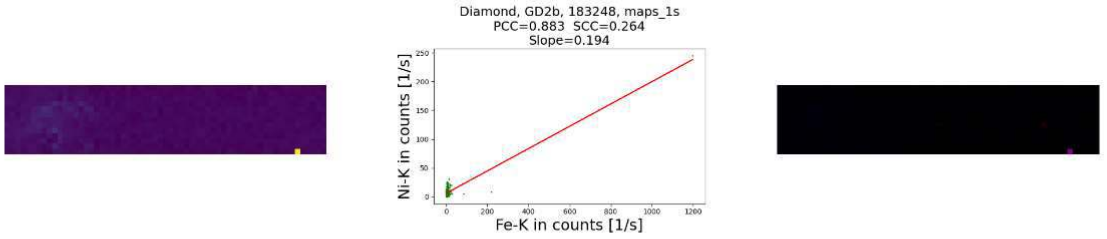
The linear correlation is approx. 1, but Cr is under the threshold level, and it can not be shown if Cr is present.

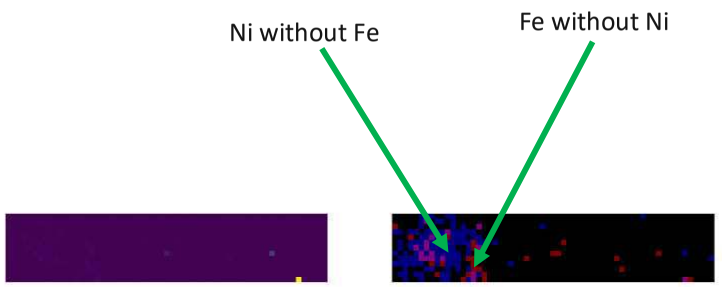


The linear correlation is approx. 0.9, but Cr is under the threshold level, and it can not be shown if Cr is present.



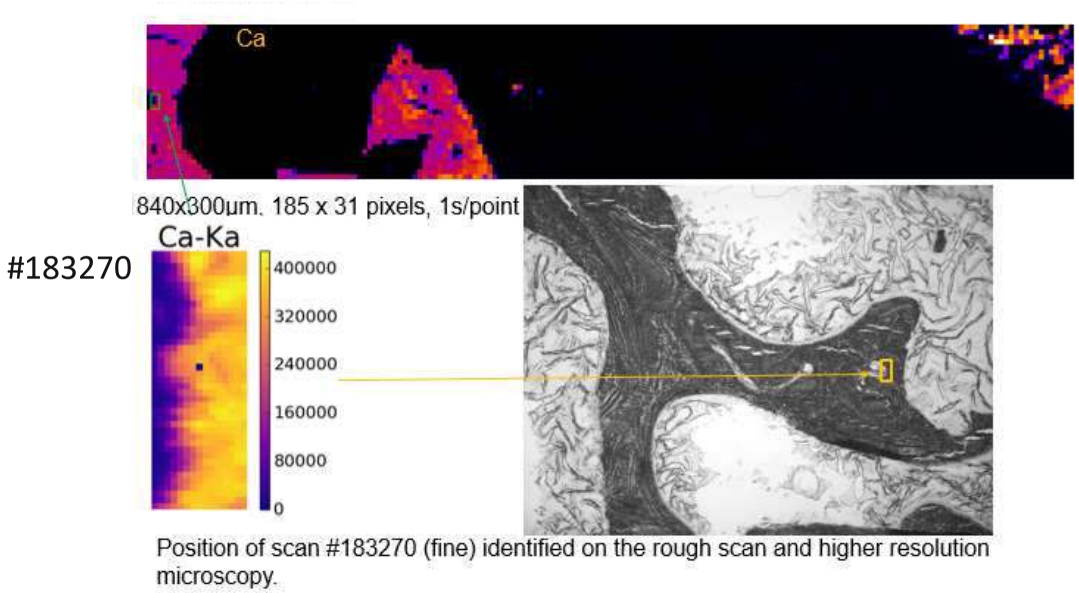
The linear correlation is approx. 0.9. The overlap regions are very low, in both directions.





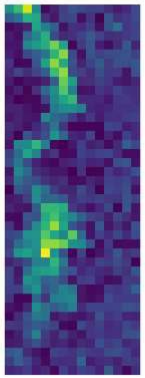
(Red) Image A: Fe-Kmask.png
(Blue) Image B: Ni-Kmask.png
M=0.24
M1=0.409 (fraction of A overlapping B)
M2=0.141 (fraction of B overlapping A)

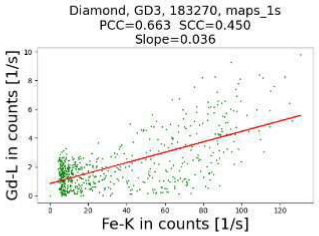
GD3



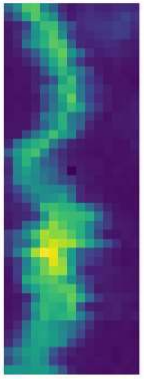
Position of scan #183270 (fine) identified on the rough scan and higher resolution microscopy.

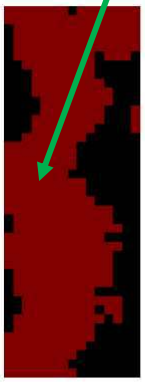
The linear correlation is approx. 0.7. Gd is under the threshold level, and it can not be shown if Gd is present.





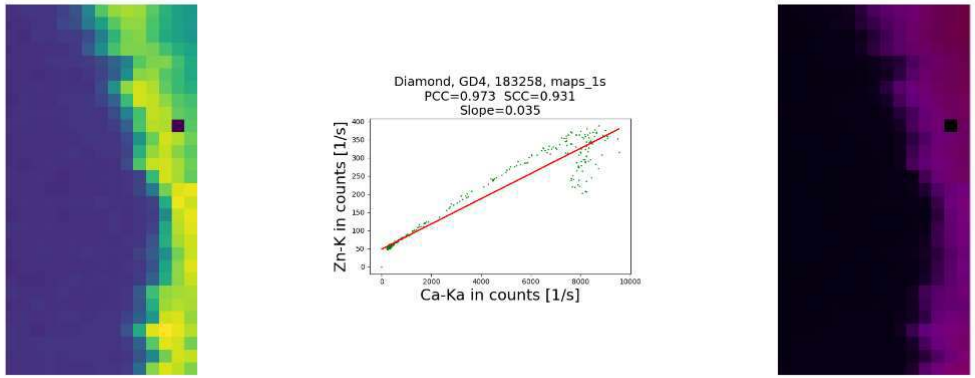
Fe without Gd





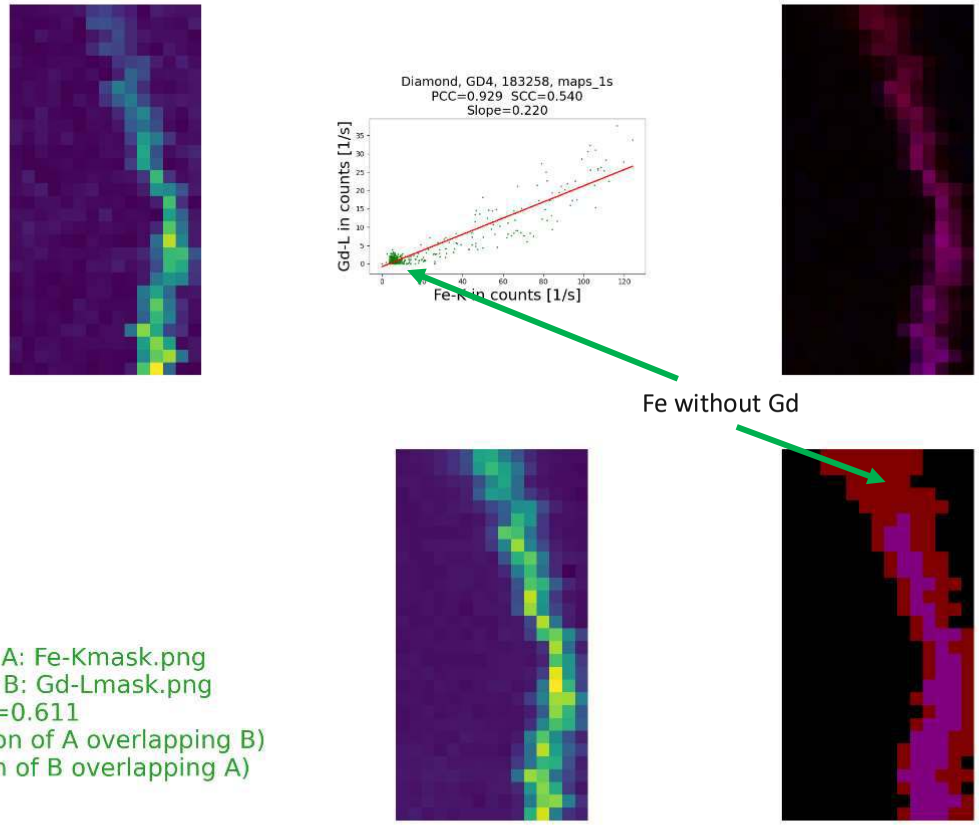
(Red) Image A: Fe-Kmask.png
(Blue) Image B: Gd-Lmask.png
M=0.0
M1=0.0 (fraction of A overlapping B)
M2=0.0 (fraction of B overlapping A)

The linear correlation is approx. 1. The full overlap in both direction could be shown.

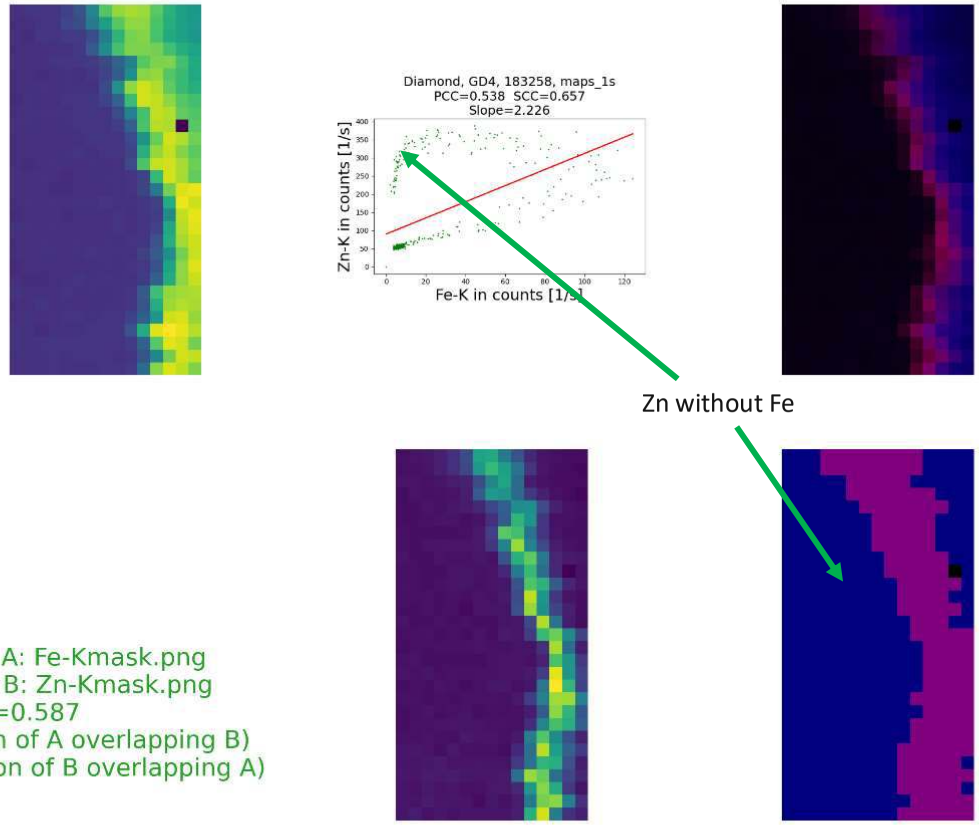


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=1.0
M1=1.0 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The linear correlation is approx. 1. Fe without Gd can be shown. Gd overlaps Fe to 100%.



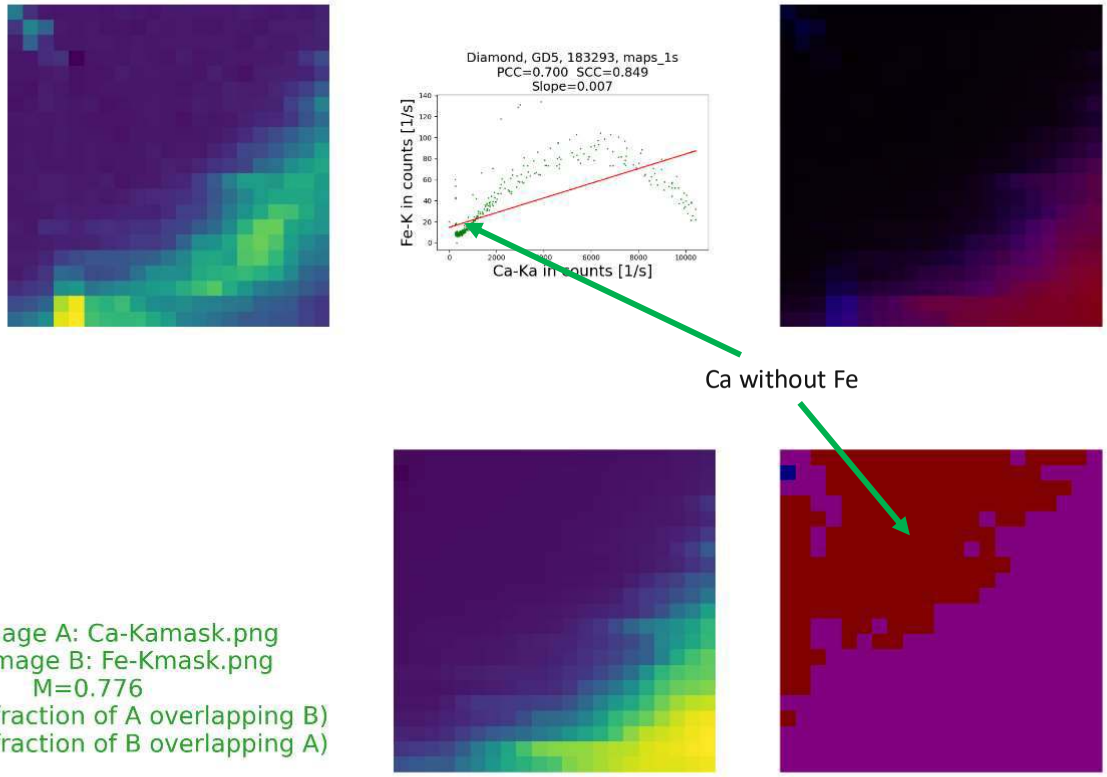
The non linear correlation is approx. 0.7. Zn without Fe can be shown. Fe overlaps Zn to 100%.



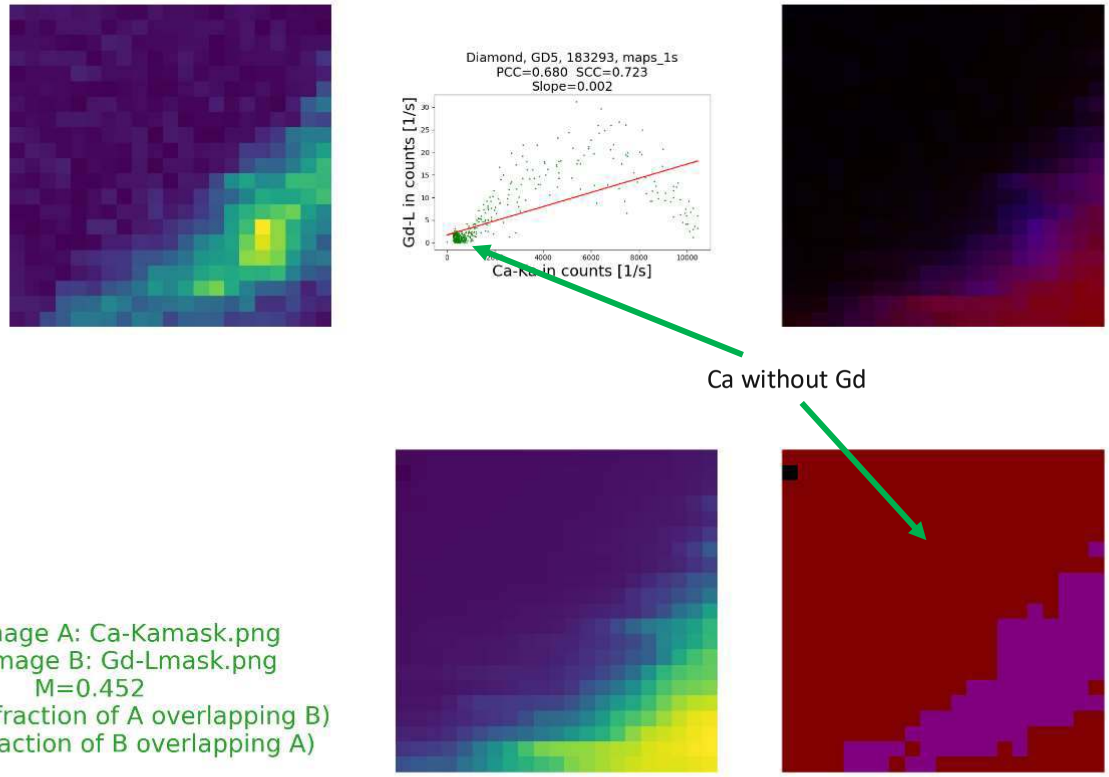
(Red) Image A: Fe-Kmask.png
(Blue) Image B: Zn-Kmask.png
M=0.587
M1=1.0 (fraction of A overlapping B)
M2=0.345 (fraction of B overlapping A)

GD5

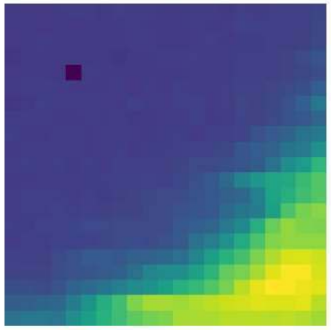
The non linear correlation is approx. 0.9. Fe overlaps Ca near to 100%.

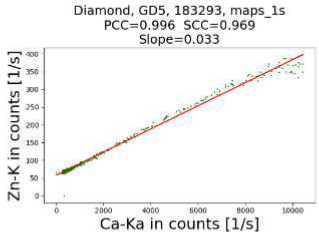


The non linear correlation is approx. 0.7. Gd overlaps Ca to 100%.



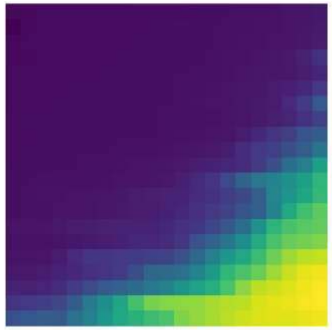
The linear correlation is approx. 1. The full overlap in both directions can be shown.

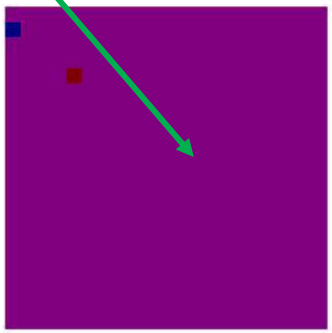




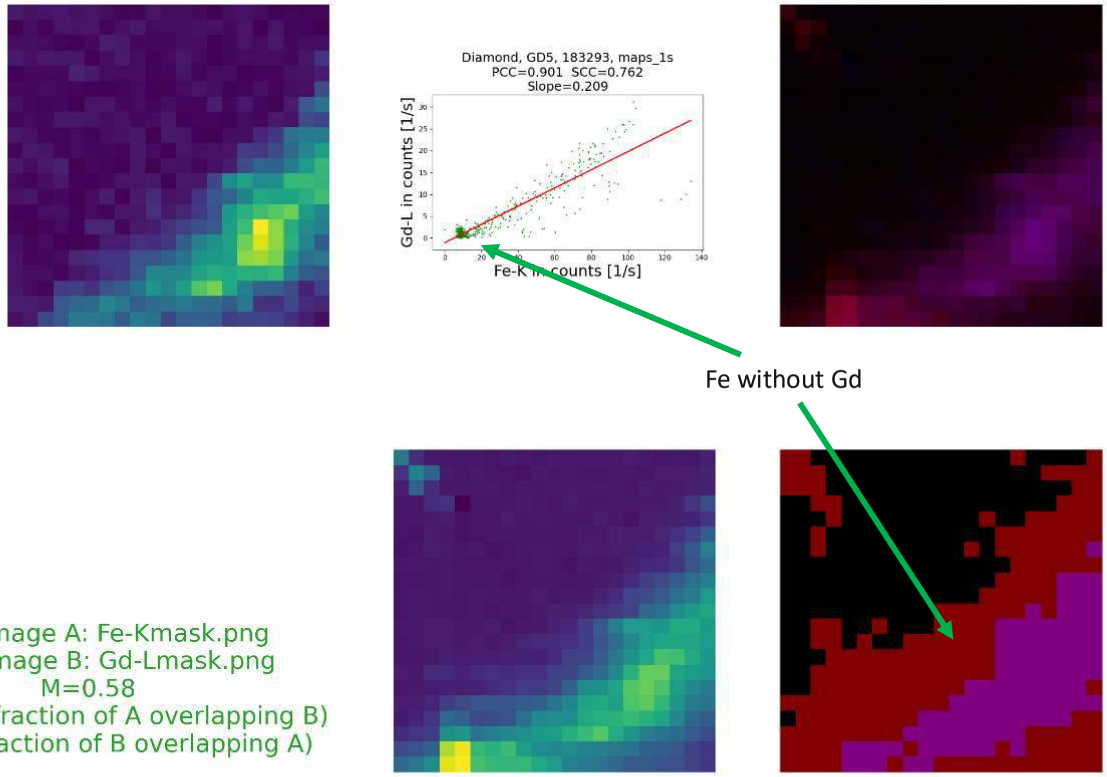
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.997
M1=0.997 (fraction of A overlapping B)
M2=0.997 (fraction of B overlapping A)

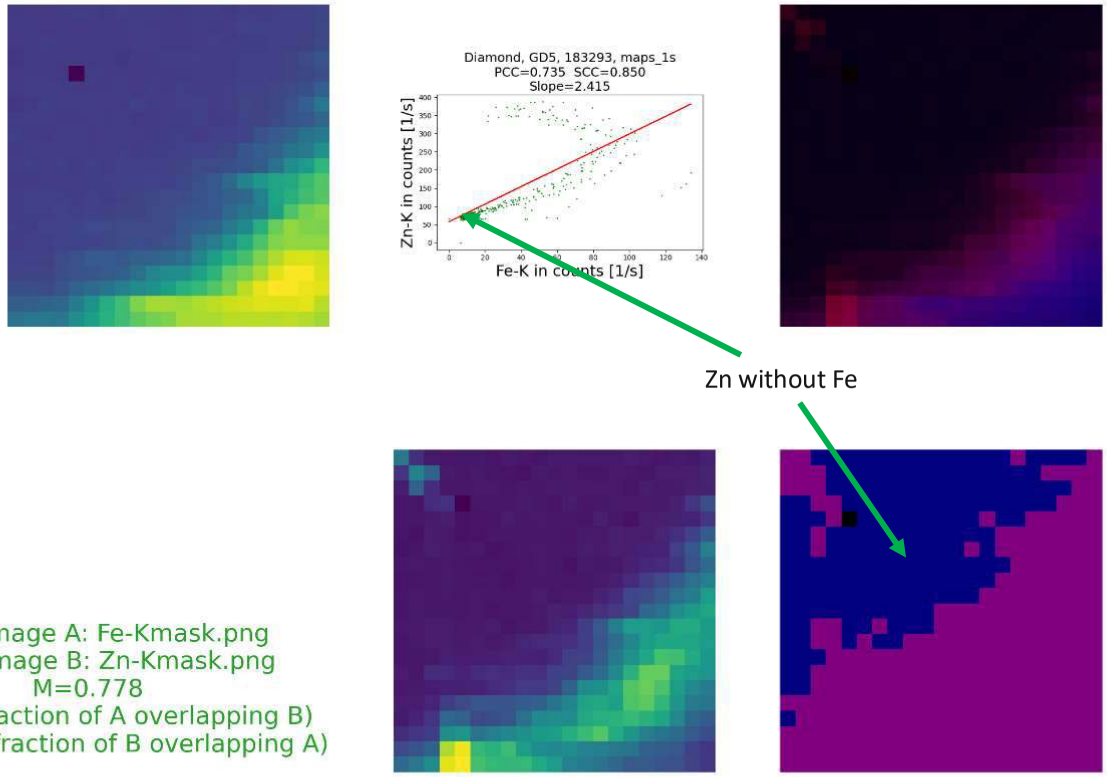




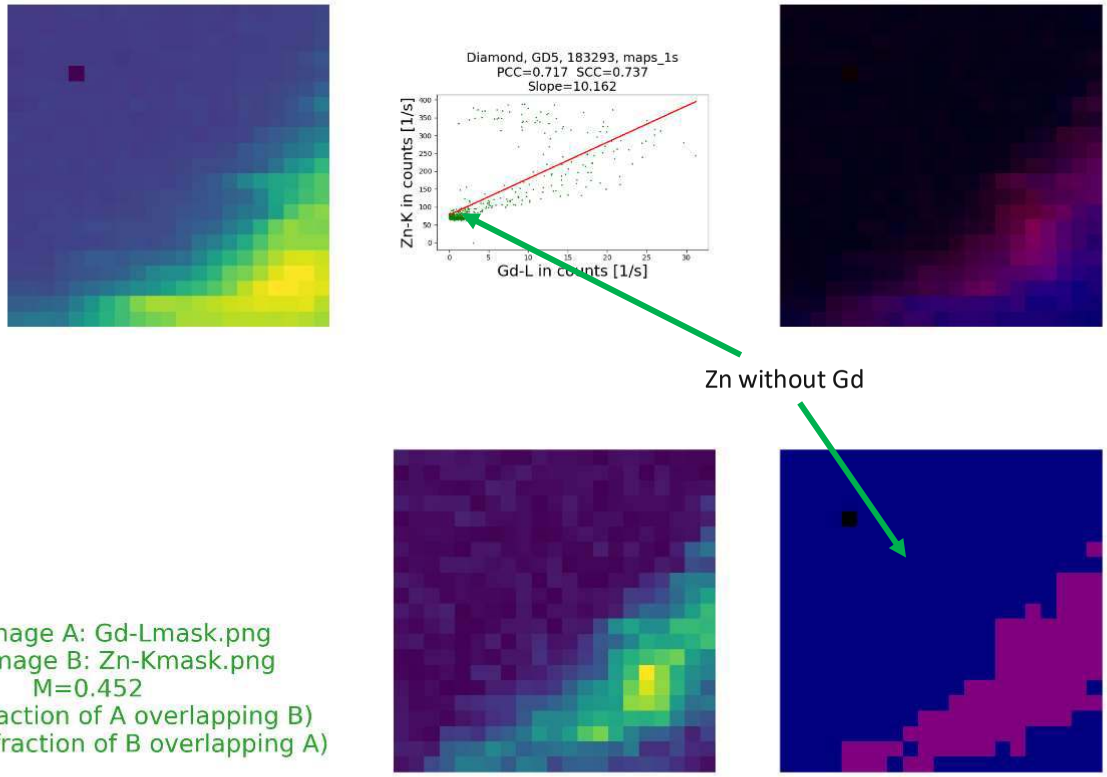
The linear correlation is approx. 0.9. Fe without Gd can be shown. Gd overlaps Fe to 100%.



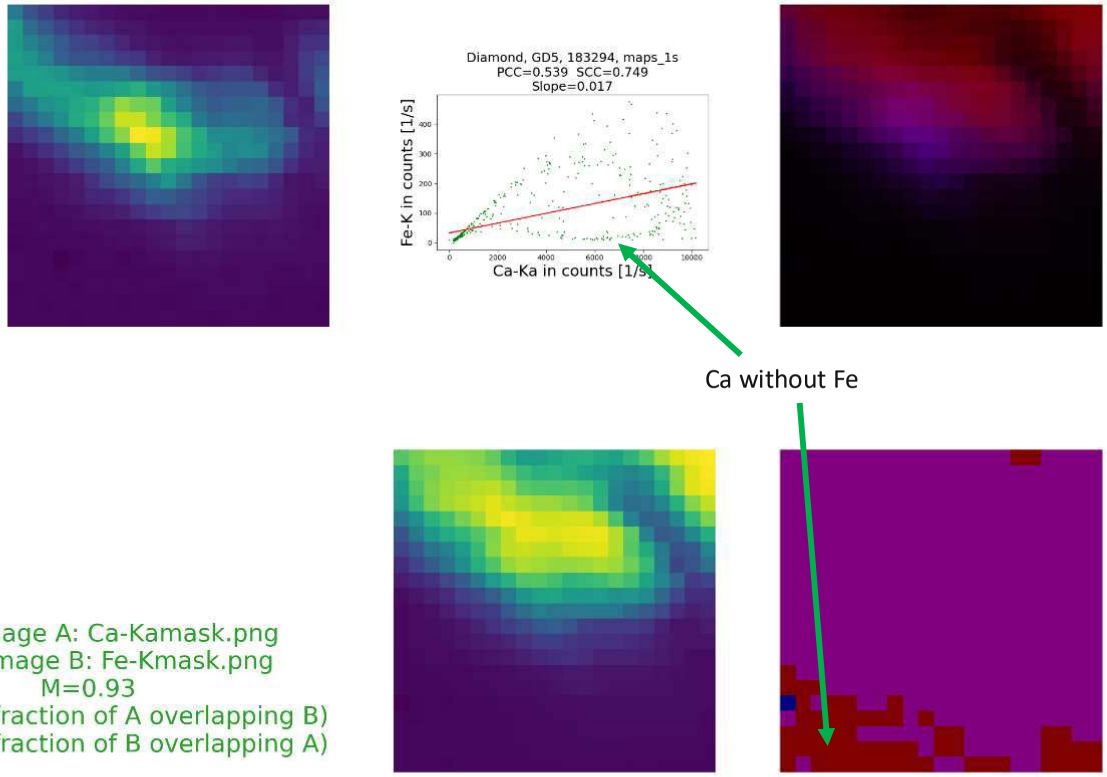
The non linear correlation is approx. 0.9. Zn without Fe can be shown. Fe overlaps Zn to 100%.



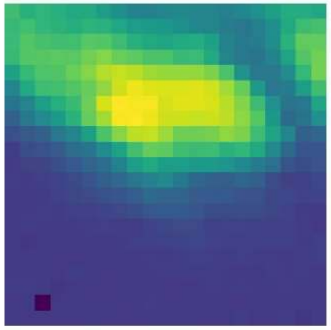
The linear correlation is approx. 0.7. Zn without Gd can be shown. Gd overlaps Zn to 100%.

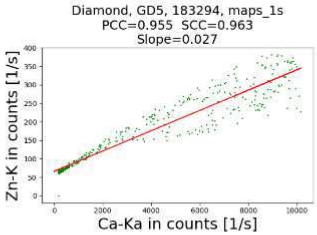


The non linear correlation is approx. 0.7. Fe overlaps Ca near to 100%.



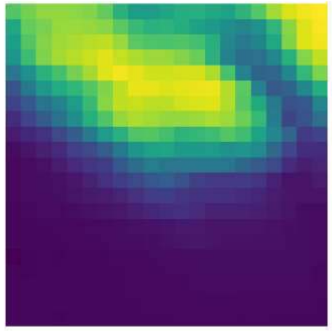
The linear correlation is approx. 1. The full overlap in both directions can be shown.

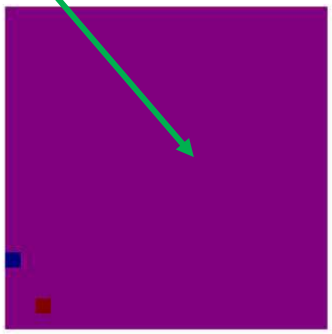




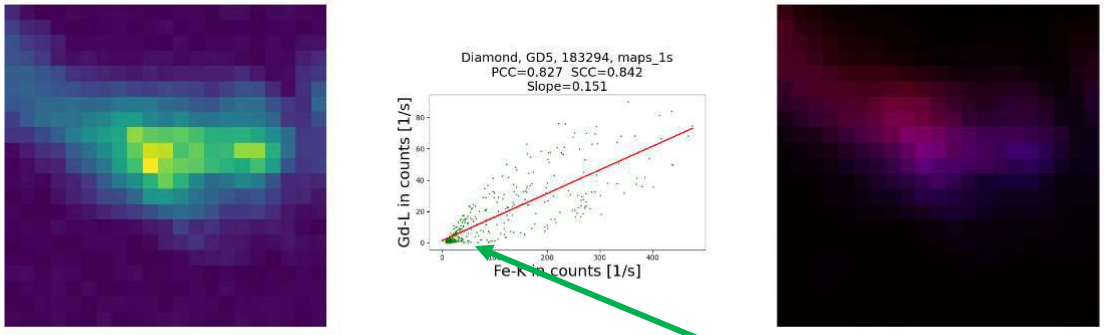
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.997
M1=0.997 (fraction of A overlapping B)
M2=0.997 (fraction of B overlapping A)

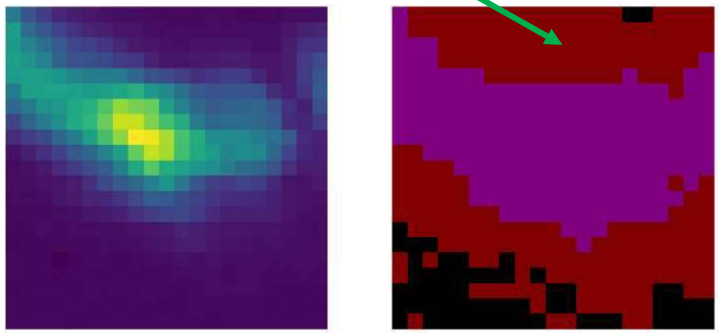




The non linear correlation is approx. 0.8. Gd overlaps Fe to 100%.

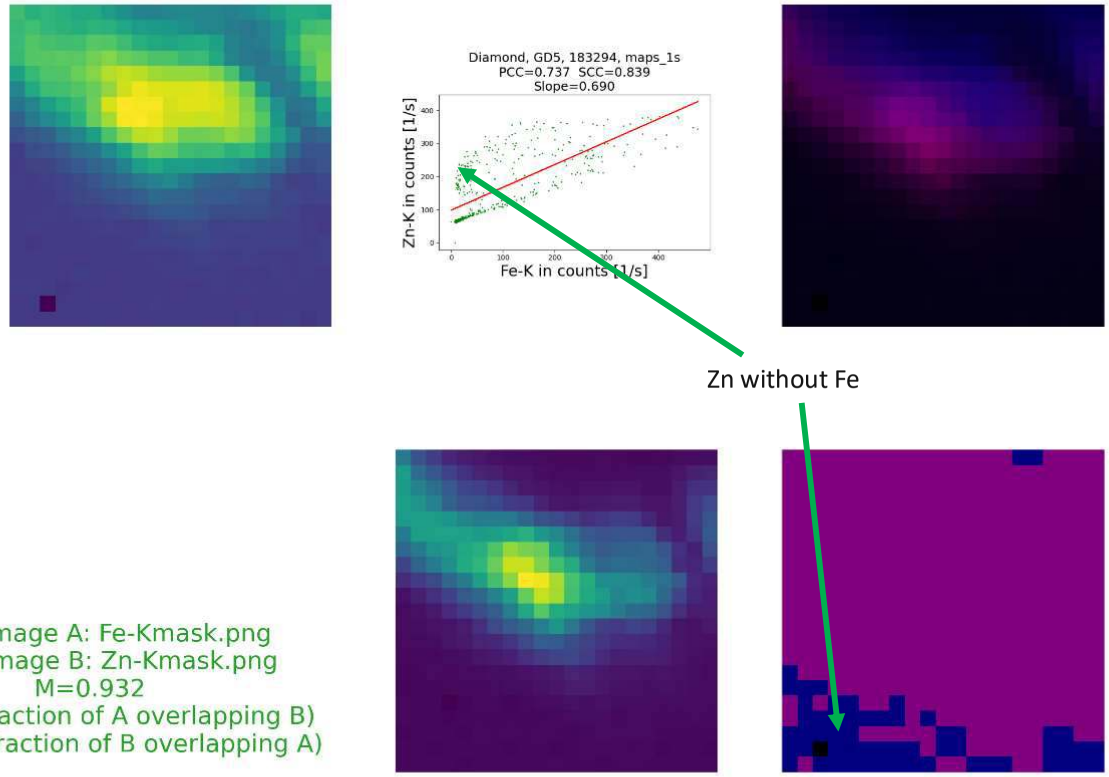


Fe without Gd

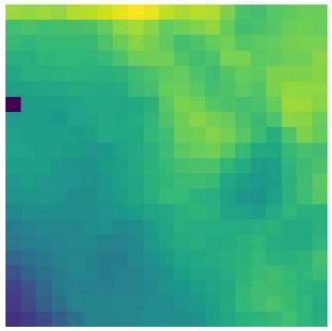


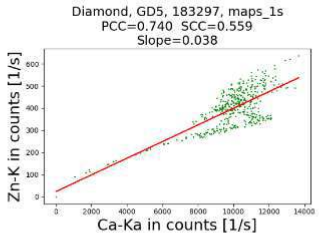
(Red) Image A: Fe-Kmask.png
(Blue) Image B: Gd-Lmask.png
M=0.685
M1=0.469 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The non linear correlation is approx. 0.8. Fe overlaps Zn to 100%.



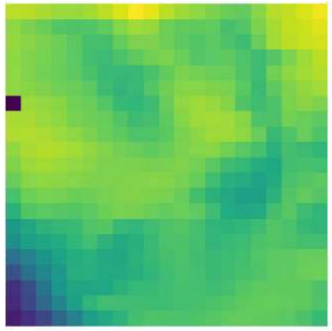
The linear correlation is approx. 0.7. The full overlap in both directions can be shown.

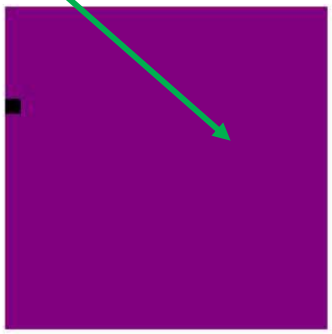




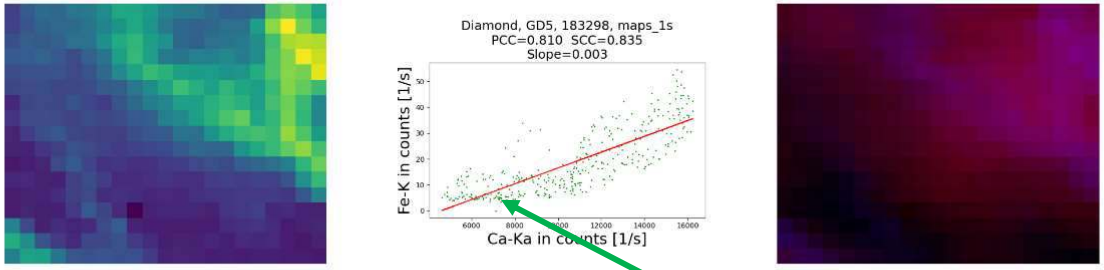
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=1.0
M1=1.0 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

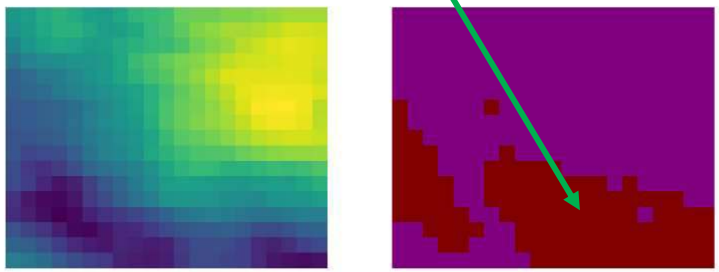




The non linear correlation is approx. 0.8. Fe overlaps Ca approx. to 100%.

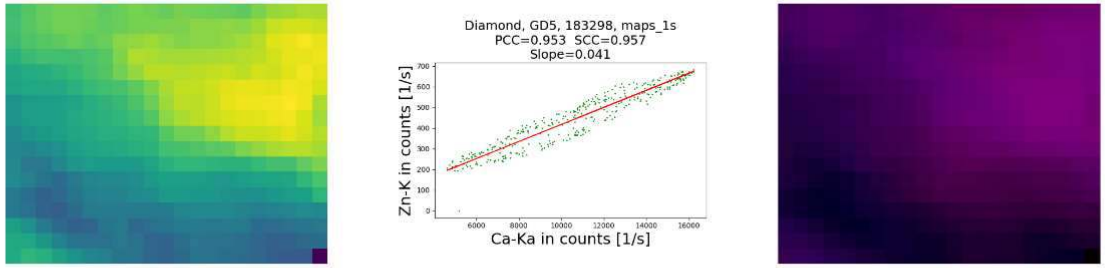


Ca without Fe

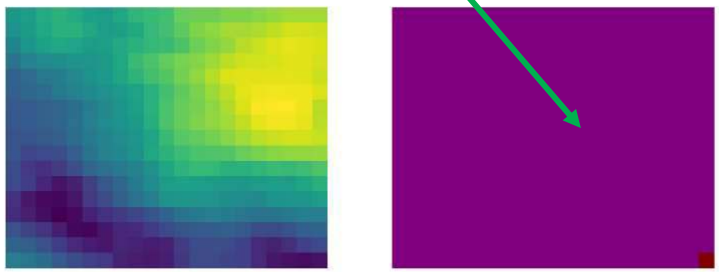


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Fe-Kmask.png
M=0.833
M1=0.694 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The linear correlation is approx. 1. The full overlap in both directions can be shown.

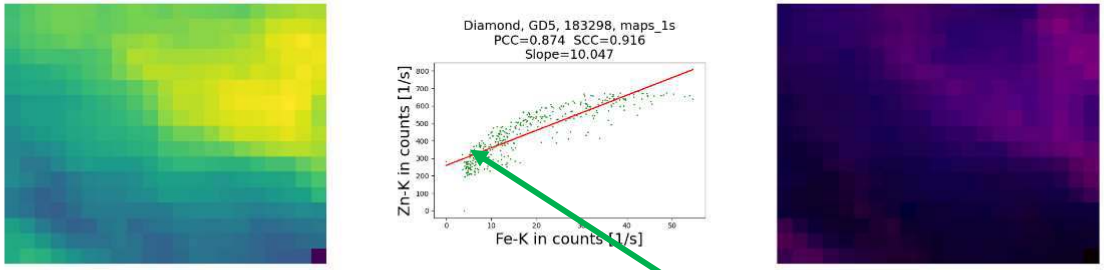


Full overlap in both directions

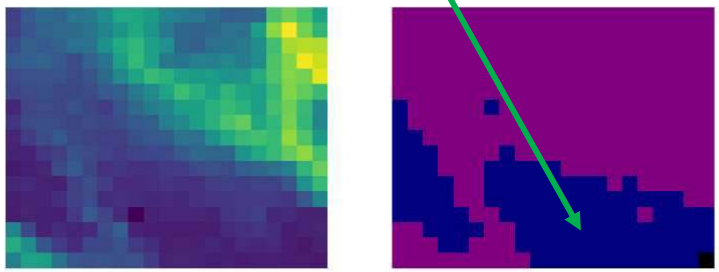


(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.998
M1=0.997 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The non linear correlation is approx. 0.9. Fe overlaps Zn to 100%.

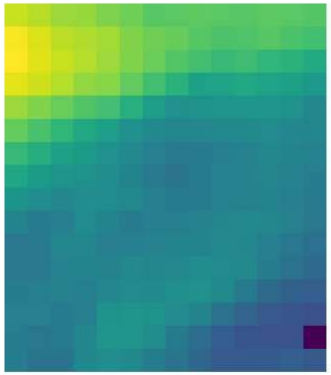


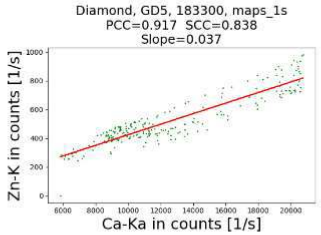
Zn without Fe



(Red) Image A: Fe-Kmask.png
(Blue) Image B: Zn-Kmask.png
M=0.834
M1=1.0 (fraction of A overlapping B)
M2=0.696 (fraction of B overlapping A)

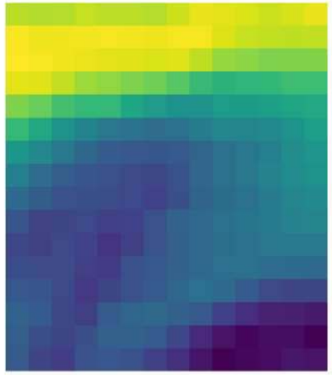
The linear correlation is approx. 0.9. The full overlap in both directions can be shown.

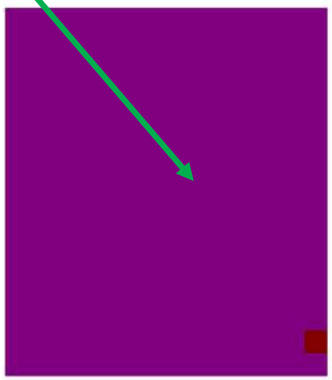




Full overlap in both directions

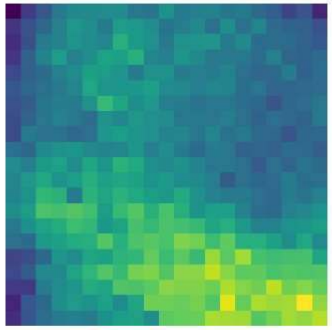
(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.997
M1=0.995 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

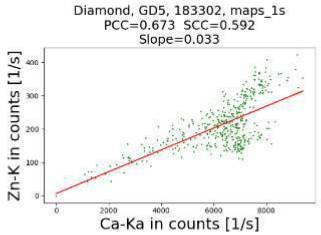




GD5

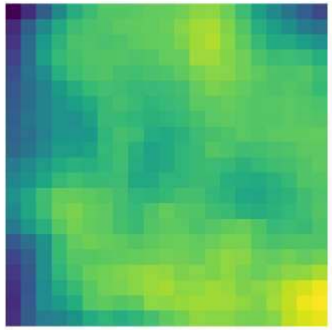
The linear correlation is approx. 0.7. The full overlap in both directions can be shown.

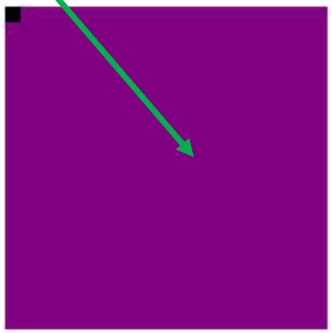




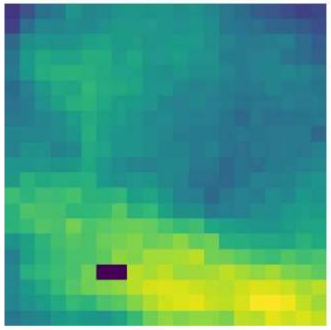
Full overlap in both directions

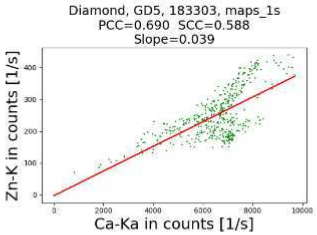
(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=1.0
M1=1.0 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)





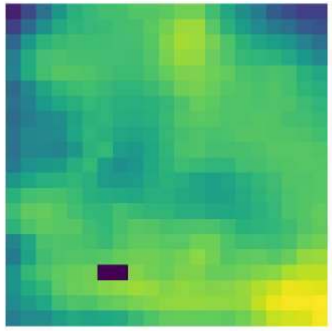
The linear correlation is approx. 0.7. The full overlap in both directions can be shown.

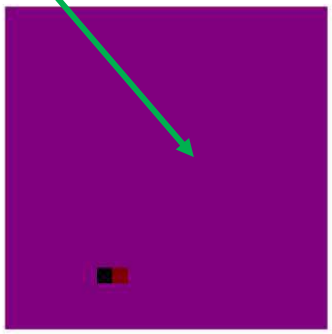




Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.998
M1=0.997 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)





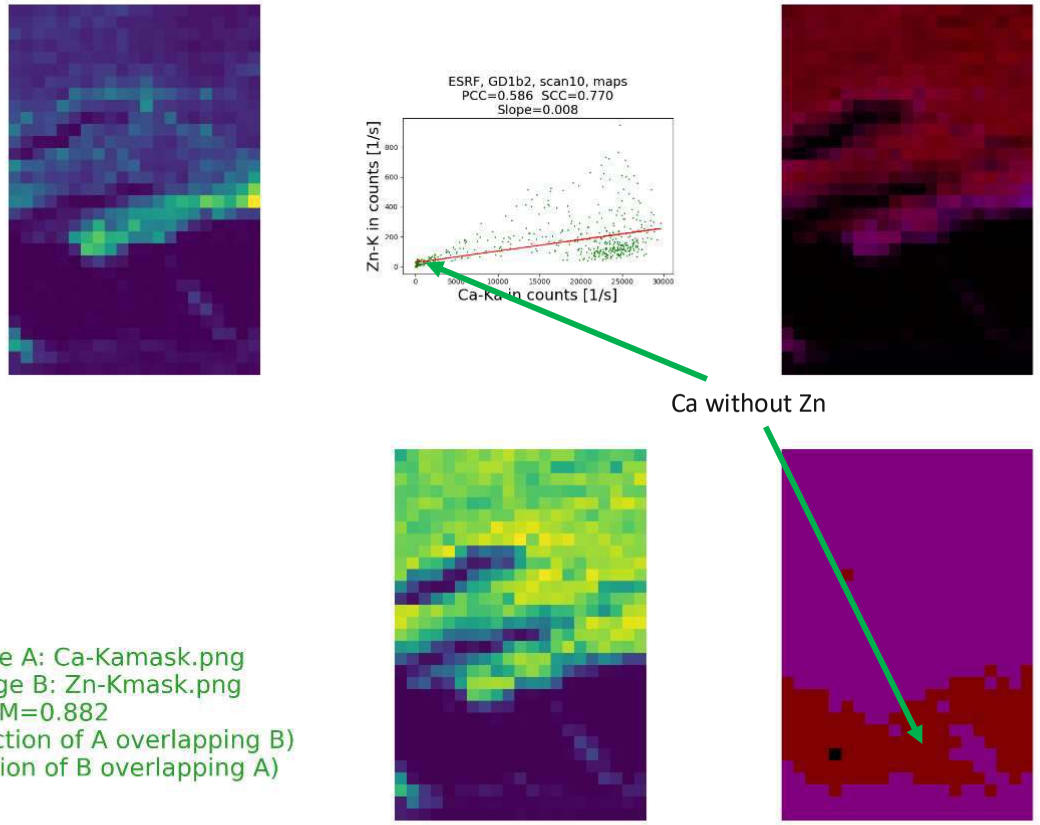
GD1

No Data for Microscopic Images

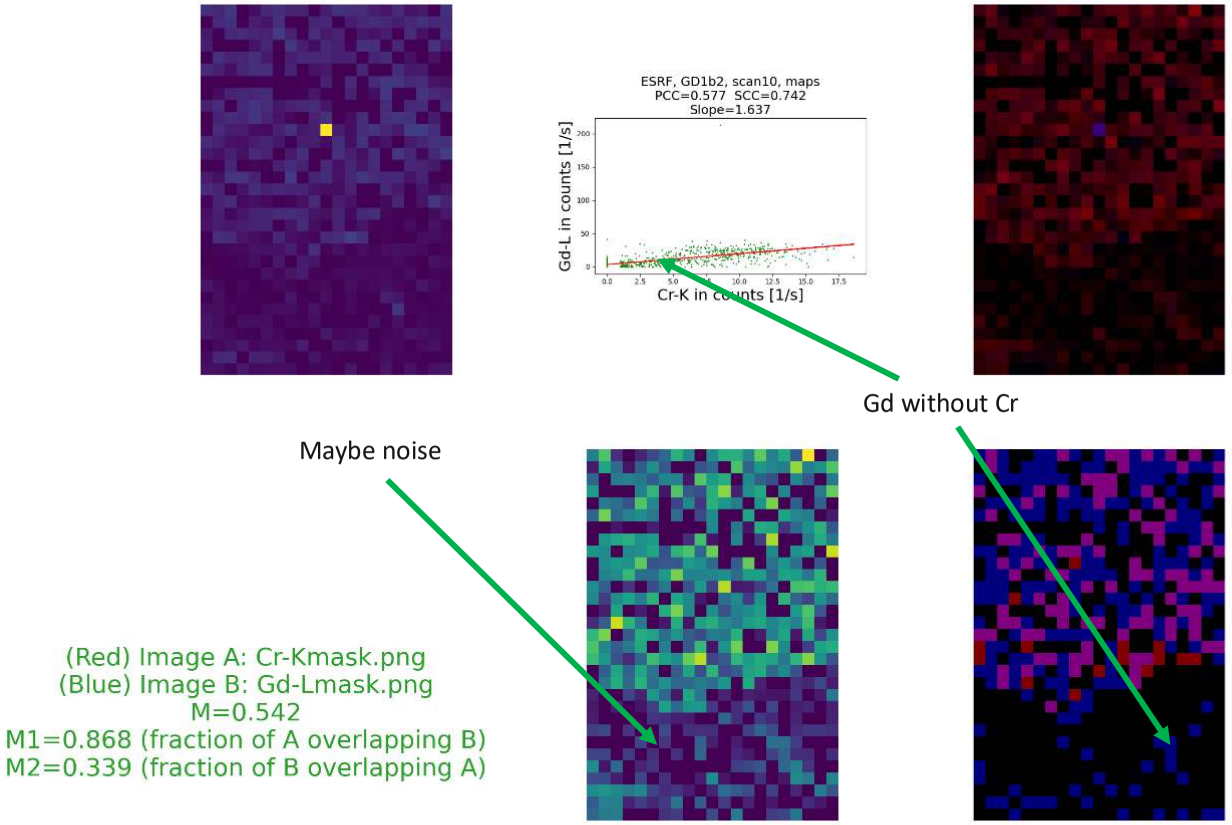
No Data for Rough Scans

GD1b2 scan10

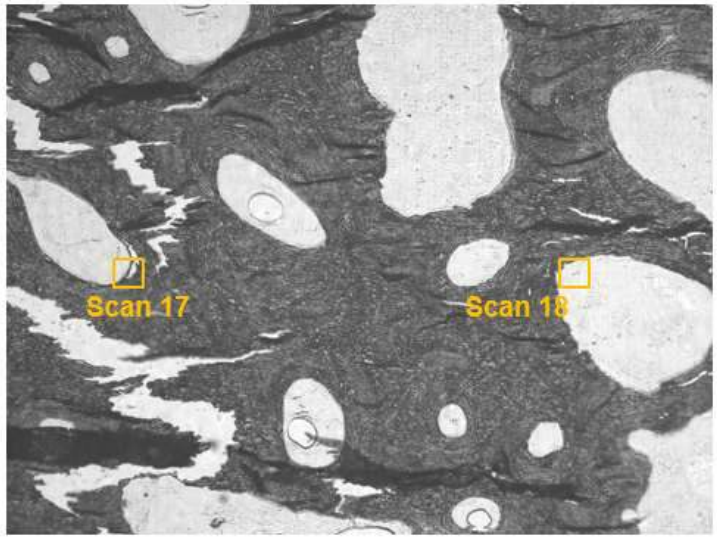
The non linear correlation is approx. 0.8. Zn overlaps Ca to 100%.



The non linear correlation is approx. 0.7. Cr overlaps Gd approx. to 90%. A lot of the Cr signal is under the threshold level.



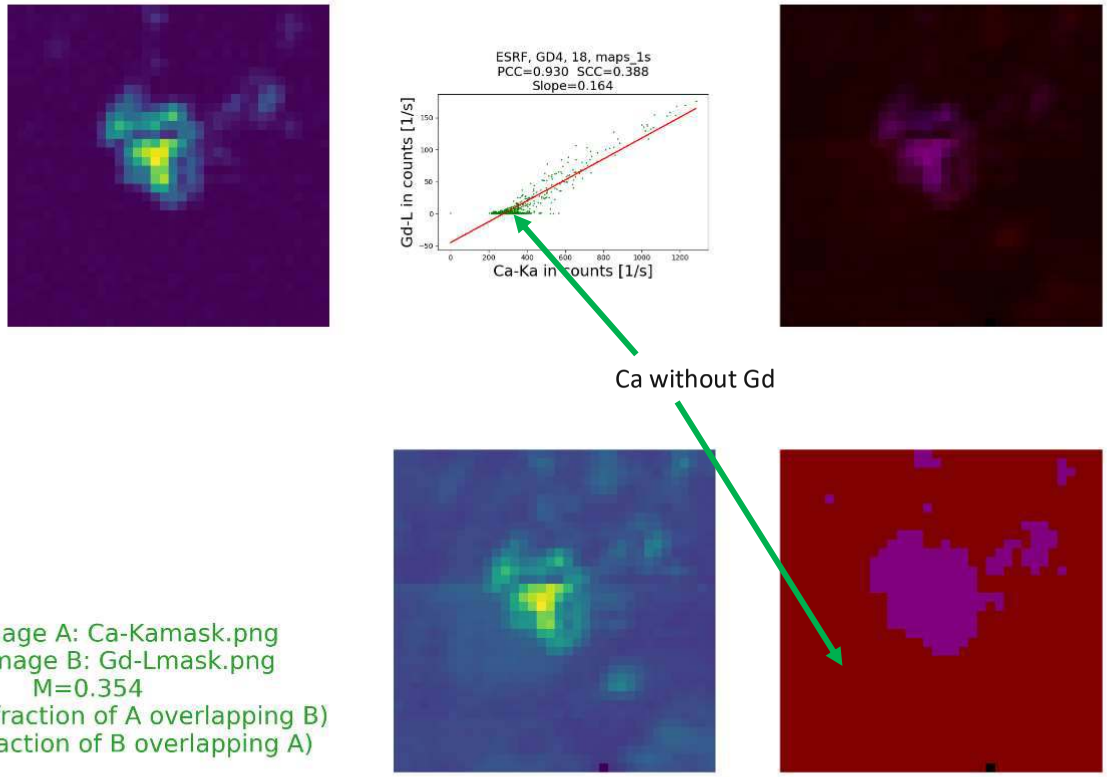
GD4



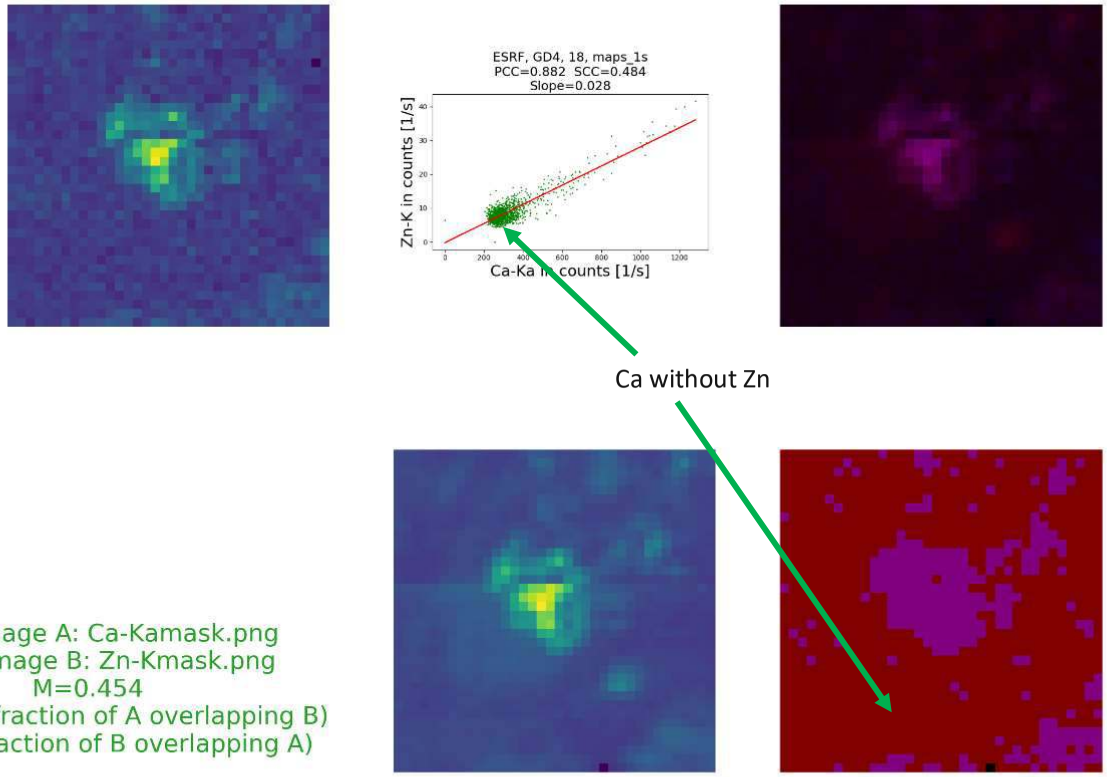
No Data for Rough Scans

GD4 scan18

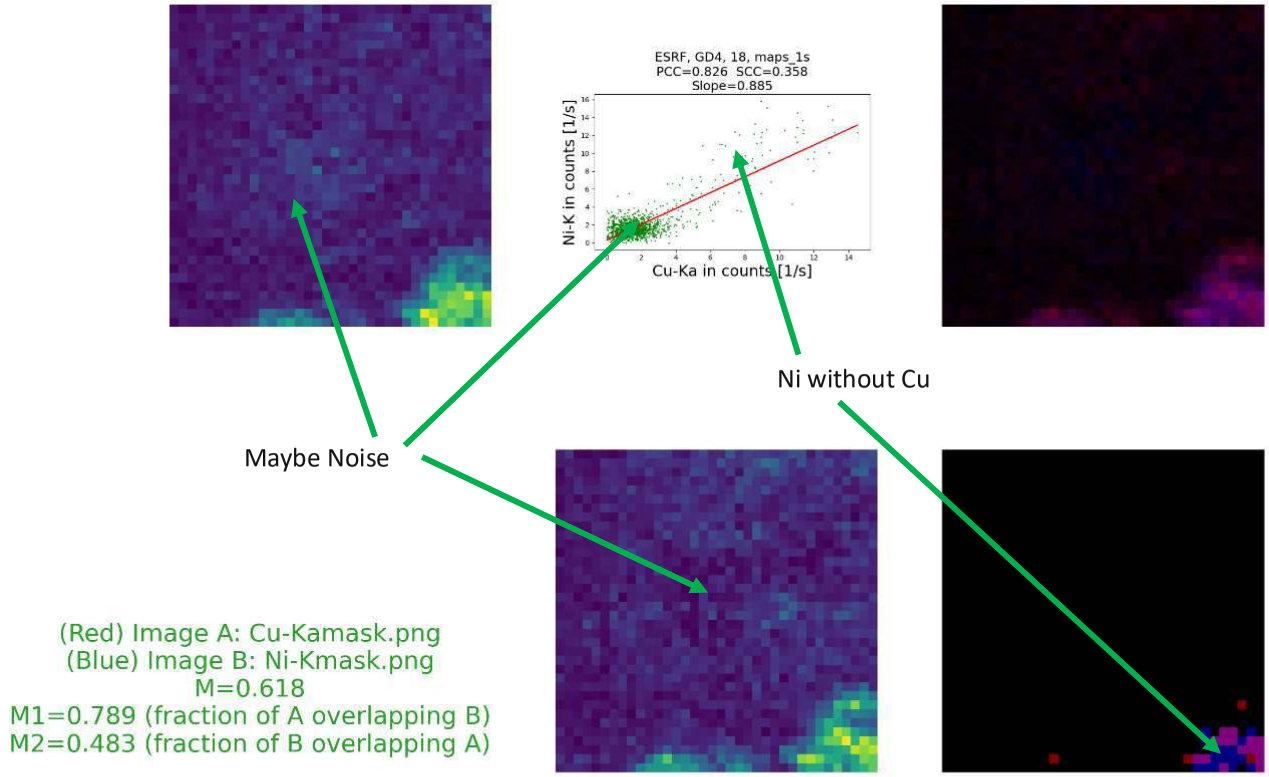
The linear correlation is approx. 0.9. Gd overlaps Ca to 100%



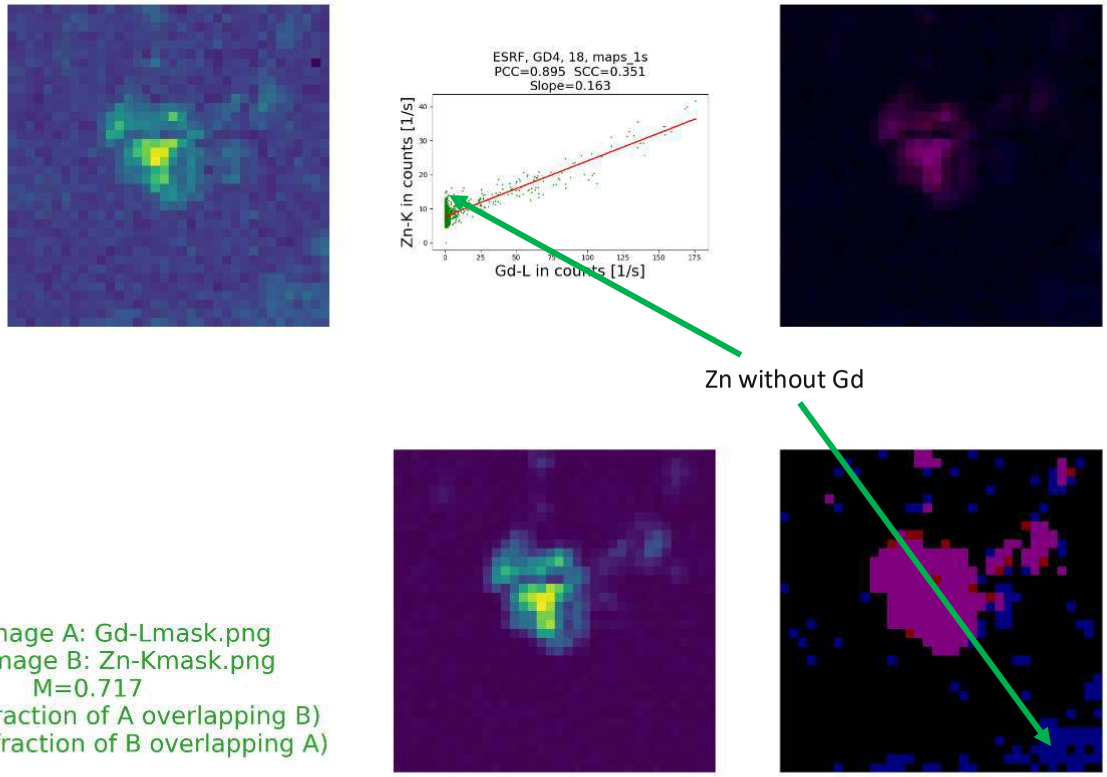
The linear correlation is approx. 0.9. Zn overlaps Ca to 100%



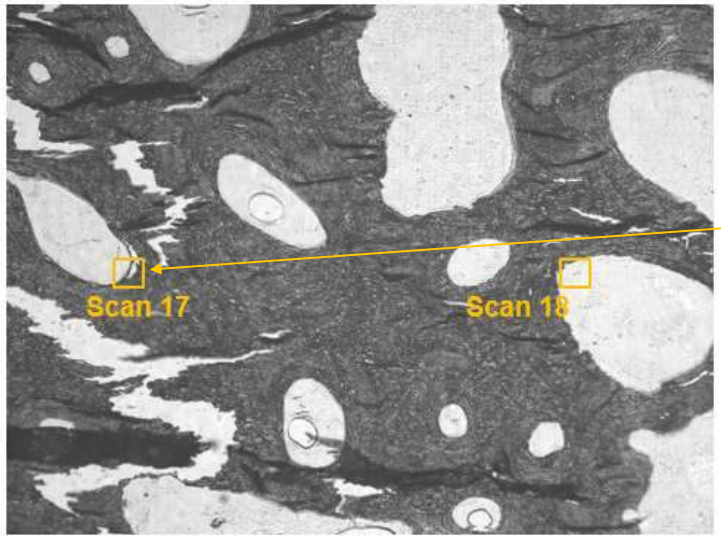
The linear correlation is approx. 0.8. Ni overlaps Cu to approx. 80%. A lot of the Ni and Cu signal is under the threshold level.

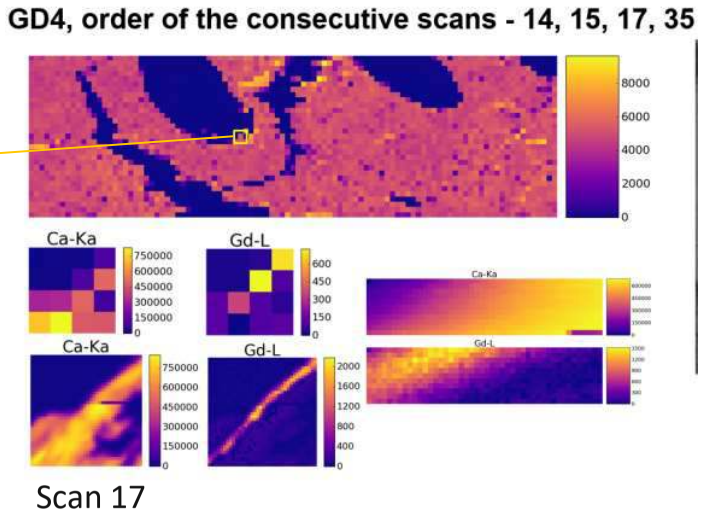


The linear correlation is approx. 0.9. Zn without Gd can be shown. Gd overlaps Zn to 100%.



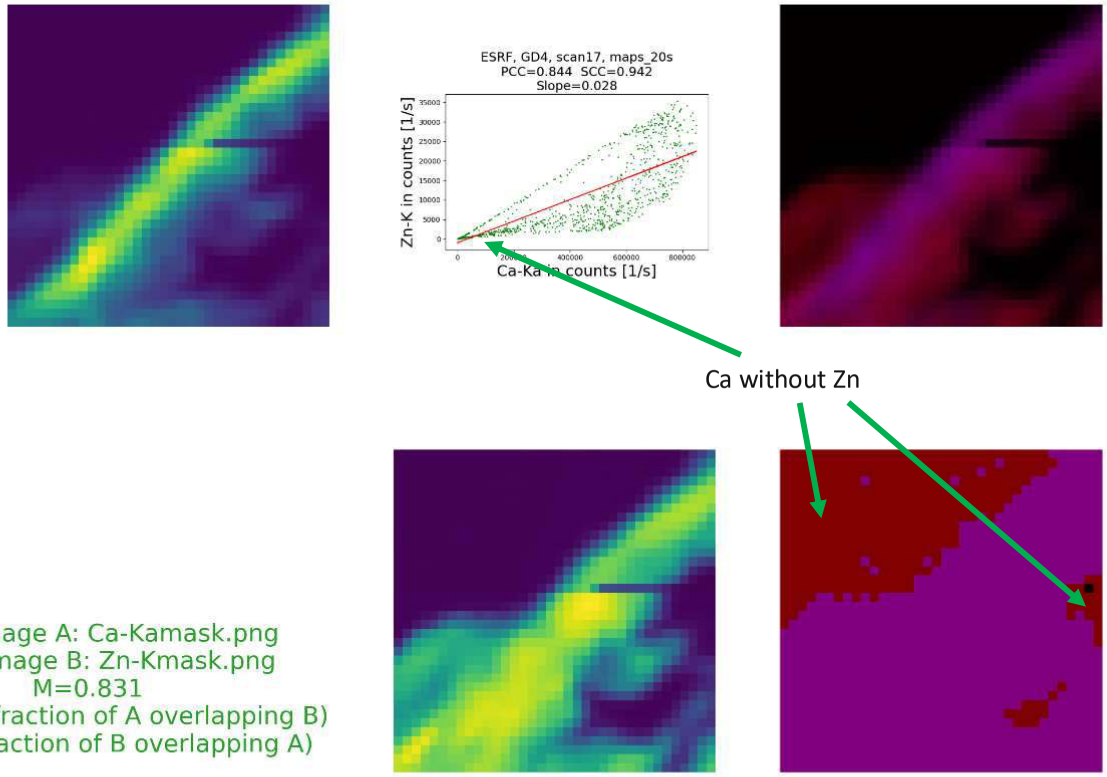
GD4



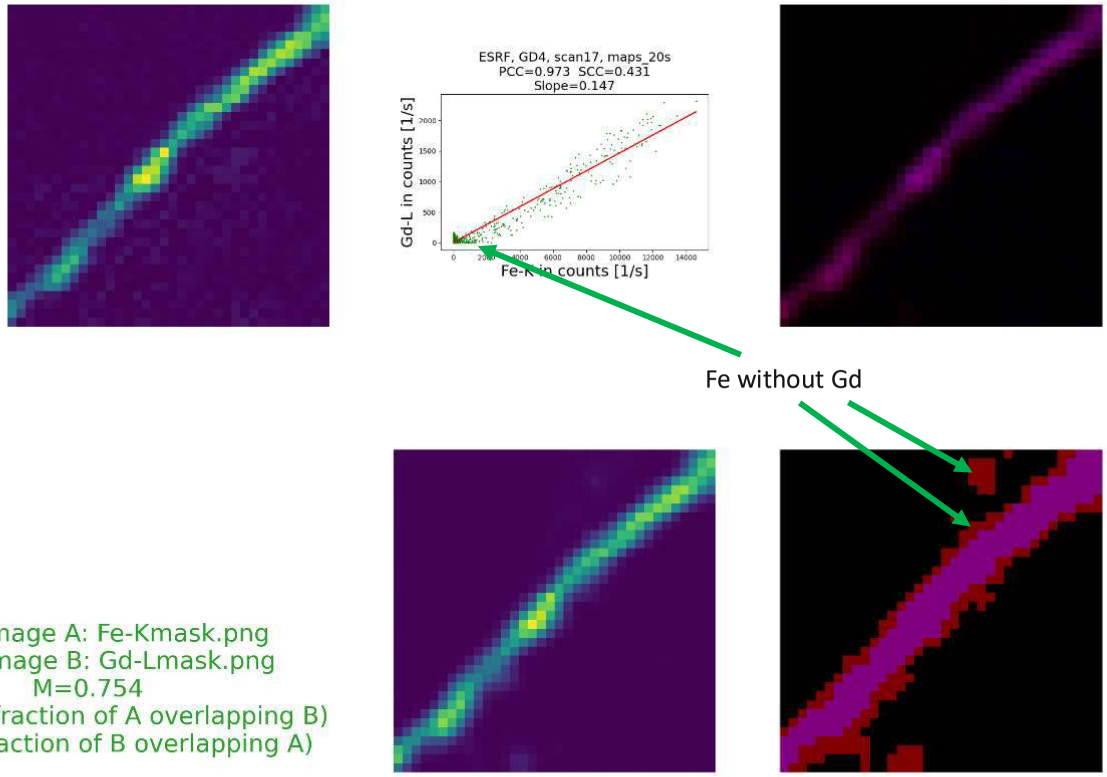


Scan 17

The non linear correlation is approx. 1. Zn overlaps Ca to 100%.



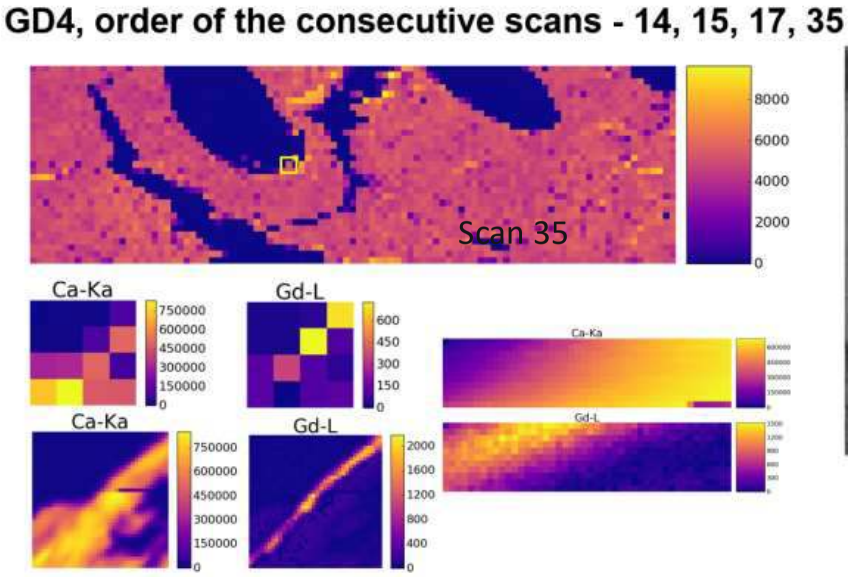
The linear correlation is approx. 1. Gd overlaps Fe to 100%.



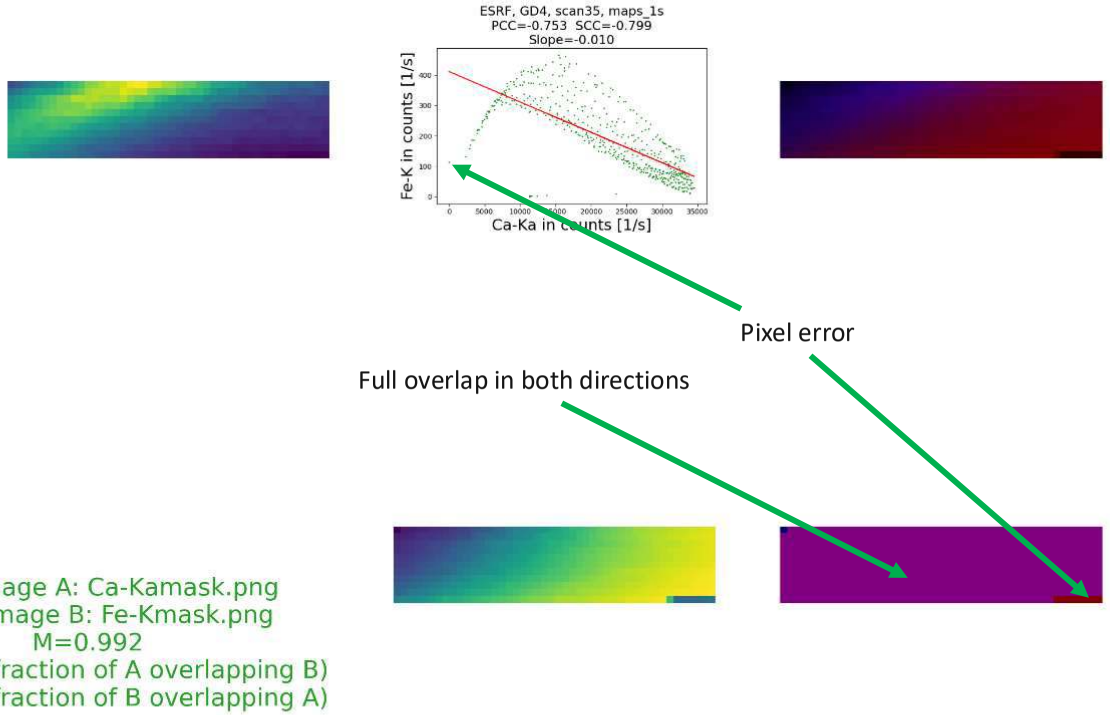
GD4

No Data for Microscopic Images

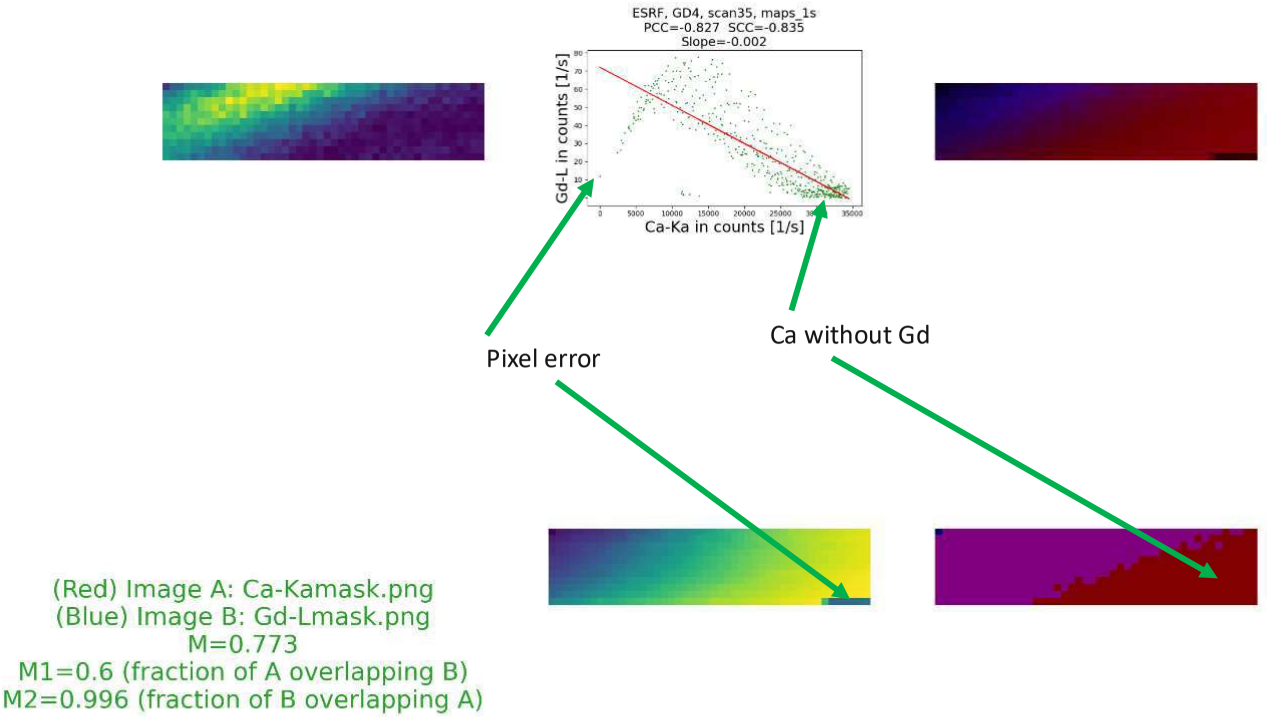
GD4 scan35



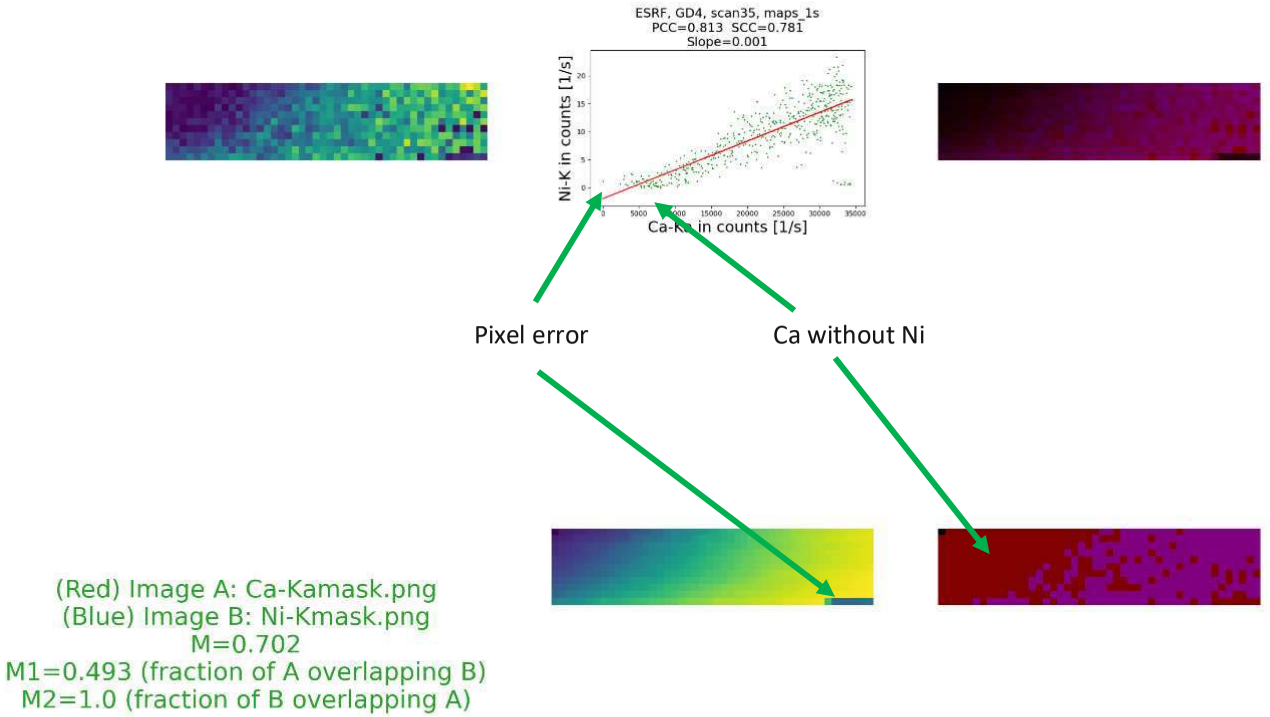
The non linear correlation is approx. -0.8. The full overlap in both directions can be shown.



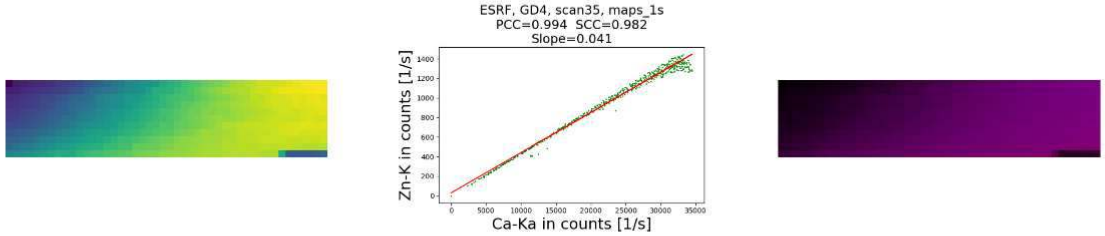
The linear correlation is approx. -0.8. Gd overlaps Ca approx. to 100%



The linear correlation is approx. 0.8. Ni overlaps Ca approx. to 100%



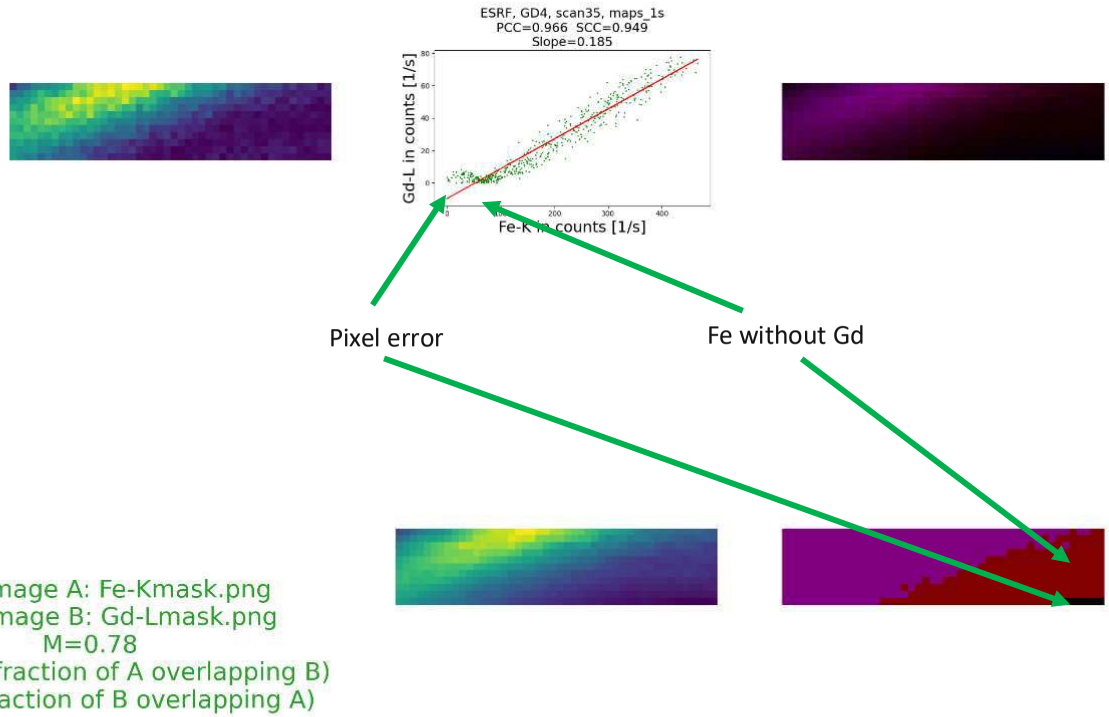
The linear correlation is approx. 1. The full overlap in both directions can be shown.



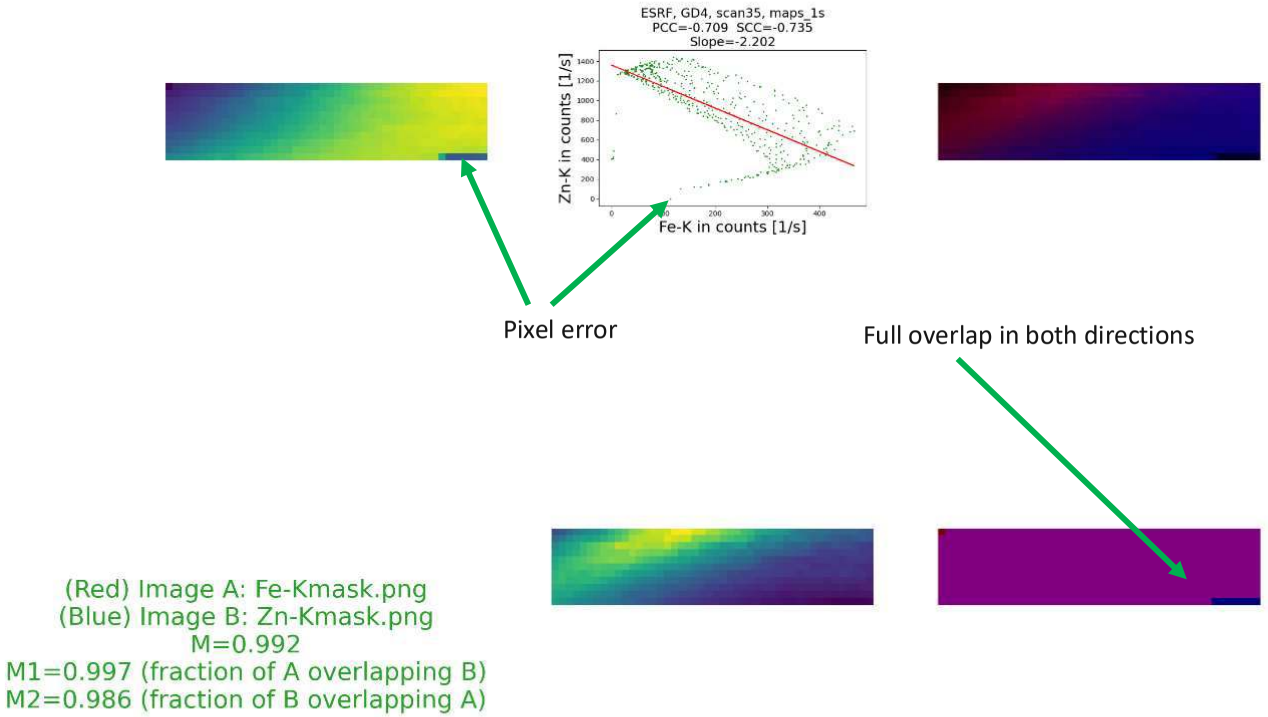
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=1.0
M1=1.0 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

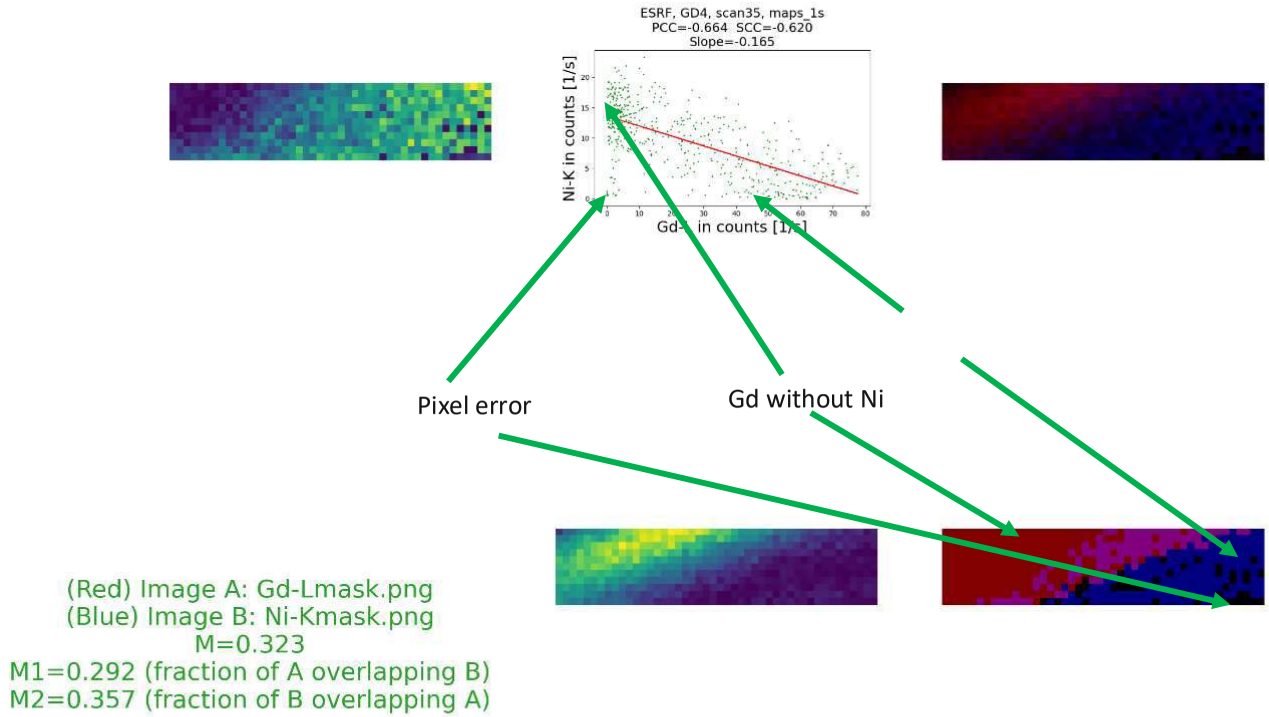
The linear correlation is approx. 1. Gd overlaps Fe to 100%.



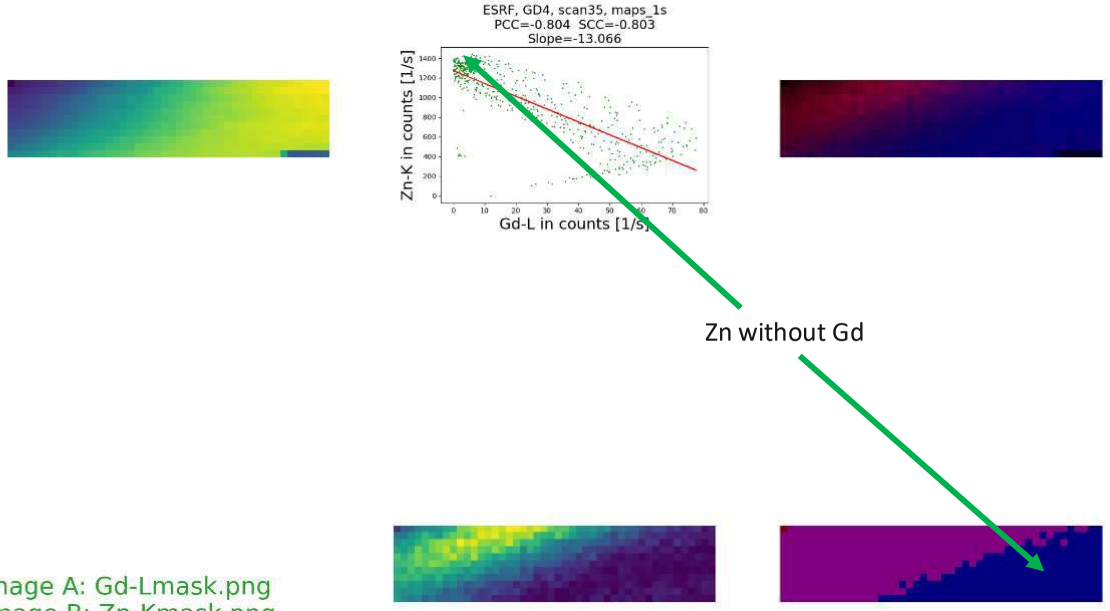
The linear correlation is approx. -0.7. The full overlap in both directions can be shown.



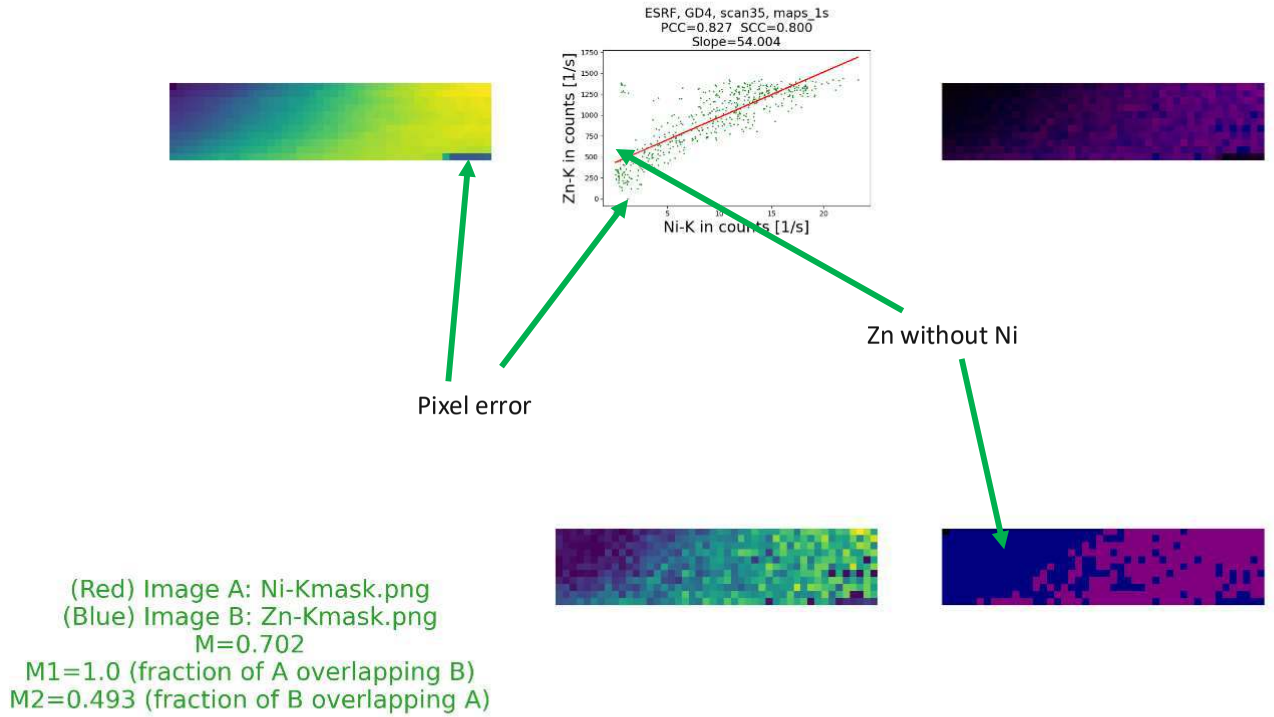
The linear correlation is approx. 1. Gd overlaps Fe to 100%.



The linear correlation is approx. -0.8. Zn without Gd can be shown. Gd overlaps Zn approx. to 100%.

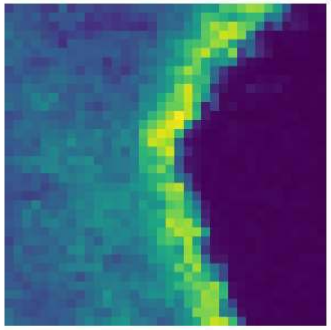


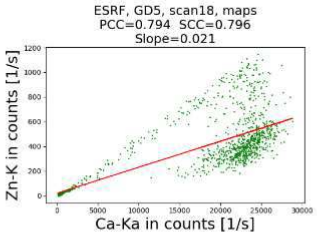
The linear correlation is approx. 0.8. Ni overlaps Zn to 100%.

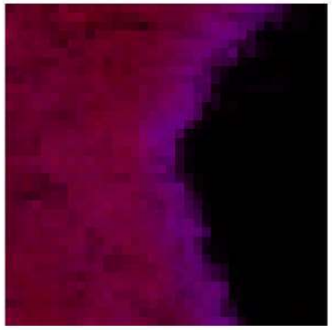


GD5

The linear correlation is approx. 0.8. Zn overlaps Ca to approx. 100%.

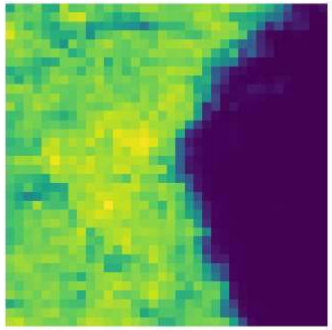


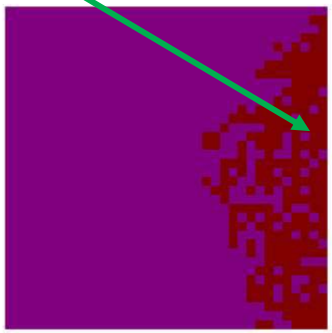




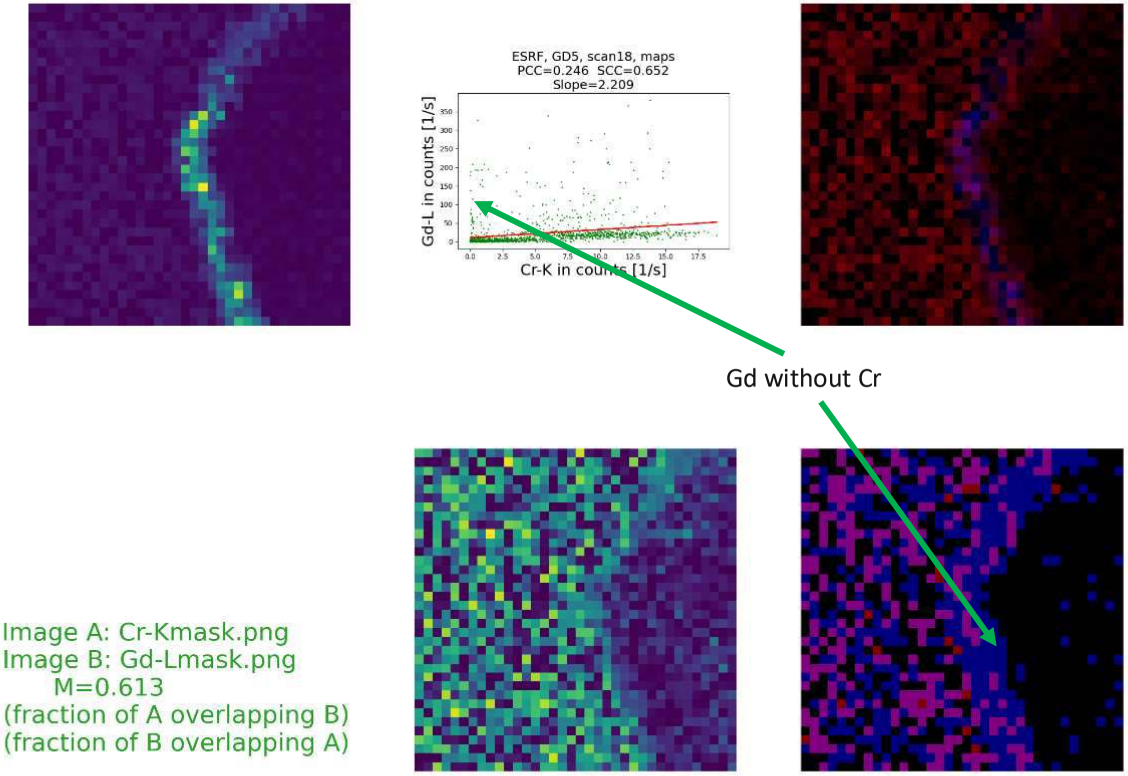
Ca without Zn

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.907
M1=0.824 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)



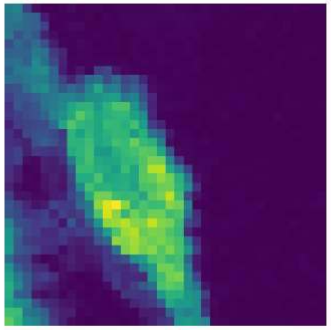


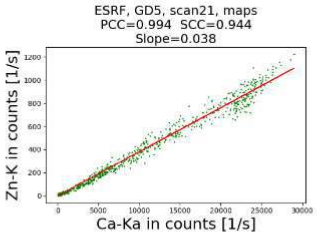
The non linear correlation is approx. 0.7. Cr overlaps Gd approx. to 90%.

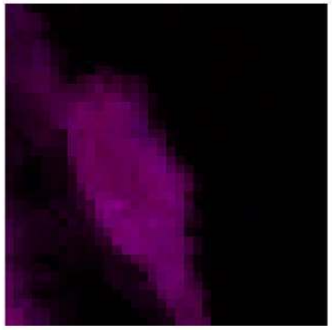


GD5

The linear correlation is approx. 1. Zn overlaps Ca to approx. 100%.

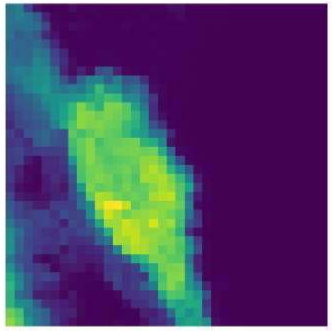


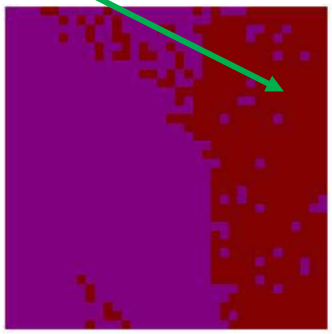




Ca without Zn

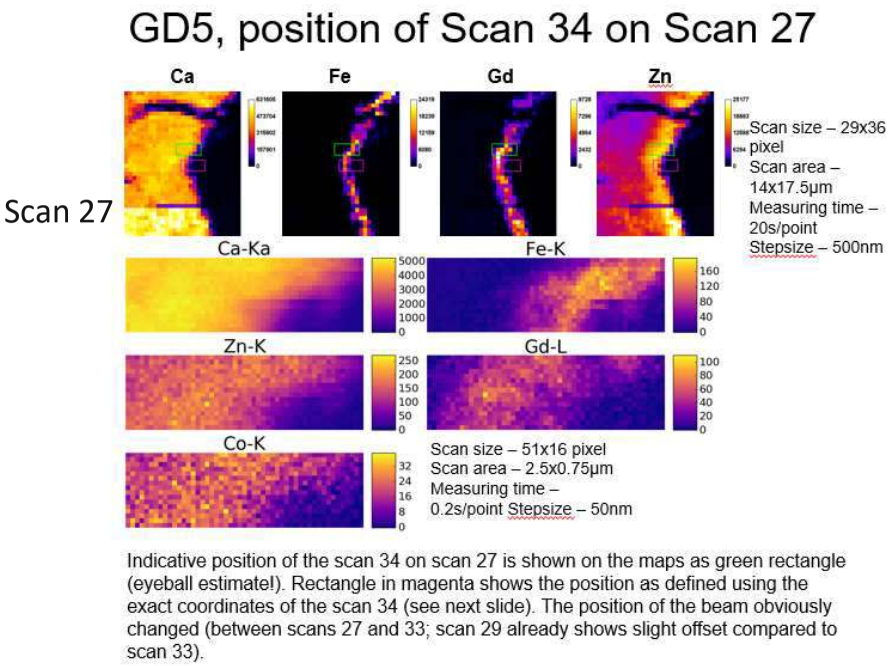
(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.787
M1=0.619 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)





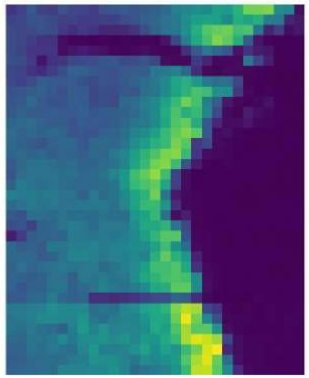
GD5

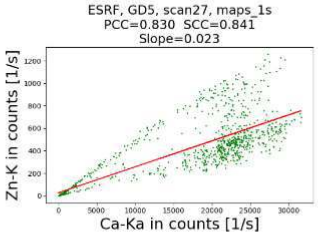
No Data for Microscopic Images

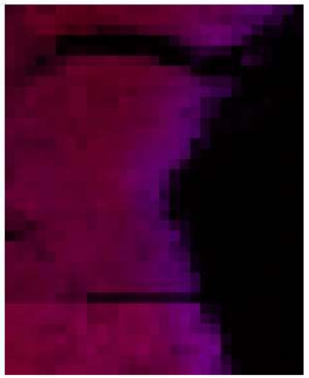


Indicative position of the scan 34 on scan 27 is shown on the maps as green rectangle (eyeball estimate!). Rectangle in magenta shows the position as defined using the exact coordinates of the scan 34 (see next slide). The position of the beam obviously changed (between scans 27 and 33; scan 29 already shows slight offset compared to scan 33).

The linear correlation is approx. 0.8. Zn overlaps Ca to 100%.

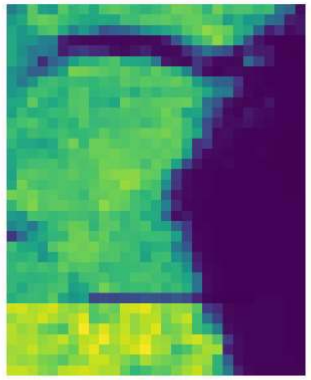


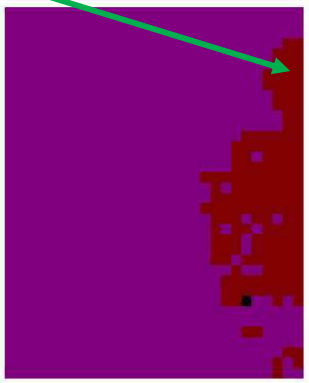




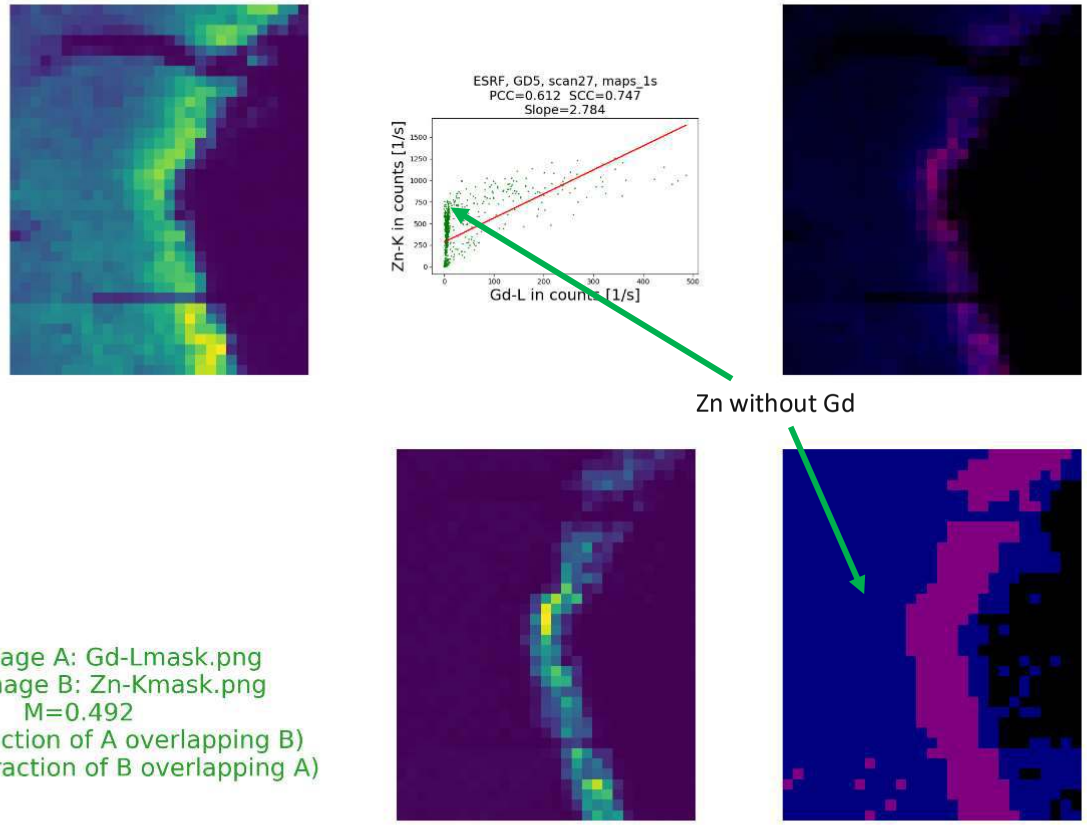
Ca without Zn

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.92
M1=0.847 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)





The non linear correlation is approx. 0.8. Zn without Gd can be shown. Gd overlaps Zn to 100%.

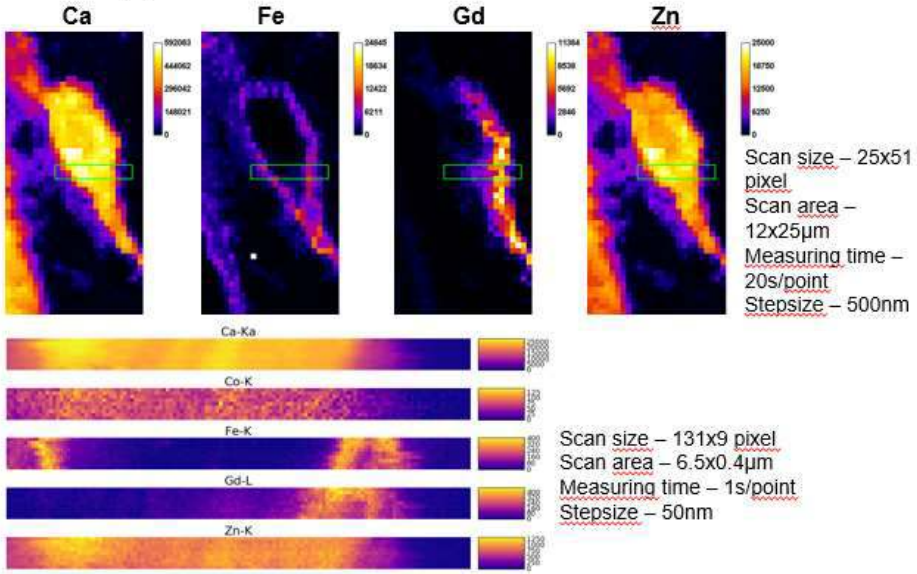


GD5

No Data for Microscopic Images

GD5, position of Scan 32 on Scan 28

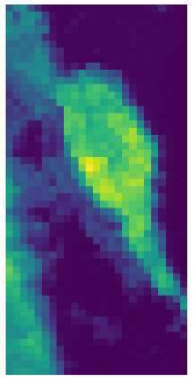
Scan 28

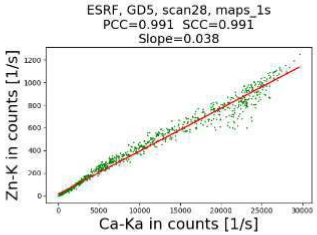


For indicative purposes only! The position of the beam have shifted between long-run scans 27/28 and following repetition of the area in scan 30. See next slide.

The non linear correlation is approx. 0.6. Gd overlaps Ca to 100%

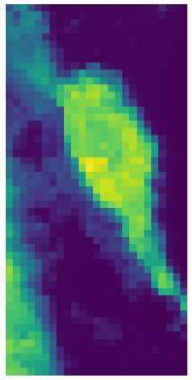
The linear correlation is approx. 1. Zn overlaps Ca to approx. 100%.

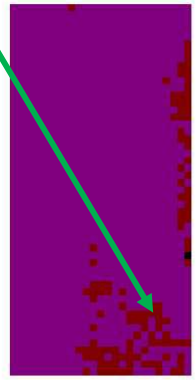




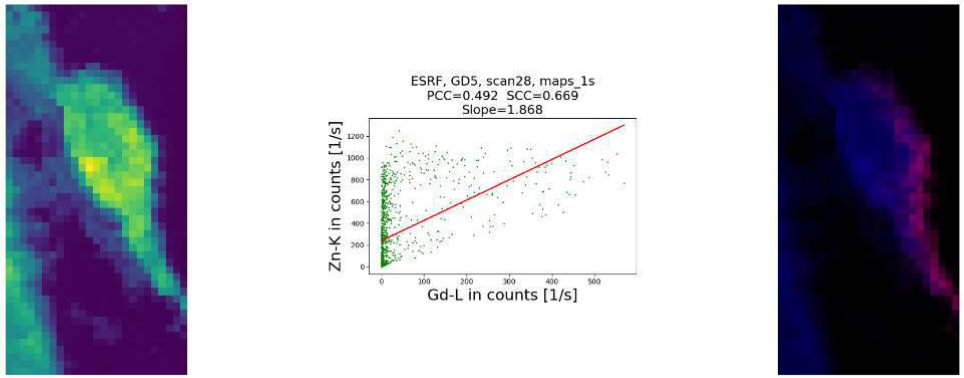
Ca without Zn

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.953
M1=0.909 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)





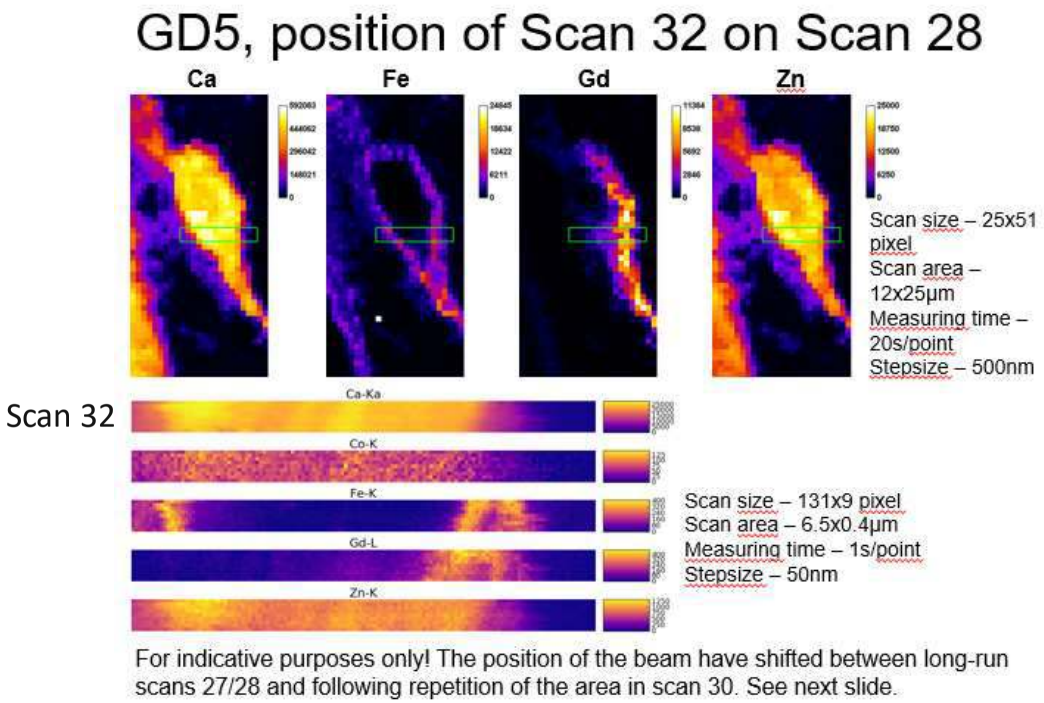
The non linear correlation is approx. 0.7. Zn without Gd can be shown. Gd overlaps Zn to 100%.



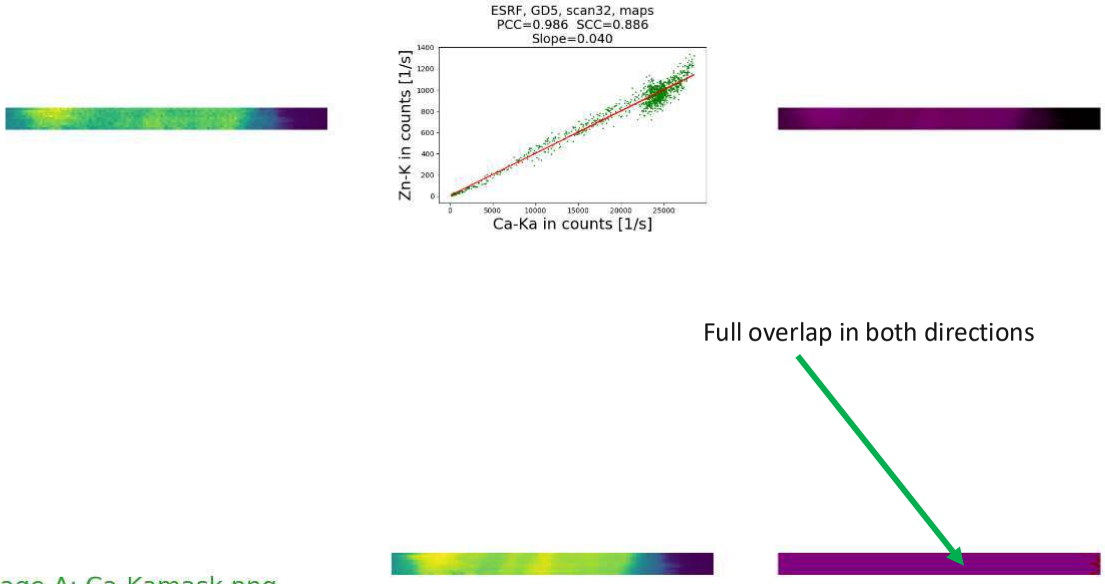
(Red) Image A: Gd-Lmask.png
(Blue) Image B: Zn-Kmask.png
M=0.586
M1=1.0 (fraction of A overlapping B)
M2=0.344 (fraction of B overlapping A)

GD5

No Data for Microscopic Images



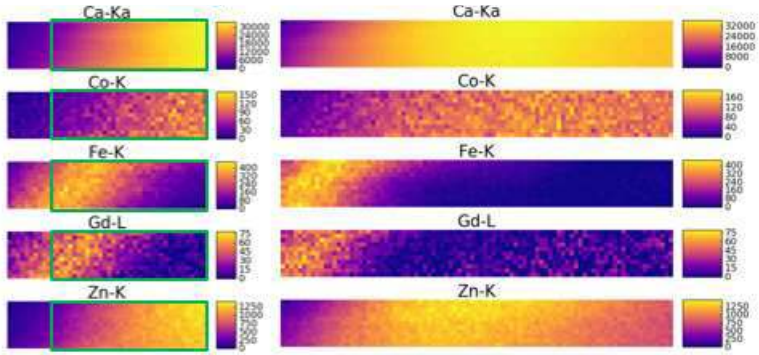
The linear correlation is approx. 1. The full overlap in both directions can be shown.



GD5

No Data for Microscopic Images

Scan 34



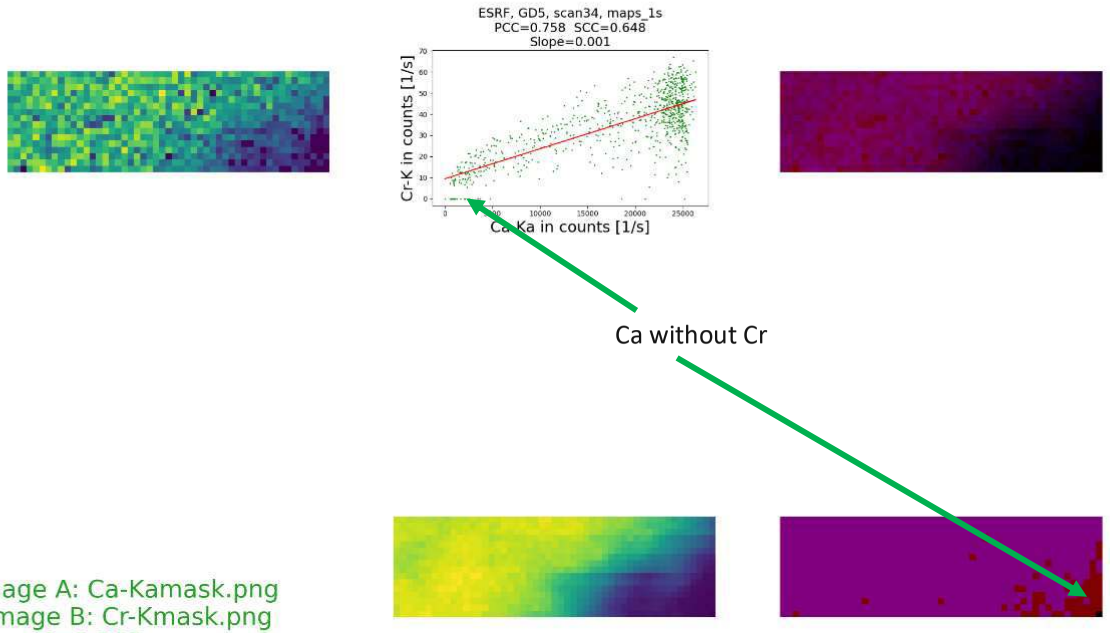
Green rectangle marks the overlap of two areas according to the motor positions

Scan size – 46x11 pixel
Scan area – 2.25x0.5µm
Measuring time – 1s/point

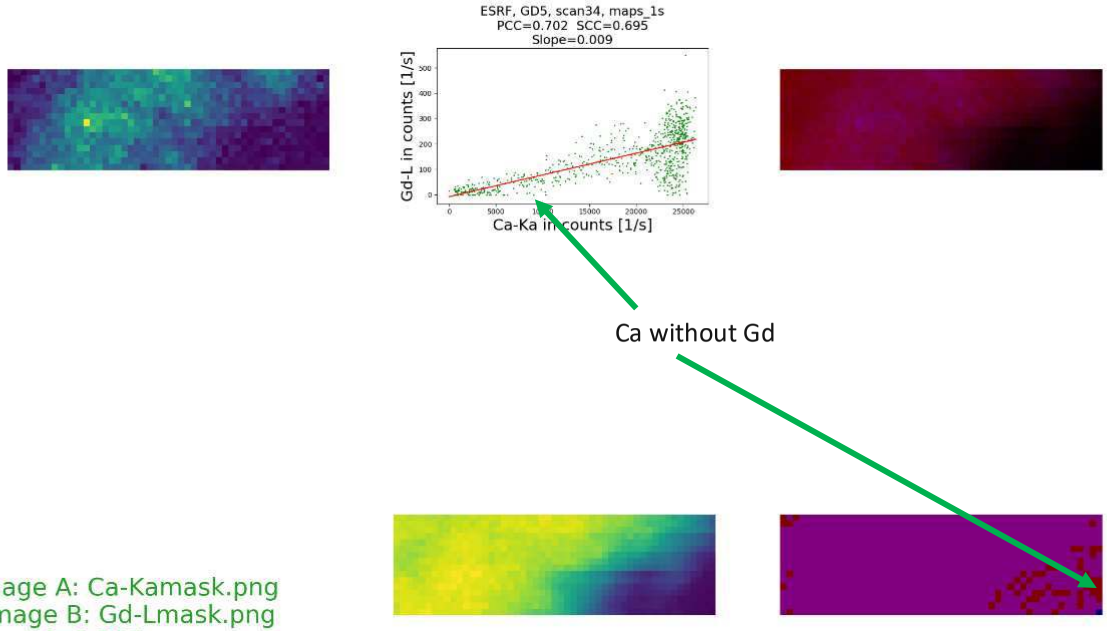
Stepsize – 50nm

Scan size – 91x11 pixel
Scan area – 4.5x0.5µm
Measuring time – 1s/point

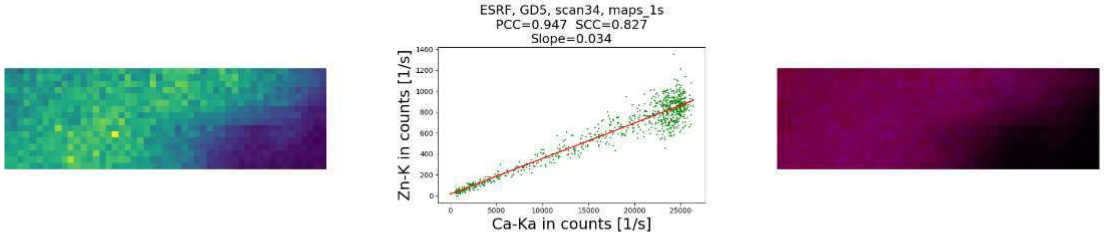
The linear correlation is approx. 0.8. Cr overlaps Ca to 100%.



The linear correlation is approx. 0.7. Gd overlaps Ca approx. to 100%



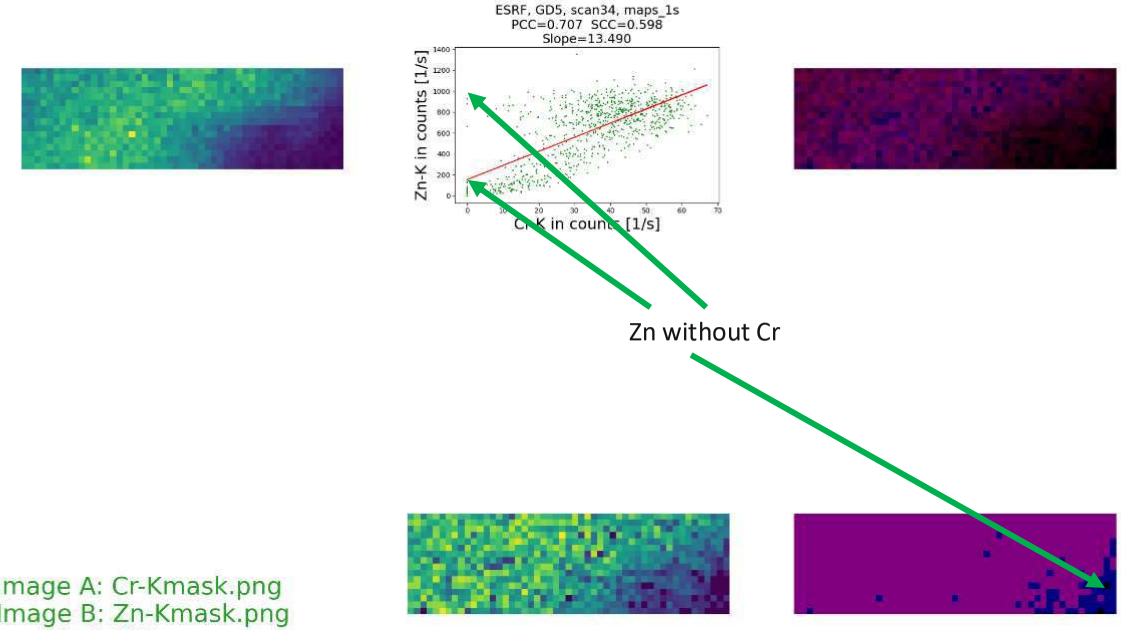
The linear correlation is approx. 1. The full overlap in both directions can be shown.



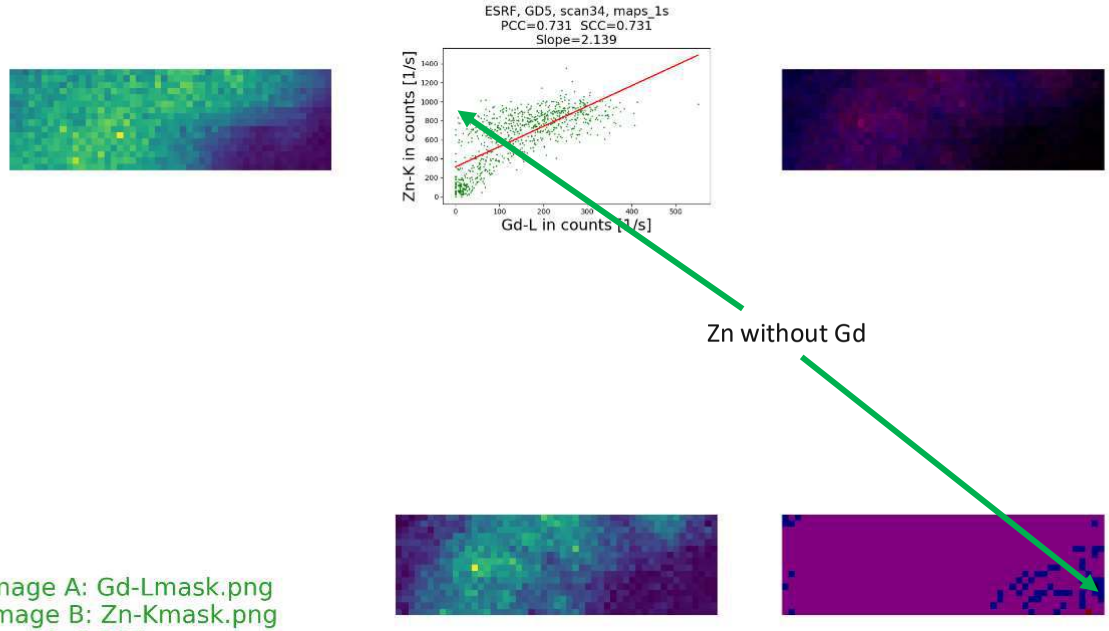
Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.998
M1=0.997 (fraction of A overlapping B)
M2=0.998 (fraction of B overlapping A)

The linear correlation is approx. 0.7. Cr overlaps Zn to 100%.



The linear correlation is approx. 0.7. Zn without Gd can be shown. Gd overlaps Zn and Zn overlaps Gd approx. to 100%.



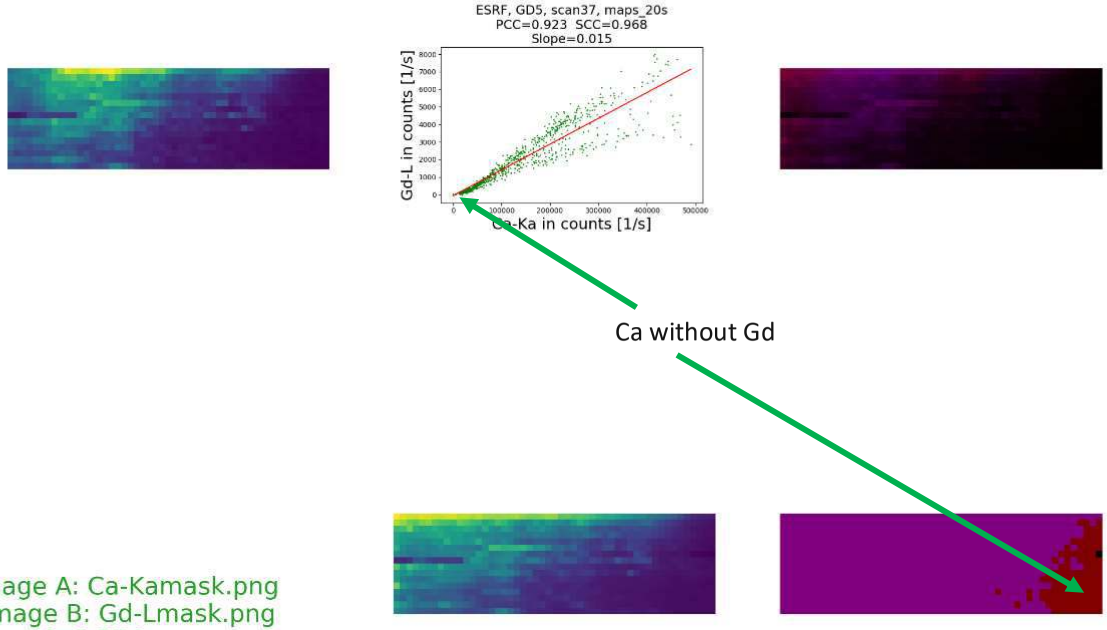
GD5

No Data for Microscopic Images

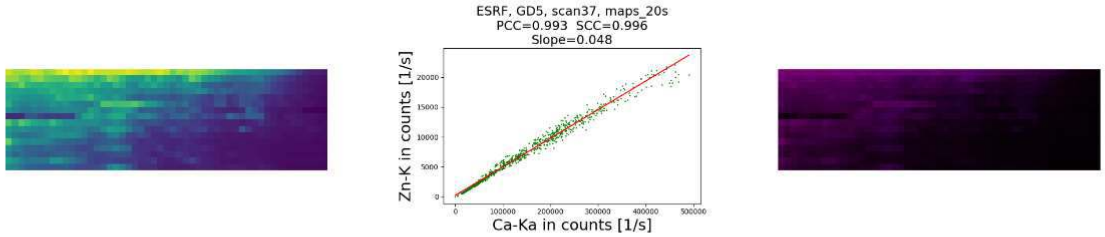
No Data for Rough Scans

GD5 scan37

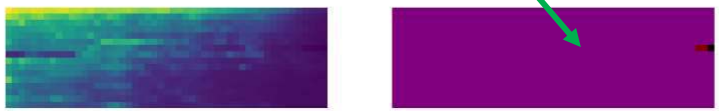
The non linear correlation is approx. 1. Gd overlaps Ca to 100%



The linear correlation is approx. 1. The full overlap in both directions can be shown.

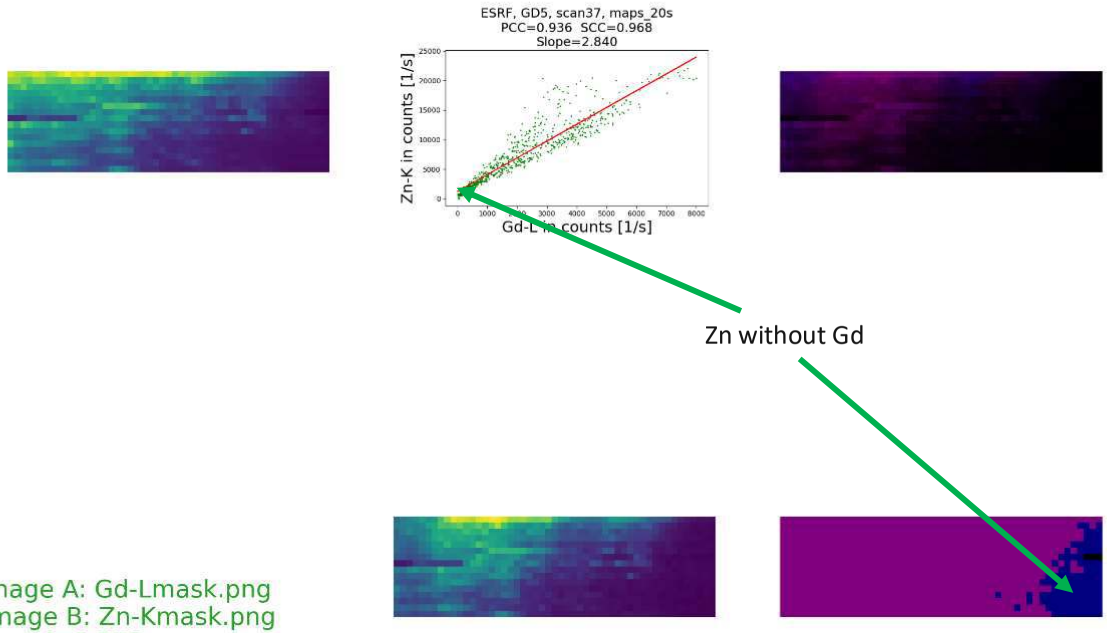


Full overlap in both directions



(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.998
M1=0.997 (fraction of A overlapping B)
M2=1.0 (fraction of B overlapping A)

The non linear correlation is approx. 1. Zn without Gd can be shown. Gd overlaps Zn to 100%.



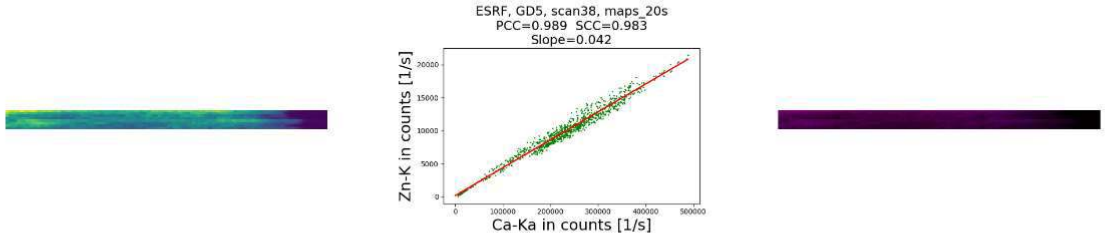
GD5

No Data for Microscopic Images

No Data for Rough Scans

GD5 scan38

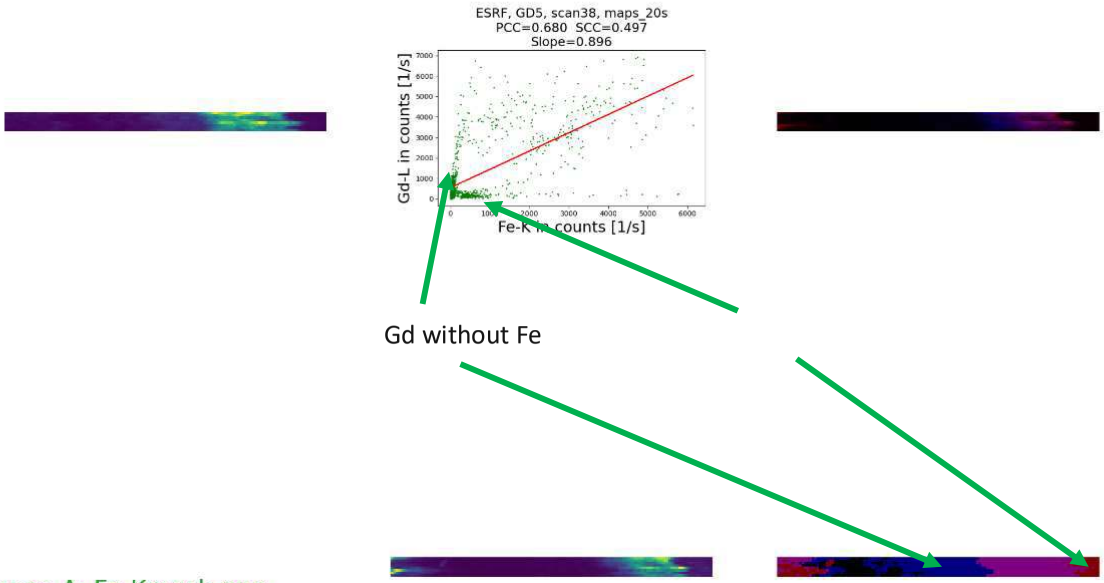
The linear correlation is approx. 1. The full overlap in both directions can be shown.



Full overlap in both directions

(Red) Image A: Ca-Kamask.png
(Blue) Image B: Zn-Kmask.png
M=0.999
M1=0.999 (fraction of A overlapping B)
M2=0.999 (fraction of B overlapping A)

The linear correlation is approx. 0.7. Fe overlaps Gd to approx. 70%.



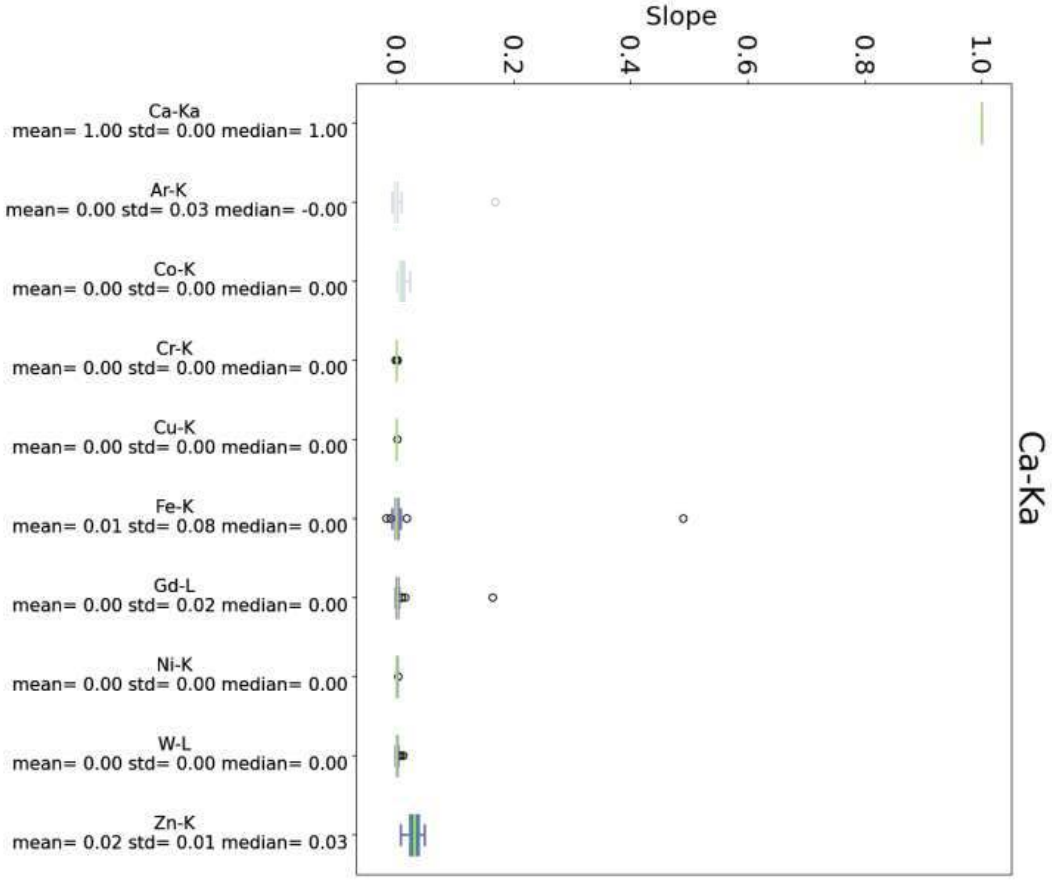
Data Interpretation

Statistic of PCC, SCC and Slope

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

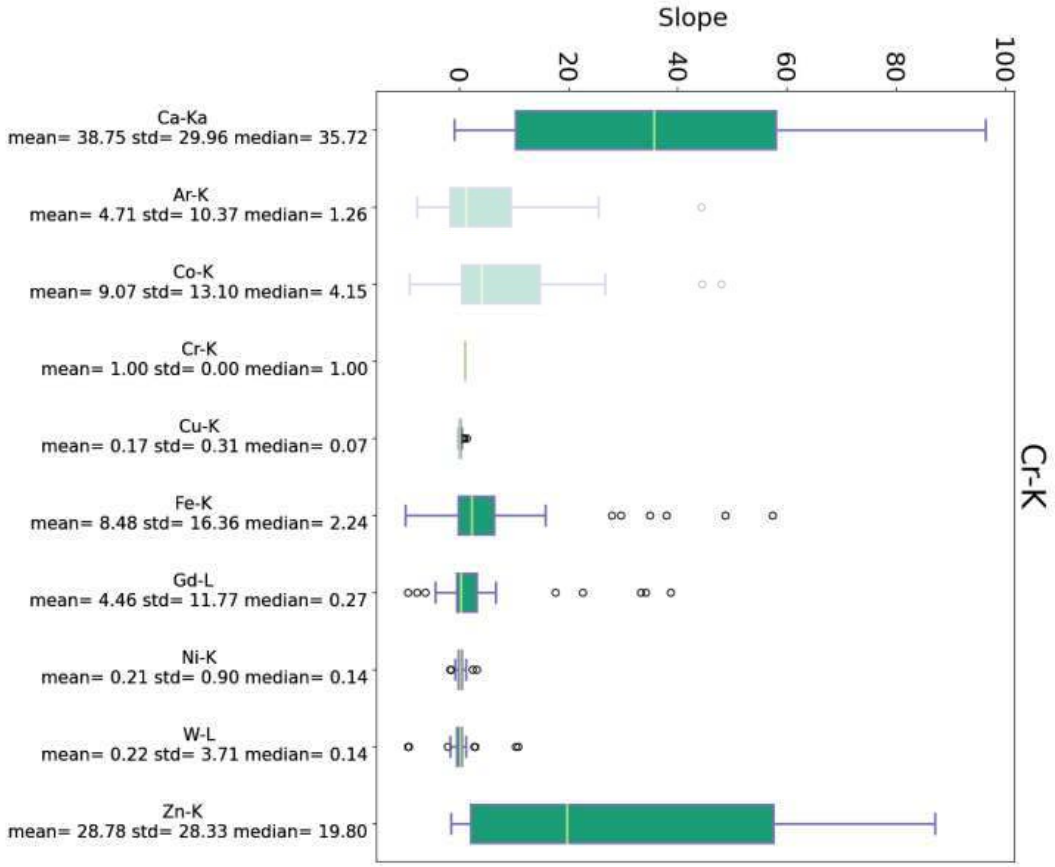
The linear correlation between Ca and Zn is approx. 0.8. Ca correlates to other elements very low and even negative. For example Fe shows high positive and high negative correlations with Ca.

The non linear correlation between Ca and Zn is approx. 0.8. Ca correlates to other elements very low. In compare to linear correlation, the deviation is a little bit higher. For example Fe shows high positive and high negative correlations with Ca.

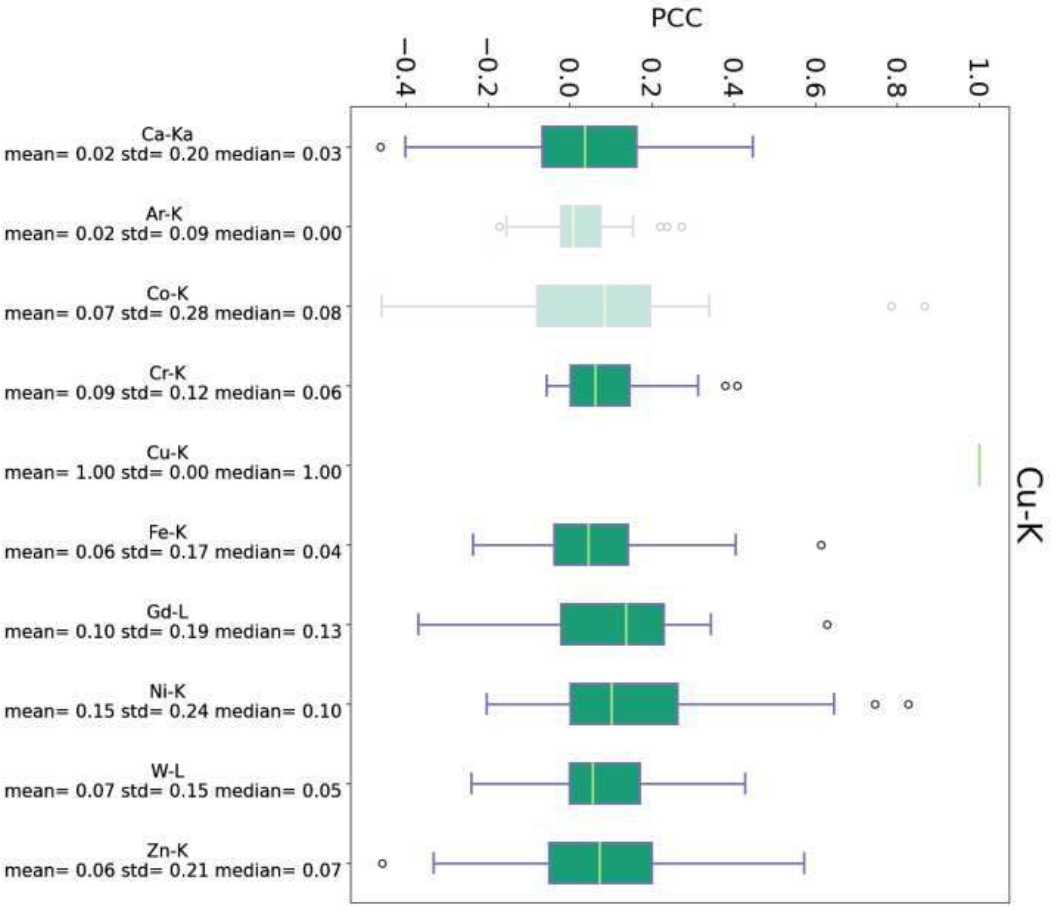


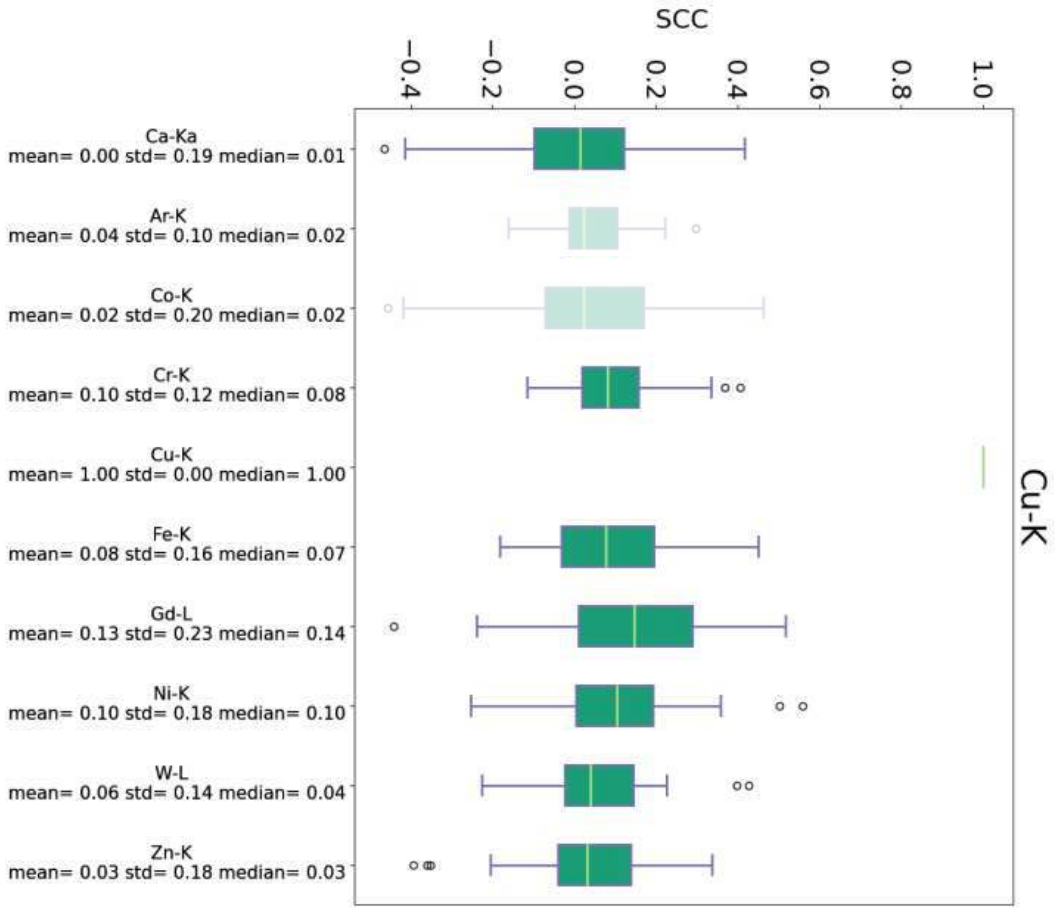
The linear correlation between Cr and other elements is very low and the deviation is very high and even negative.

The non linear correlation between Cr and other elements is very low and the deviation is very high and even negative. No special differences to the linear correlation.

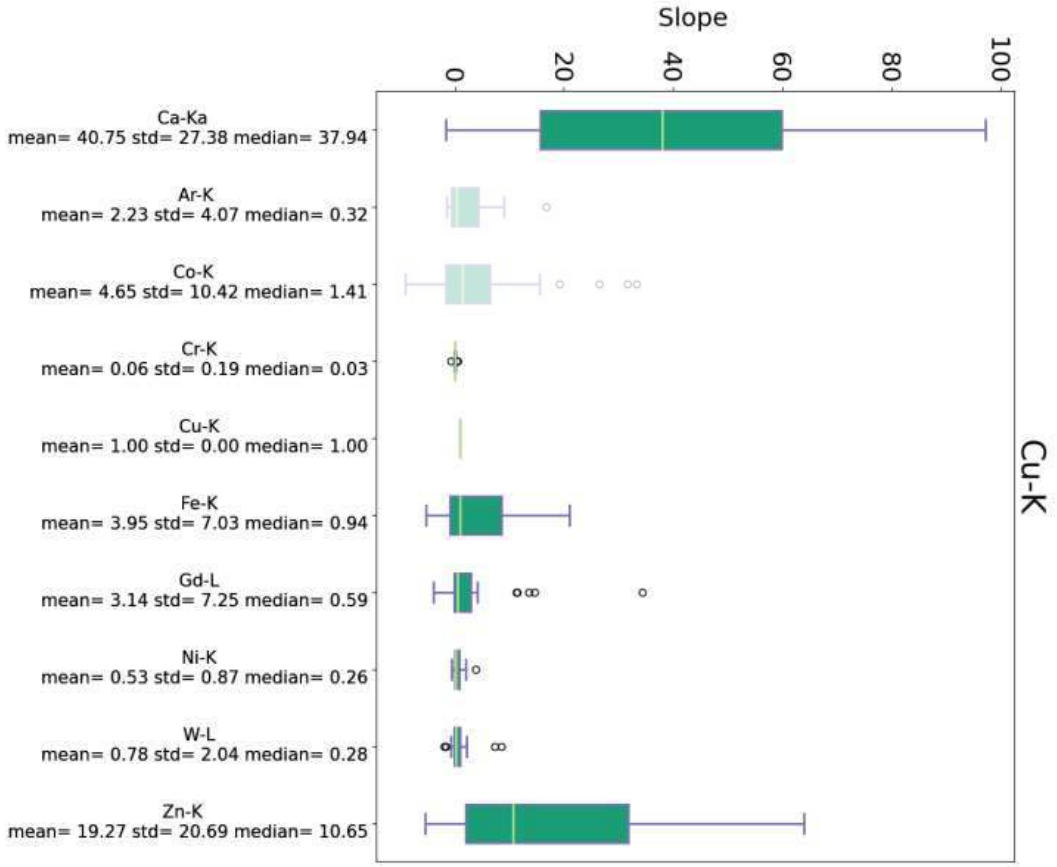


The linear correlation between Cu and other elements is very low and the deviation is very high and even negative.



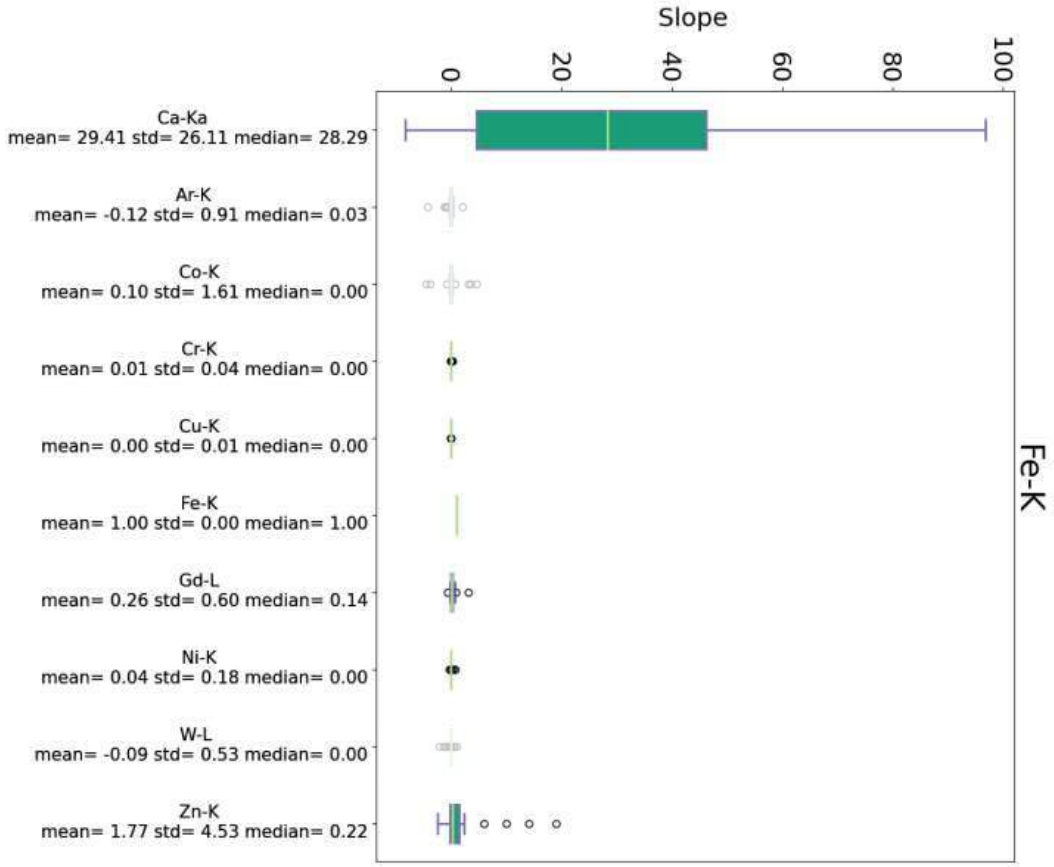


The non linear correlation between Cu and other elements is very low and the deviation is very high and even negative. No special differences to the linear correlation.



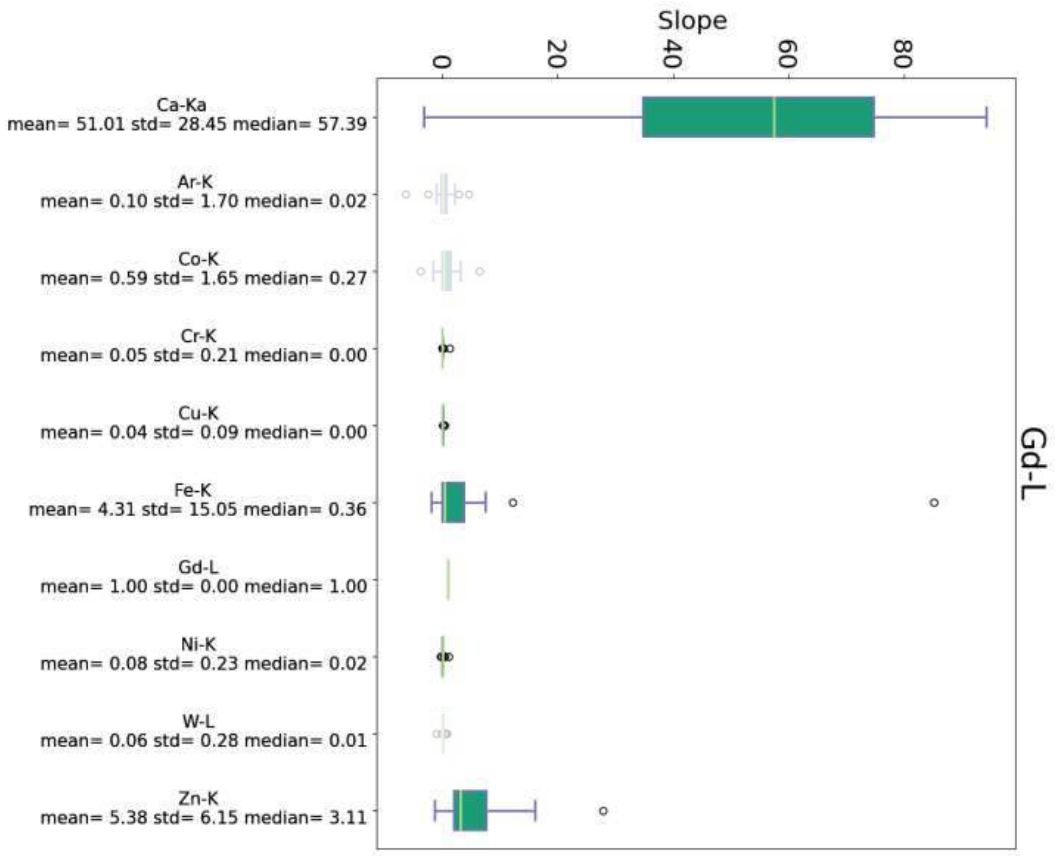
The linear correlation between Fe and other elements is very low and the deviation is very high and even negative. Gd shows a small deviance with a higher correlation and a higher deviation more on the positive side.

The non linear correlation between Fe and other elements is very low and the deviation is very high and even negative. Gd shows a small deviance with a higher correlation and a higher deviation more on the positive side. No special differences to the linear correlation.



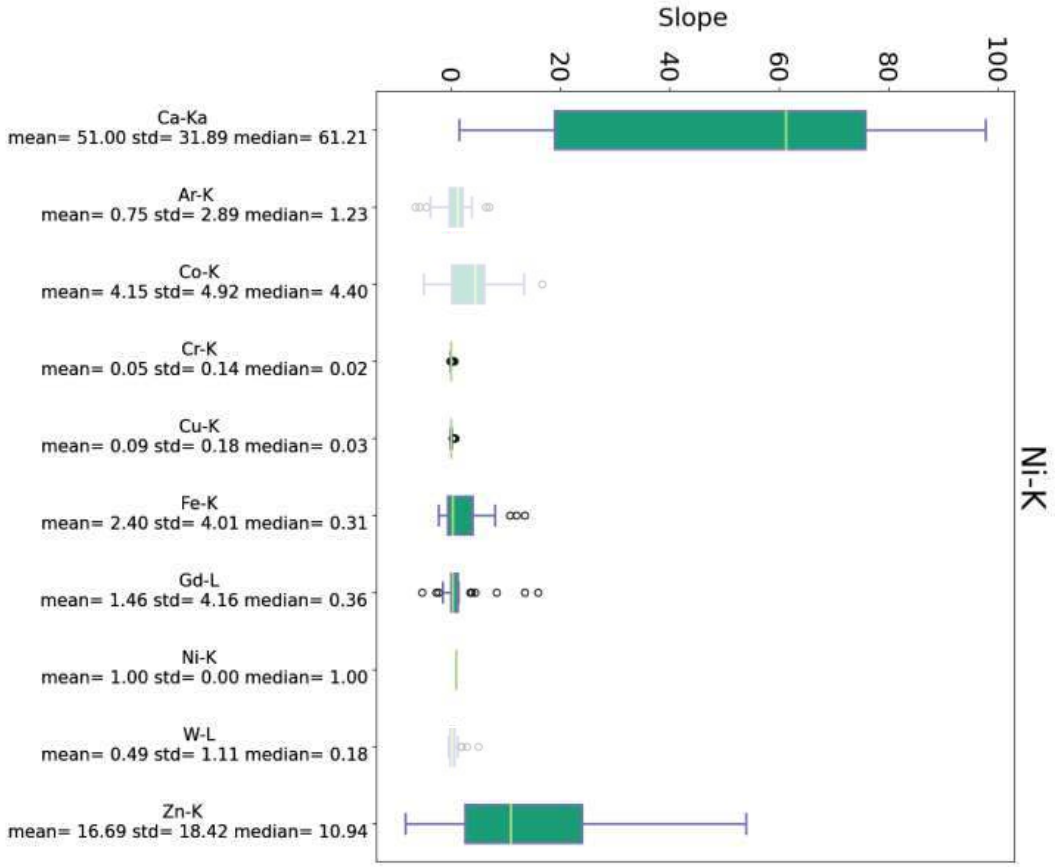
The linear correlation between Gd and other elements is very low and the deviation is very high and even negative, but more on the positive side. Ca, Fe and Zn show a higher correlation and reach higher values than Cr, Cu and Ni.

The linear correlation between Gd and other elements is very low and the deviation is very high and even negative, but more on the positive side. Ca, Fe and Zn show a higher correlation and reach higher values than Cr, Cu and Ni.



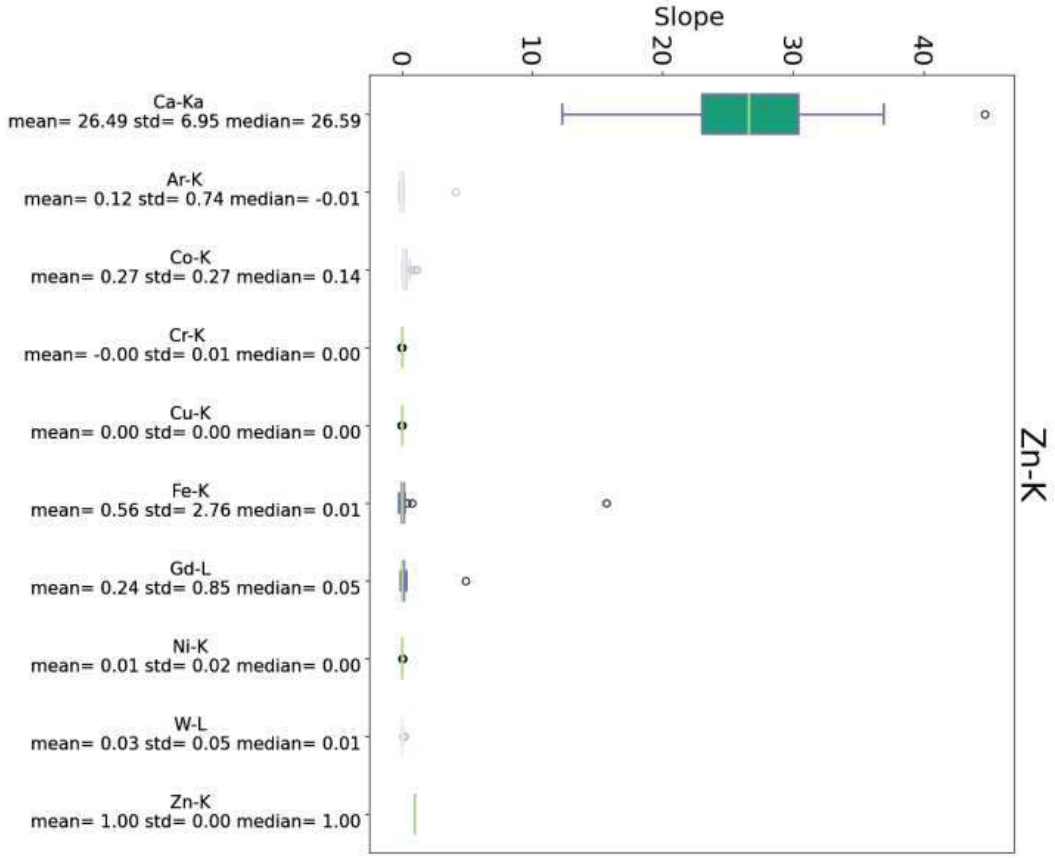
The linear correlation between Ni and other elements is very low and the deviation is very high and even negative, but more on the positive side. Ca and Zn show a higher correlation and reach higher values than Cr, Cu, Fe and Gd.

The non linear correlation between Ni and other elements is very low and the deviation is very high and even negative, but more on the positive side. Ca and Zn show a higher correlation and reach higher values than Cr, Cu, Fe and Gd.



The linear correlation between Zn and Ca is very high and is approx. 0.8. The correlation between Zn and Gd is approx. 0.4 but the deviation is high and also negative. For other elements, the correlation is very low and the deviations is very high and even negative.

The non linear correlation between Zn and Ca is very high and is approx. 0.9. The correlation between Zn and Gd is approx. 0.4 but the deviation is high and also negative. For other elements, the correlation is very low and the deviations is very high and even negative. No special differences to the linear correlation.



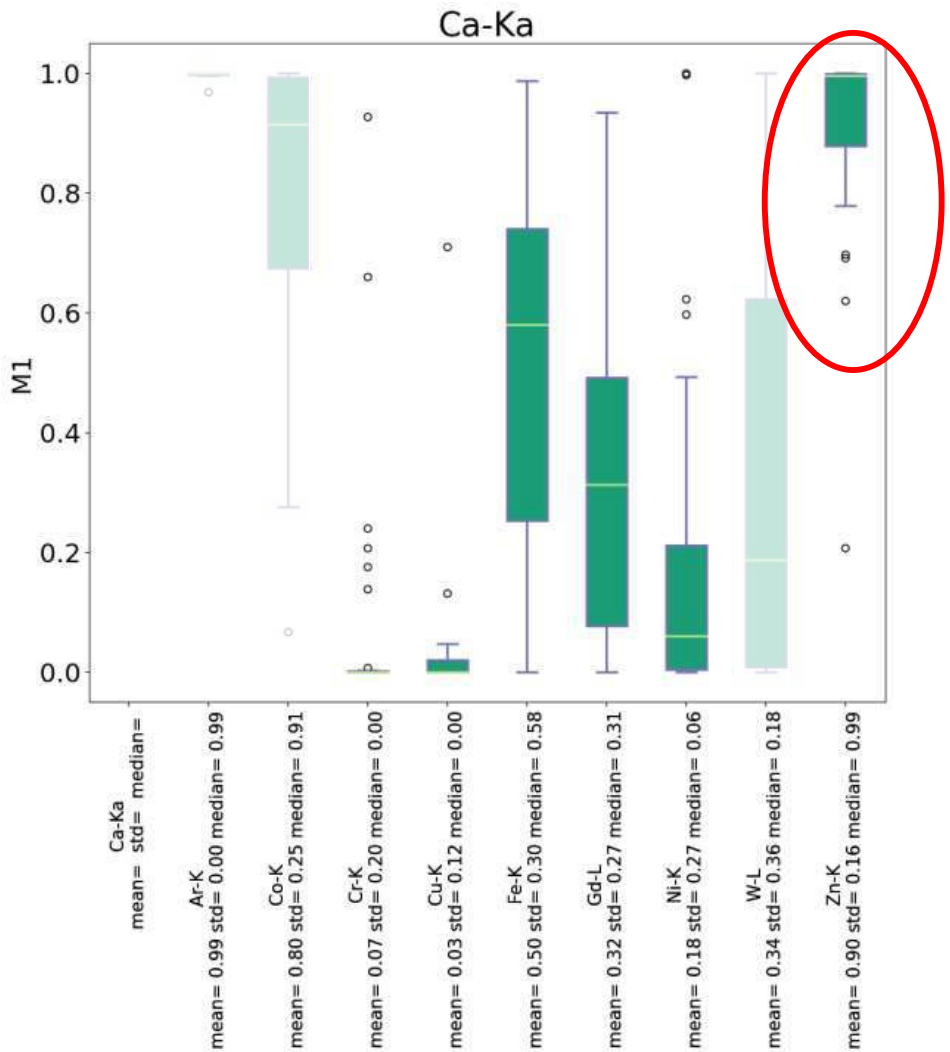
Data Interpretation

Statistic of MOC

M1: head element overlaps x-Axis

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

Ca overlaps Zn in the most areas. But it can be shown, Ca has only a small number of images where it is full covered with other elements. Ca overlaps Cr and Cu only in small areas. Ca overlaps Fe and Gd in big areas but the deviation is also high. Ca overlaps Ni in small areas and the deviation is high.



Cr does not overlap other elements. The average value for all elements is 0. The high deviation maybe can be traced back to noise.

Cu does not overlap other elements. The average value for all elements is 0. The high deviation maybe can be traced back to noise.

Fe overlaps Ca and Zn in the most areas. Fe overlaps Gd approx. to 40% but the high deviation must be noted. For Cr, Cu and Ni the overlap is very low and for Ni the deviation is also very high.

Ni overlaps Ca and Zn in the most areas, especially Ca. Ni overlaps Fe approx. to 50% but the high deviation must be noted. Ni overlaps Gd approx. to 30% and the high deviation must be also noted. For Cr and Cu the overlap is very low.

Zn overlaps Ca in the most areas. Zn overlaps Fe approx. to 60% but the high deviation must be noted. Zn overlaps Gd approx. to 30% and the high deviation must be also noted. Zn overlaps Ni very low and the deviation is high. For Cr and Cu the overlap is very low.

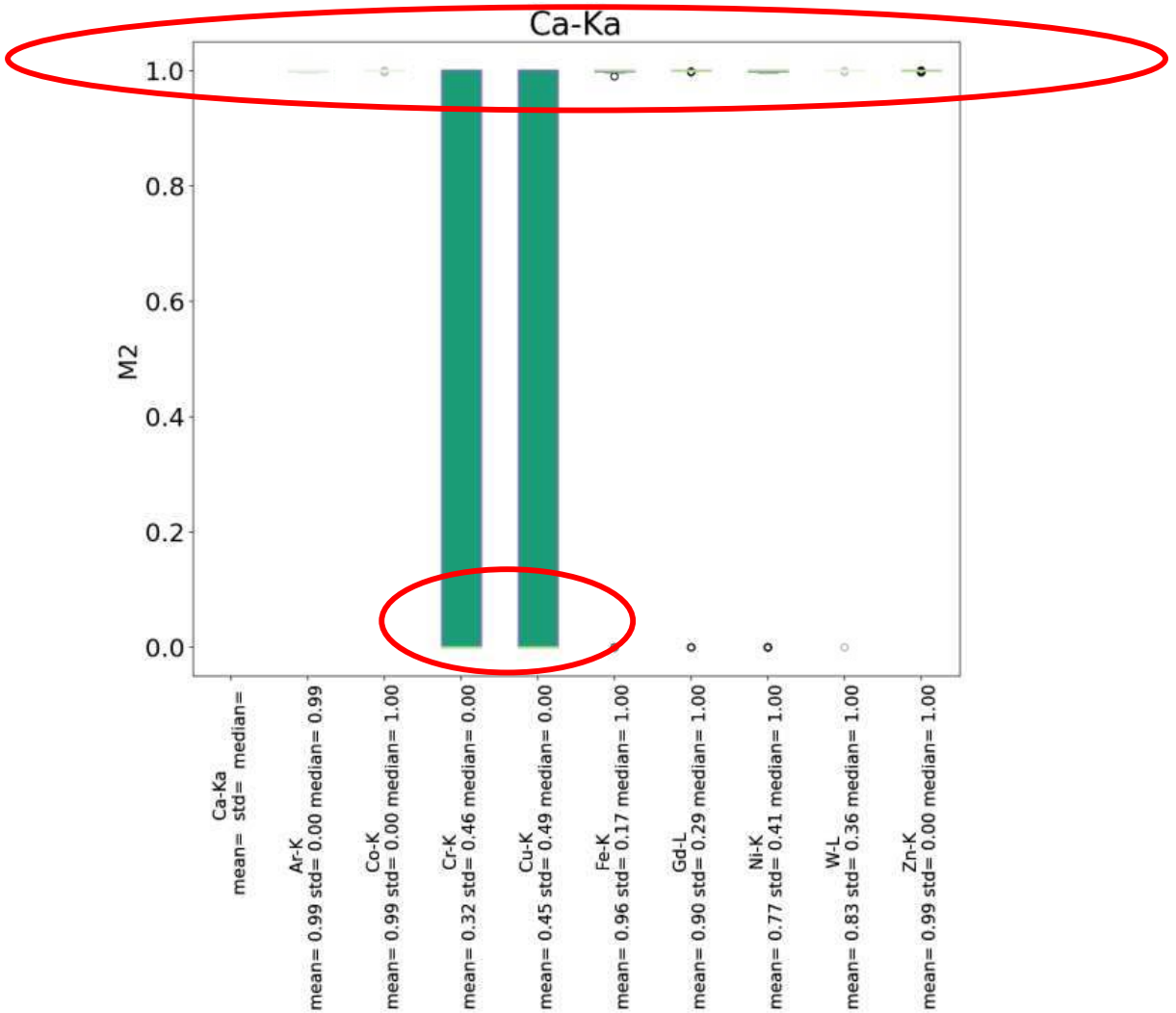
Data Interpretation

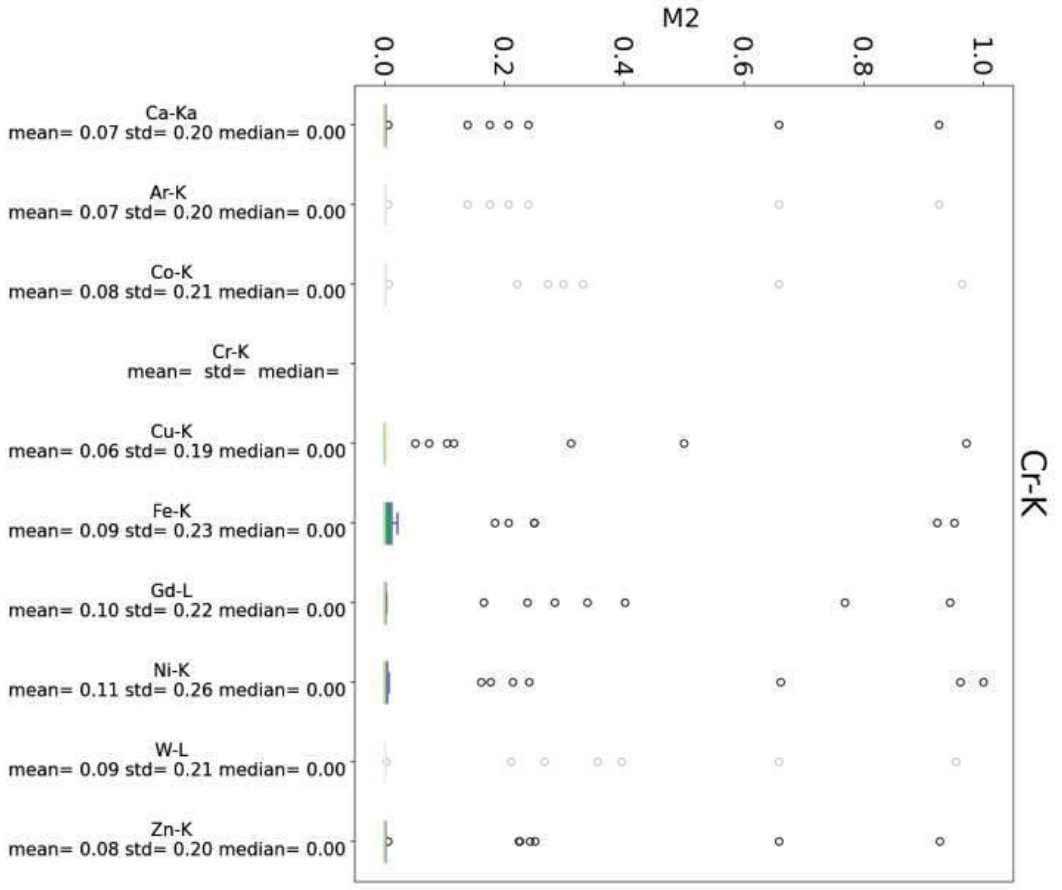
Statistic of MOC

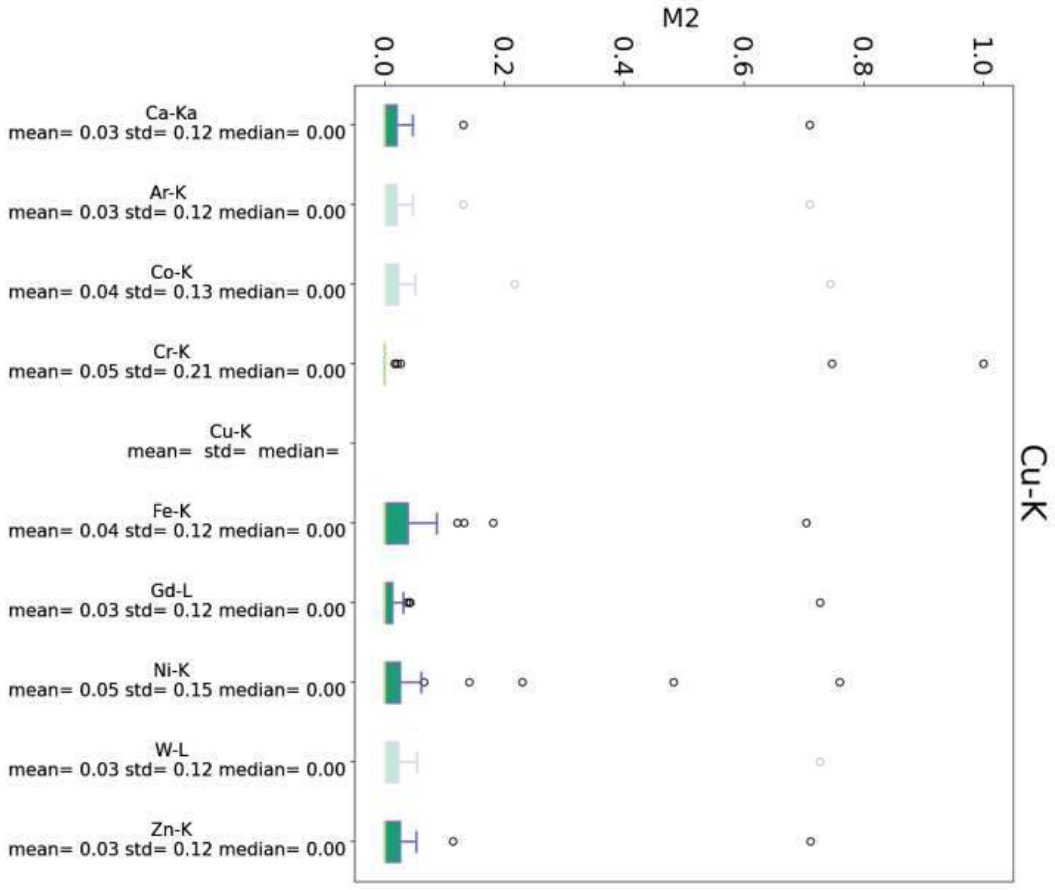
M2: x-Axis overlaps the head element

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

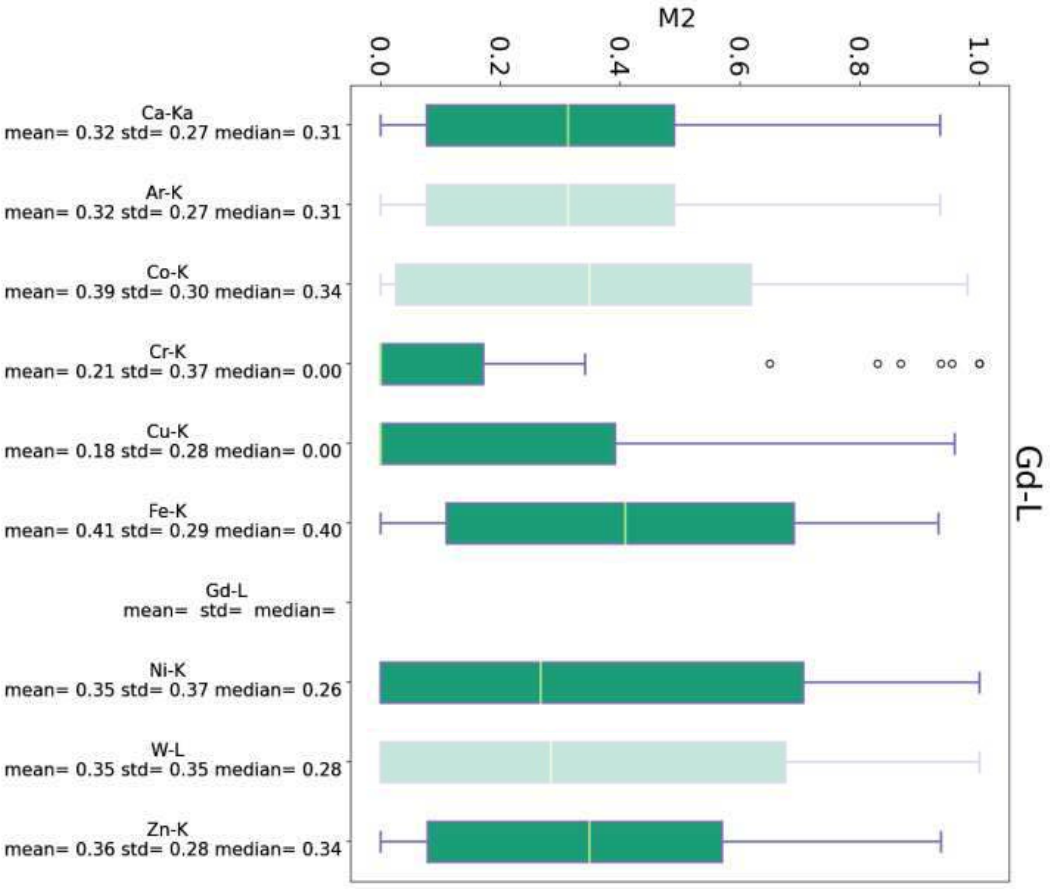
Fe, Gd, Ni and Zn overlap Ca to nearly 100%. Cr and Co do not overlap Ca. The high deviation maybe is traced back to noise.







Gd overlaps Fe approx. To 70%, but the deviation is very high. Ca, Ni and Zn overlap Fe approx. 60 % and the deviation is also high. Cr and Cu do not overlap Fe in the average value.



Ca, Fe, Ni and Zn overlap Gd approx. to 30 % and the deviation is very high. Cr and Cu do not overlap Gd in the average value.

Ca, Fe, Gd and Zn overlap Ni approx. to 10 % and the deviation is very high. Cr and Cu do not overlap Ni in the average value.

Ca, Fe, Gd and Ni overlap Zn approx. to 100 % and the deviation is low. Cr and Cu do not overlap Zn in the average value.

Data Interpretation

Statistic of MOC

M: M1 and M2 combined

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

The overlap between Ca and Zn is very high. The overlap between Ca and Fe is approx. 80% but the deviation is very high. The overlap between Ca and Gd is approx. 50% and the deviation is also high. The deviation between Ca and Ni is low and the deviation is also high. The overlap between Ca and Cr and Cu is nearly 0%.

The overlap between Cr and all other elements is nearly 0%.

The overlap between Cu and all other elements is nearly 0%.

The overlap between Fe and Ca and Zn is approx. 80% and the deviation is very high. The overlap between Fe and Gd is approx. 50% but the deviation is also very high. The overlap between Fe and Ni is approx. 20%. The overlap between Fe and Cr and Cu is nearly 0%.

The overlap between Gd and Ca, Fe and Zn is very high and the deviation is also very high. The overlap between Gd and Ni is approx. 10% but the deviation is very high. The overlap between Gd and Cr and Cu is nearly 0%.

The overlap between Ni and Ca, Fe, Gd and Zn is approx. 30% and the deviation is high. The overlap between Ni and Cr and Cu is nearly 0%.

The overlap between Zn and Ca is approx. 100% and the deviation is low. The overlap between Zn and Fe is approx. 70% but the deviation is very high. The overlap between Zn and Gd is approx. 60% and the deviation is also high. The deviation between Zn and Ni is approx. 30% and the deviation is also high. The overlap between Zn and Cr and Cu is nearly 0%.

Data Interpretation

Statistic
PCC, SCC and Slope

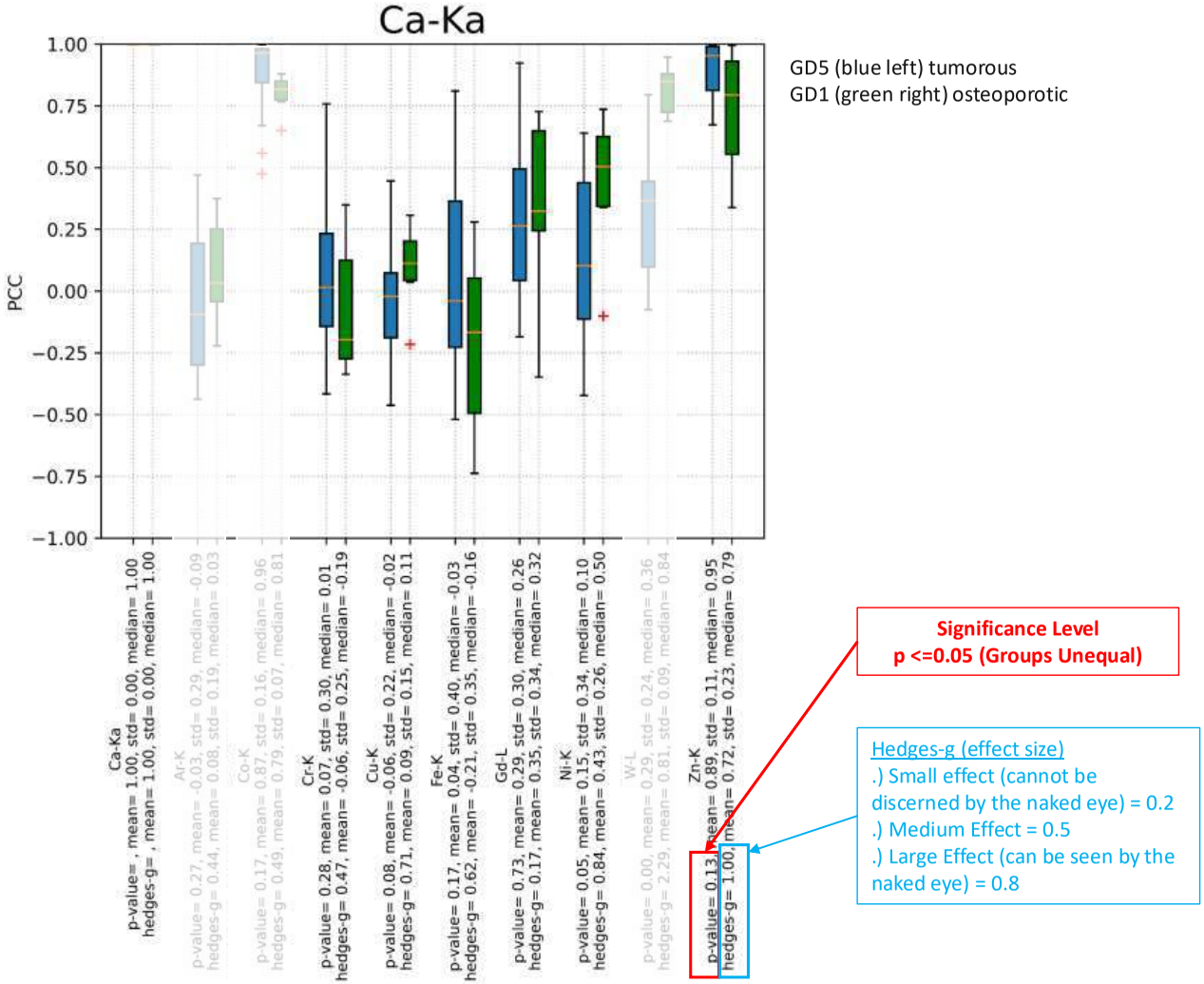
Compare
GD1 (n=7) & GD5 (n=15)

Significance Level
p <=0.05 (Groups Unequal)

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

Ni shows a significance difference (p-value= 0.05).

For all elements, no significant differences can be shown.



GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Significance Level
p <=0.05 (Groups Unequal)

Hedges-g (effect size)
.) Small effect (cannot be discerned by the naked eye) = 0.2
.) Medium Effect = 0.5
.) Large Effect (can be seen by the naked eye) = 0.8

Source: https://www.statisticshowto.com/hedges-g/

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ni shows a significance difference (p-value= 0.01).

For all other elements, no significant differences can be shown.

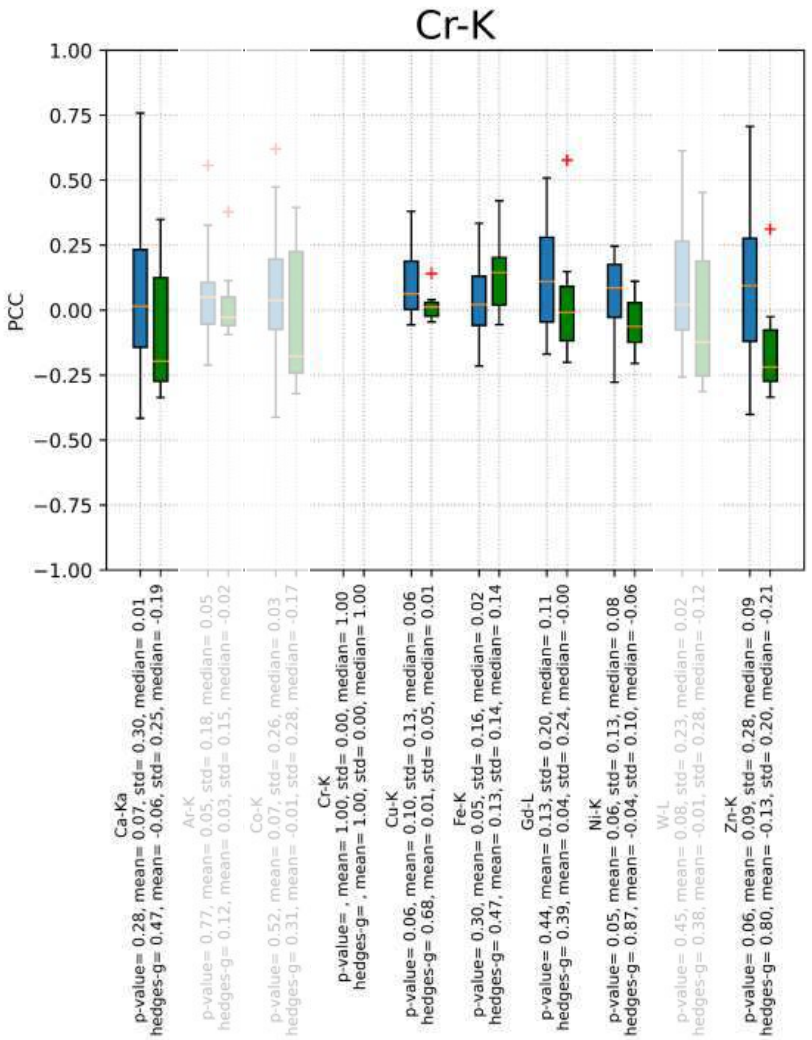
GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ni (p-value= 0.04) and Zn (p-value= 0.00) show a significance difference. For all other elements, no significant differences can be shown.

Cu shows a significance difference (p-value= 0.06).

Ni shows a significance difference (p-value= 0.05).

For all other elements, no significant differences can be shown.



GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

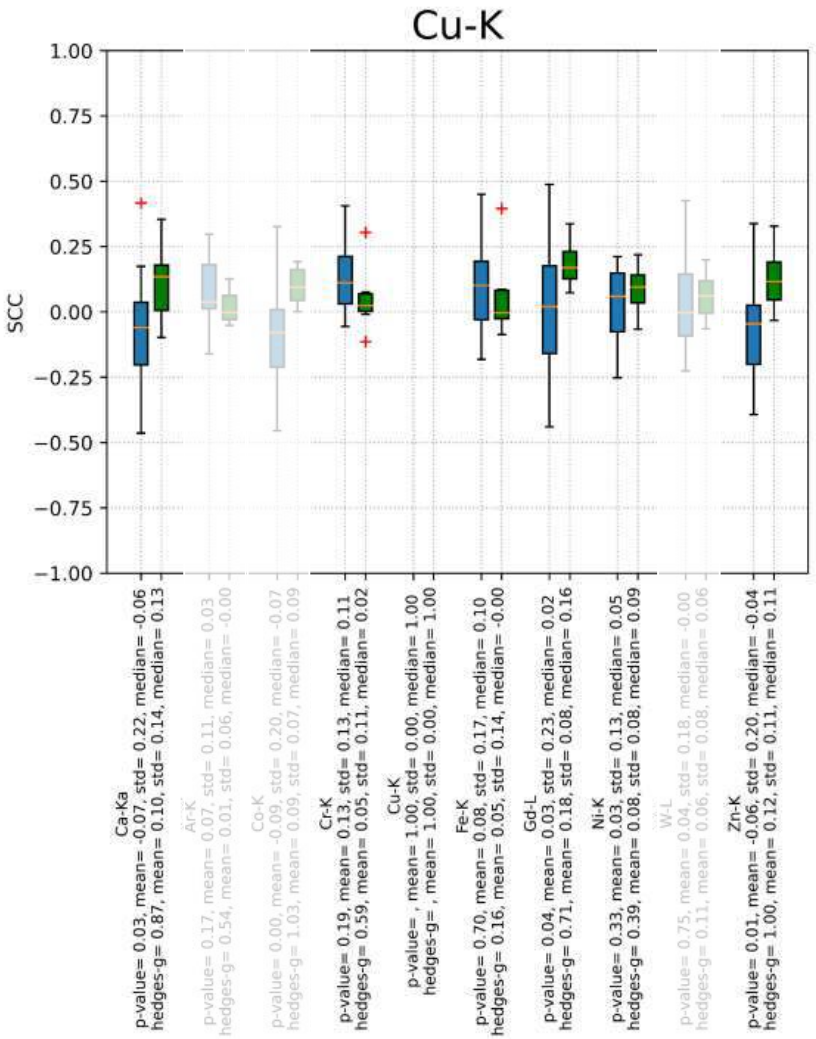
GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Cr shows a significance difference (p-value= 0.06).

For all other elements, no significant differences can be shown.

Ca (p-value= 0.03) and Zn (p-value= 0.01) show a significance difference. For all other elements, no significant differences can be shown.



GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ni shows a significance difference (p-value= 0.05).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Zn shows a significance difference (p-value= 0.03).

For all other elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

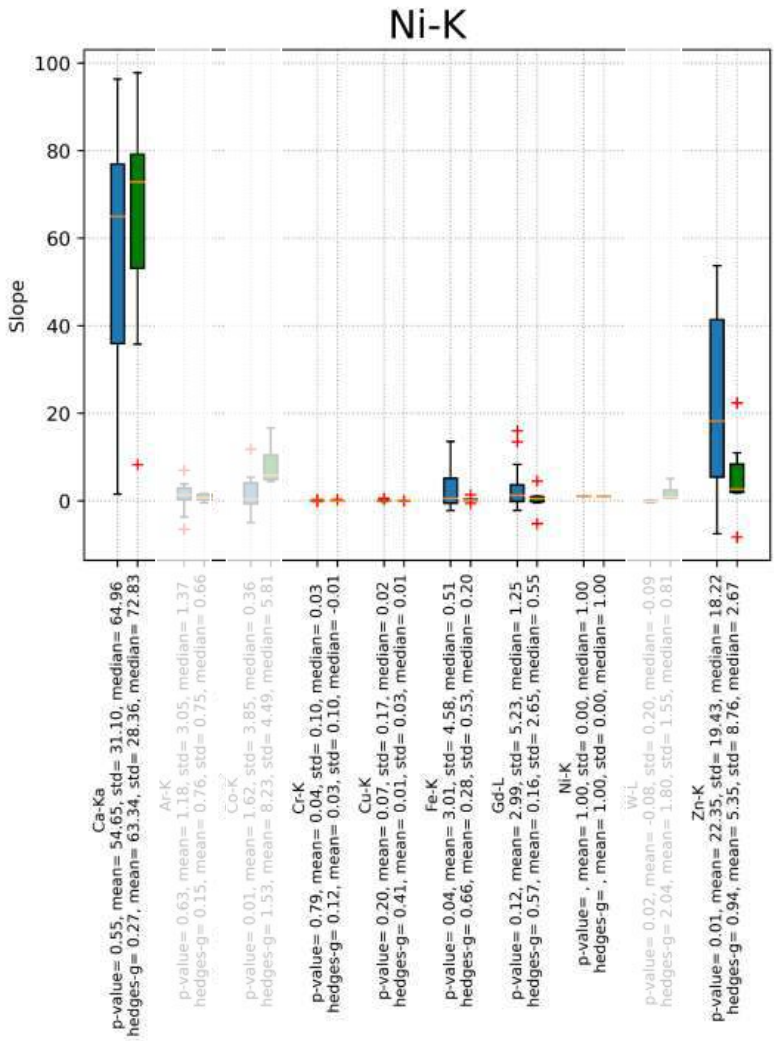
For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ca (p-value= 0.01) and Zn (p-value= 0.03) show a significance difference. For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe (p-value= 0.04) and Zn (p-value= 0.01) show a significance difference. For all other elements, no significant differences can be shown.



GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe shows a significance difference (p-value= 0.03).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Cu (p-value= 0.01) and Ni (p-value= 0.03) show a significance difference. For all other elements, no significant differences can be shown.

Ni shows a significance difference (p-value= 0.04).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Data Interpretation

M1: head element
overlaps x-Axis

Statistic
of MOC

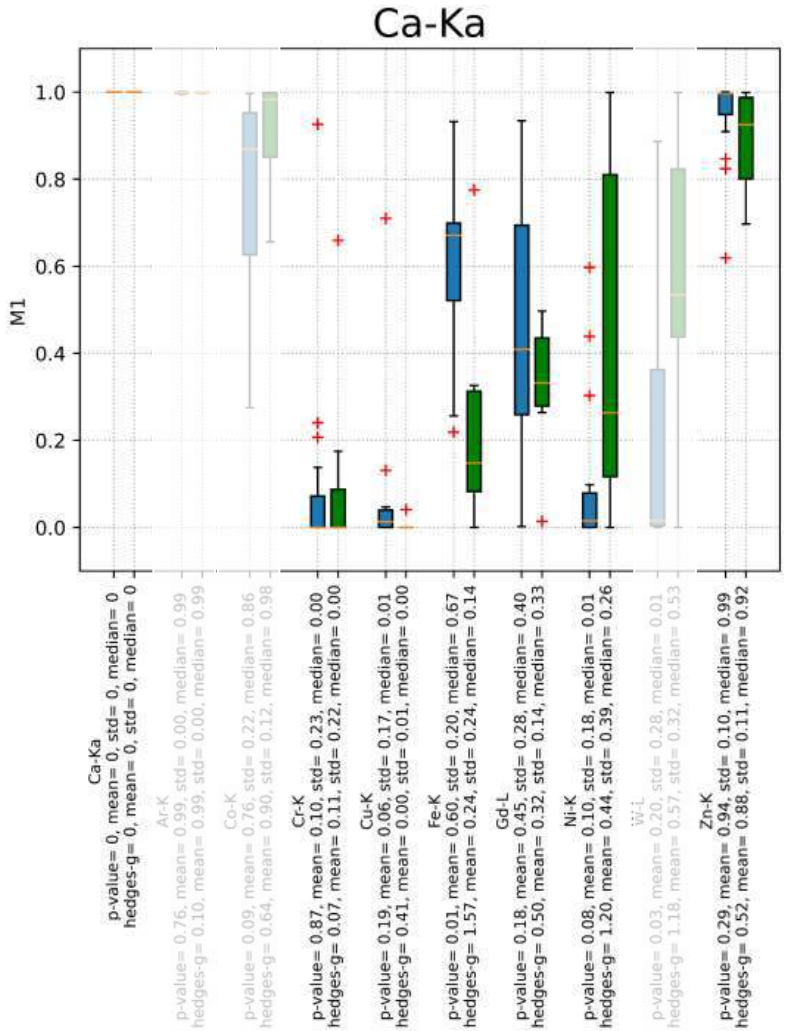
Compare
GD1 (n=7) & GD5 (n=15)

Significance Level
p <=0.05 (Groups Unequal)

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

Fe shows a significance difference (p-value= 0.01).

For all other elements, no significant differences can be shown.



GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ni shows a significance difference (p-value= 0.03).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe (p-value= 0.00) and Ni (p-value= 0.03) show a significance difference. For all other elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe shows a significance difference (p-value= 0.00).

For all other elements, no significant differences can be shown.

Data Interpretation

M2: x-Axis overlaps the head element

Statistic of MOC

Compare GD1 (n=7) & GD5 (n=15)

Significance Level p <=0.05 (Groups Unequal)

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ca (p-value= 0.01), Gd (p-value= 0.00) and Zn (p-value= 0.00) show a significance difference. For all other elements, no significant differences can be shown.

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe (p-value= 0.03) and Gd (p-value= 0.03) show a significance difference. For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

Data Interpretation

M: M1 and M2 combined

Statistic of MOC

Compare GD1 (n=7) & GD5 (n=15)

Significance Level
p <=0.05 (Groups Unequal)

Elements of interest: Ca, Cr, Cu, Fe, Gd, Ni, Zn
Excluded elements: W (blade), Co (blade), Ar (air)

Fe shows a significance difference (p-value= 0.01).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

For all elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Ca (p-value= 0.01), Gd (p-value= 0.02) and Zn (p-value = 0.01) show a significance difference. For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe shows a significance difference (p-value= 0.02).

For all other elements, no significant differences can be shown.

Zn shows a significance difference (p-value= 0.04).

For all other elements, no significant differences can be shown.

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

GD5 (blue left) tumorous
GD1 (green right) osteoporotic

Fe (p-value= 0.01) and Ni (p-value= 0.04) show a significance difference. For all other elements, no significant differences can be shown.

For Motivation

"Where we're going we don't need roads"

-Dr Emmett Brown