## Project Partner

In 12 posts, we report about our recent research project #BETTERWHEAT. It is coming to an end after 5 years, funded by @bmel via #innovation_ptble. It is a collaboration between @UniHohenheim, @UniMainz, four breeding companies KWS Group, LG, DSV, WvB and baking lab DIGeFa.

Combining innovative methods of genomics, proteomics and phenotyping of >100 traits, the #BETTERWHEAT project aims to elucidate the fundamentals of quality traits and their stability in the context of changing environmental and cultivation conditions along the wheat supply chain
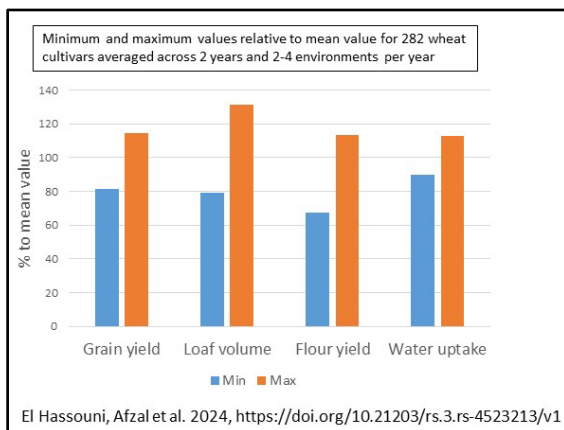
We tested >600 wheat lines across many locations and years and determined beside agronomic also numerous quality traits and dozens of minerals & sugars. All lines were genotyped with > 30K markers, all harvest samples measured with 3 spectrometers and > 6.000 proteins were quantified per sample
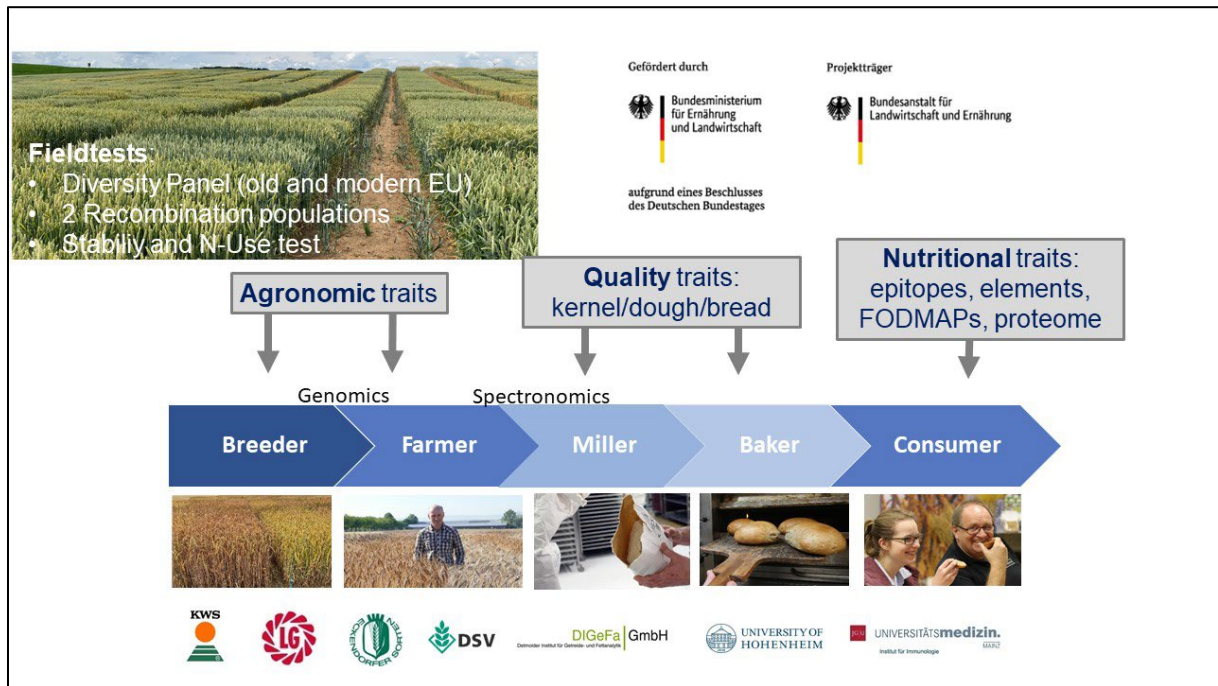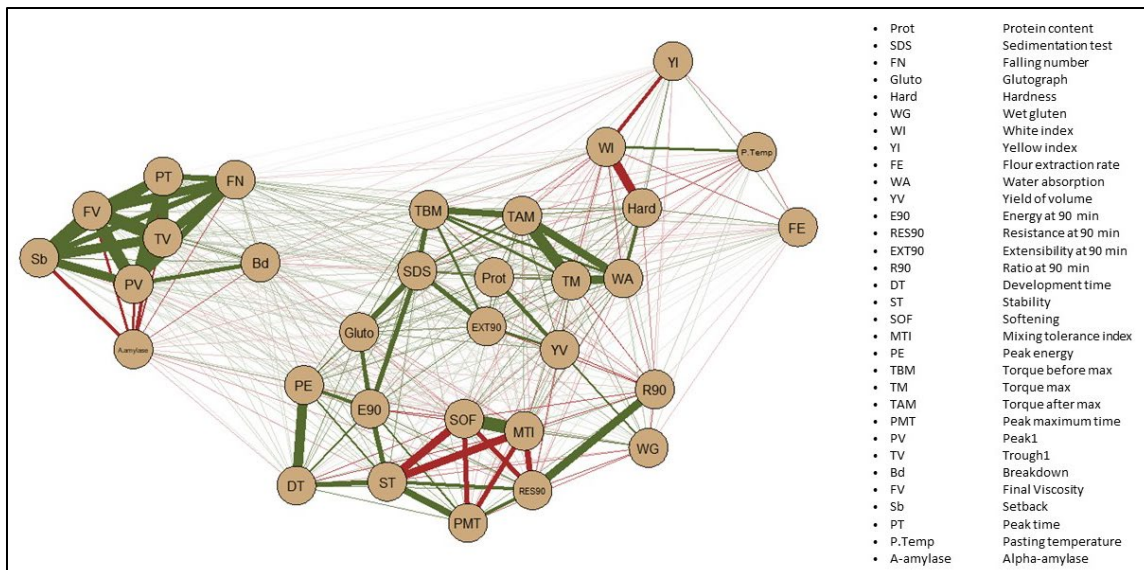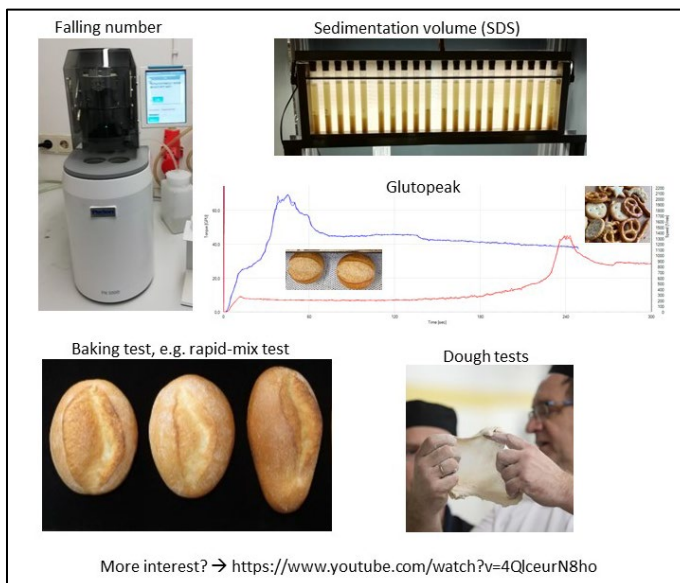
Three PostDocs and two PhD students drive the project #BETTERWHEAT. Besides tremendous coordination efforts, tasks encompass classical phenotypic analyses of field data, genome-wide association mapping, proteome profiling and the use of machine learning for prediction of baking quality by spectroscopy and numerous other traits/proteins

#Betterwheat results 1: The tested wheat cultivars originated from D and surrounding countries from different decades of wheat breeding. They largely differed in agronomic and quality traits; e.g. yield ranged from 7.3-10.7 t/ha, bread loaf volume between 472-782ml, flour yield between 35-60% & water uptake of dough between 51- 64%. This highlights the enormous potential of choosing the right cultivar for the intended use. Nevertheless, all investigated traits were influenced besides genetics also by the test environment underlining the need of monitoring wheat qualities along the supply chain.

Minimum and maximum values relative to mean value for 282 wheat cultivars averaged across 2 years and 2-4 environments per year

El Hassouni, Afzal et al. 2024, https://doi.org/10.21203/rs.3.rs-4523213/v1

#Betterwheat results 2: Loaf volume from rapid-mix-test correlated most strongly (r = 0.7) with protein content and Extensograph, followed by SDS, Glutograph and Glutopeak. Methods describing starch quality (falling, RVA, amylase activity) did not correlate with loaf volume. Dough properties 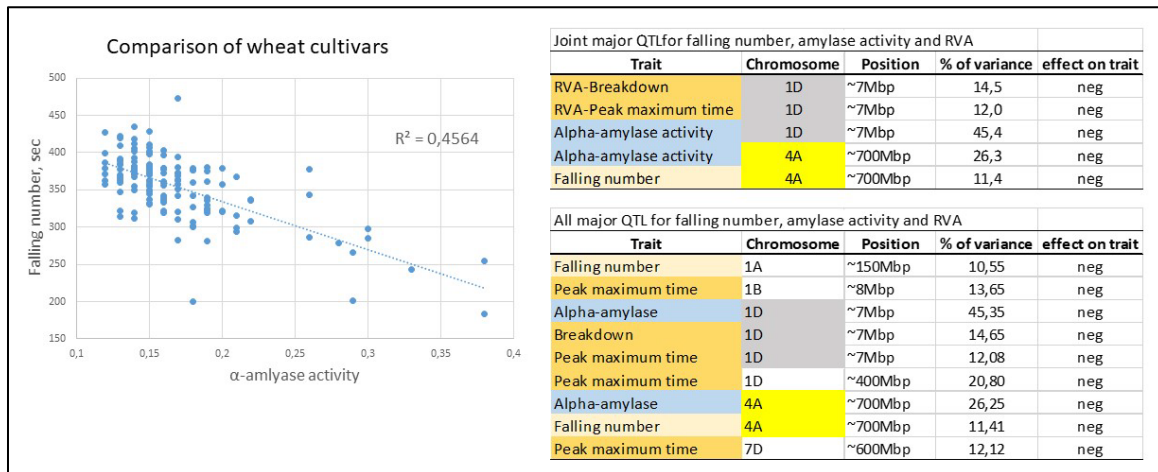measured by Extensograph correlated most strongly with SDS, Glutograph and some values of Glutopeak. Summarizing, from the different tested lab machines, Extensograph appeared to be the best suited for measuring dough and baking quality of German wheat cultivars. Please note, the rapid-mix-test of baking is the gold standard for cultivar testing in D but to my opinion, it is not realistic for practical baking.

Falling number   Sedimentation volume (SDS)

Glutopeak

Baking test, e.g. rapid-mix test   Dough tests

More interest? → https://www.youtube.com/watch?v=4QJceurN8ho

- Prot — Protein content
- SDS — Sedimentation test
- FN — Falling number
- Gluto — Glutograph
- Hard — Hardness
- WG — Wet gluten
- WI — White index
- YI — Yellow index
- FE — Flour extraction rate
- WA — Water absorption
- YV — Yield of volume
- E90 — Energy at 90 min
- RES90 — Resistance at 90 min
- EXT90 — Extensibility at 90 min
- R90 — Ratio at 90 min
- DT — Development time
- ST — Stability
- SOF — Softening
- MTI — Mixing tolerance index
- PE — Peak energy
- TBM — Torque before max
- TM — Torque max
- TAM — Torque after max
- PMT — Peak maximum time
- PV — Peak1
- TV — Trough1
- Bd — Breakdown
- FV — Final Viscosity
- Sb — Setback
- PT — Peak time
- P.Temp — Pasting temperature
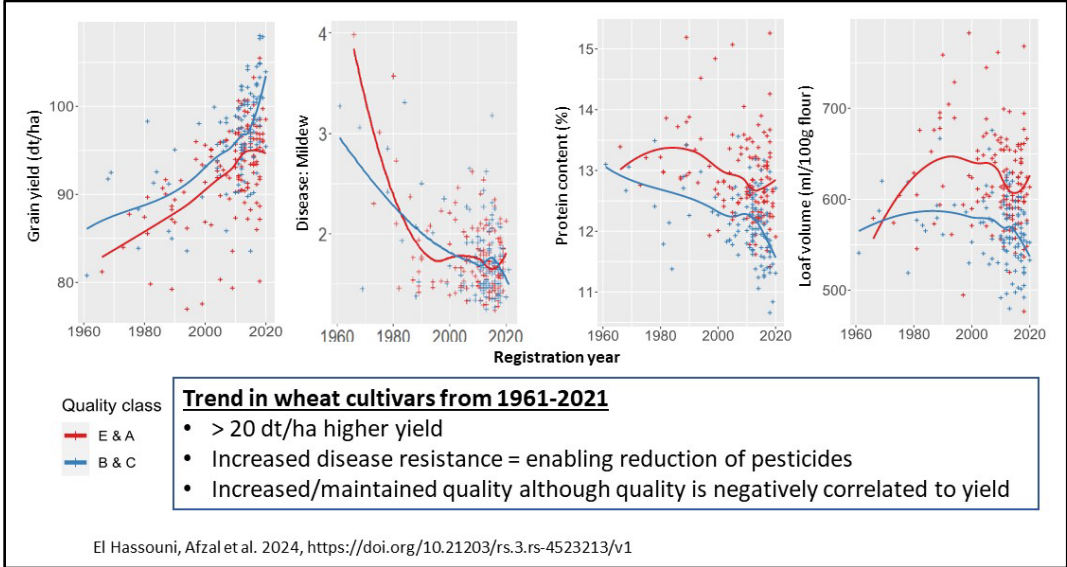- A-amylase — Alpha-amylase

#Betterwheat results 3: Near-infrared, Raman and Fluorescence spectrometry were used to predict quality parameters of wheat cultivars using either whole grain or fine flour. Correlations between predicted and measured values were surprisingly high (r > 0.8) for many quality traits like loaf volume, SDS, water uptake of dough and dough traits from different quality-testing machines. Considering the speed of spectrometry at whole grains, we highly recommend more R&D to rapidly establish a routine use of these approaches for detailed quality predictions along wheat supply chains.

- Correlation between predicted per NIRs and measured quality traits were r > 0.8 for bread loaf volume, SDS, water uptake, and few dough traits from Extensograph, Glutopeak and MicroDoughLab
- Error of prediction close to experimental lab error
- Fusion of spectra of different spectrometer hardly improved prediction
- Use of flour better than use of kernels
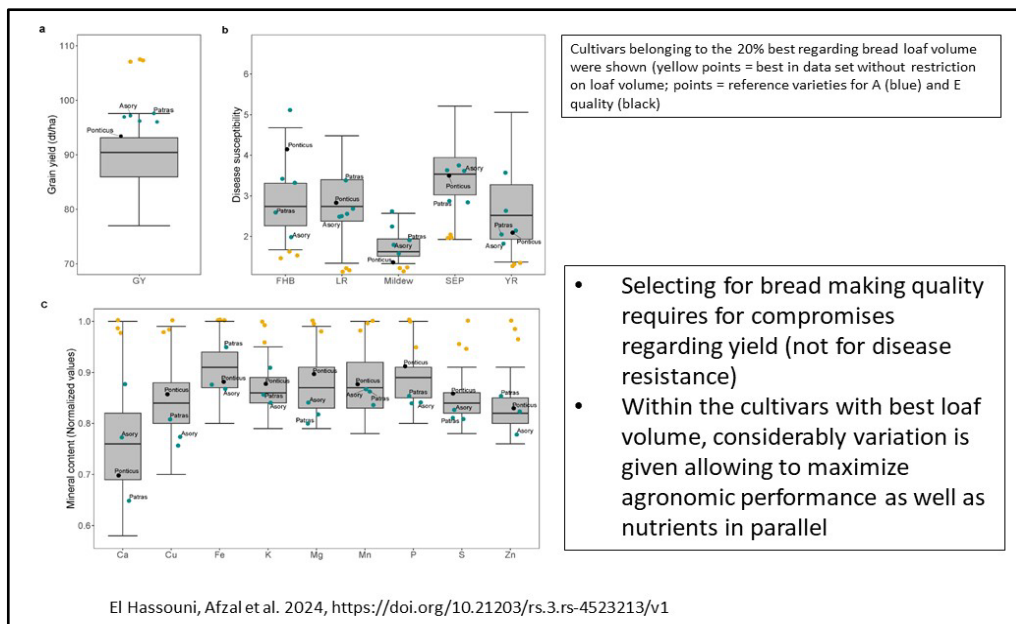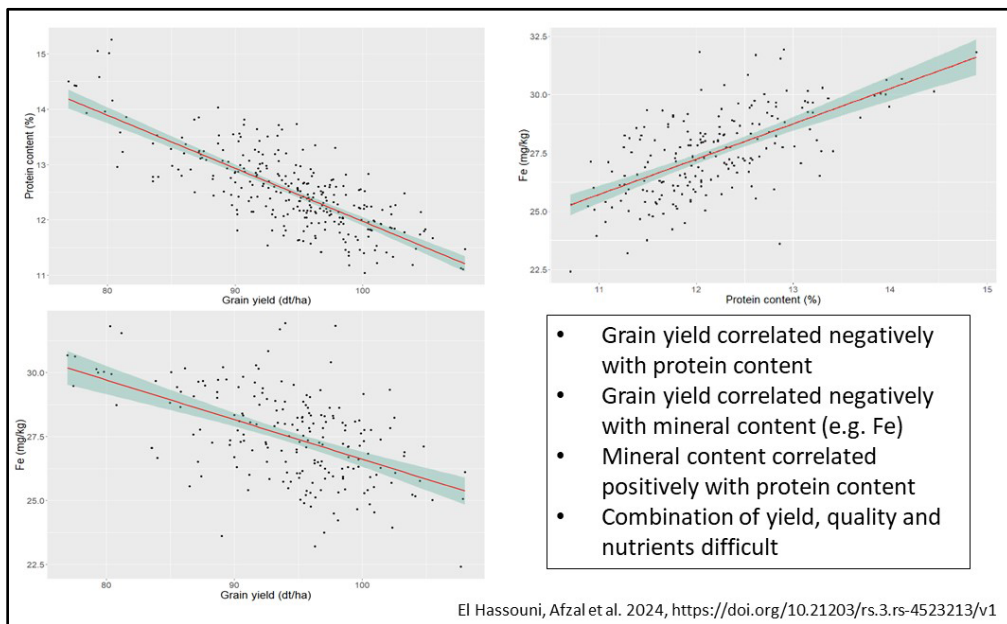
Nagel-Held et al. 2023; doi: 10.1002/cche.10732

#Betterwheat results 4: Starch properties measured by rapid visco analyser (RVA) correlated tightly with falling number (r>0.8), while correlation between α-amylase activity and falling number was lower (r = -0.68). The wide variability in falling number of wheat cultivars for the same amount of α-amylase activity should be noted as falling number is routinely used as fast method to estimate the amylase activity of flour. The phenotypic correlation between these traits were partly confirmed by identifying joint genomic regions largely influencing these traits (major QTL). Interestingly, the major QTLs for α-amylase activity on chromosome 1D were also identified for some of the RVA traits but not for falling number. No additional major QTL was identified for α-amylase activity, which segregated independently from falling number or RVA.

Comparison of wheat cultivars

$R^2 = 0,4564$

Joint major QTL for falling number, amylase activity and RVA				
Trait	Chromosome	Position	% of variance	effect on trait
RVA-Breakdown	1D	~7Mbp	14,5	neg
RVA-Peak maximum time	1D	~7Mbp	12,0	neg
Alpha-amylase activity	1D	~7Mbp	45,4	neg
Alpha-amylase activity	4A	~700Mbp	26,3	neg
Falling number	4A	~700Mbp	11,4	neg

All major QTL for falling number, amylase activity and RVA				
Trait	Chromosome	Position	% of variance	effect on trait
Falling number	1A	~150Mbp	10,55	neg
Peak maximum time	1B	~8Mbp	13,65	neg
Alpha-amylase	1D	~7Mbp	45,35	neg
Breakdown	1D	~7Mbp	14,65	neg
Peak maximum time	1D	~7Mbp	12,08	neg
Peak maximum time	1D	~400Mbp	20,80	neg
Alpha-amylase	4A	~700Mbp	26,25	neg
Falling number	4A	~700Mbp	11,41	neg
Peak maximum time	7D	~600Mbp	12,12	neg

#Betterwheat results 5: Testing 282 wheat cultivars registered during different decades of breeding, we could show that breeding has largely improved disease resistance and grain yield, while protein content was reduced as logical consequence of the tight negative correlation with yield. However, bread loaf volume was maintained mainly to largely improved protein quality (SDS). Separating the wheat cultivars in high quality (red-colored lines/points) and low quality (blue-colored lines/points) highlights the difficulties to improve grain yield and baking quality at the same time.

Quality class
E & A
B & C

Trend in wheat cultivars from 1961-2021
- > 20 dt/ha higher yield
- Increased disease resistance = enabling reduction of pesticides
- Increased/maintained quality although quality is negatively correlated to yield

El Hassouni, Afzal et al. 2024, https://doi.org/10.21203/rs.3.rs-4523213/v1

#Betterwheat results 6: Mineral content (Fe, Zn, P, S) of wheat cultivars was positively correlated with their protein content (r > 0.6), but negatively with yield (r < -0.5). Selection for baking quality needs compromise for yield. Nevertheless, within the cultivars belonging to the 20% highest in bread loaf volume, remarkable variation was determined for agronomic performance and mineral content enabling to find the best compromise maximising yield for a certain level of baking and nutritional quality. Note, most of the mineral content is found in the outer layers of wheat grain, which is removed during milling to obtain fine flour. Increasing mineral content via plant breeding makes sense only after a considerable increase of whole grain consumption is realized.

- Grain yield correlated negatively with protein content
- Grain yield correlated negatively with mineral content (e.g. Fe)
- Mineral content correlated positively with protein content
- Combination of yield, quality and nutrients difficult

El Hassouni, Afzal et al. 2024, https://doi.org/10.21203/rs.3.rs-4523213/v1

Cultivars belonging to the 20% best regarding bread loaf volume were shown (yellow points = best in data set without restriction on loaf volume; points = reference varieties for A (blue) and E quality (black)

- Selecting for bread making quality requires for compromises regarding yield (not for disease resistance)
- Within the cultivars with best loaf volume, considerably variation is given allowing to maximize agronomic performance as well as nutrients in parallel

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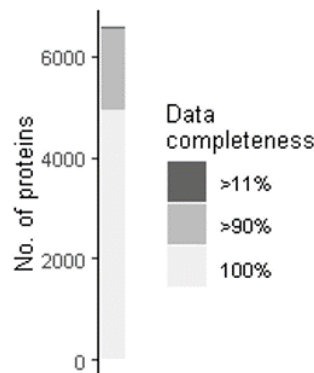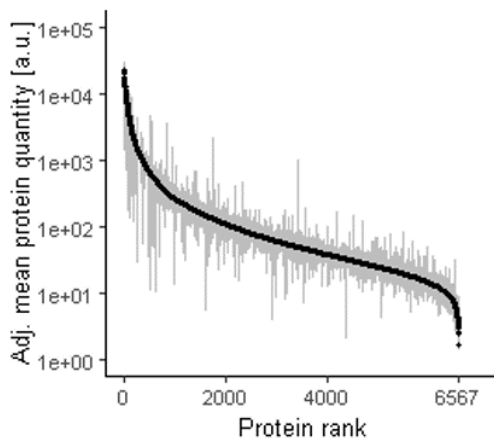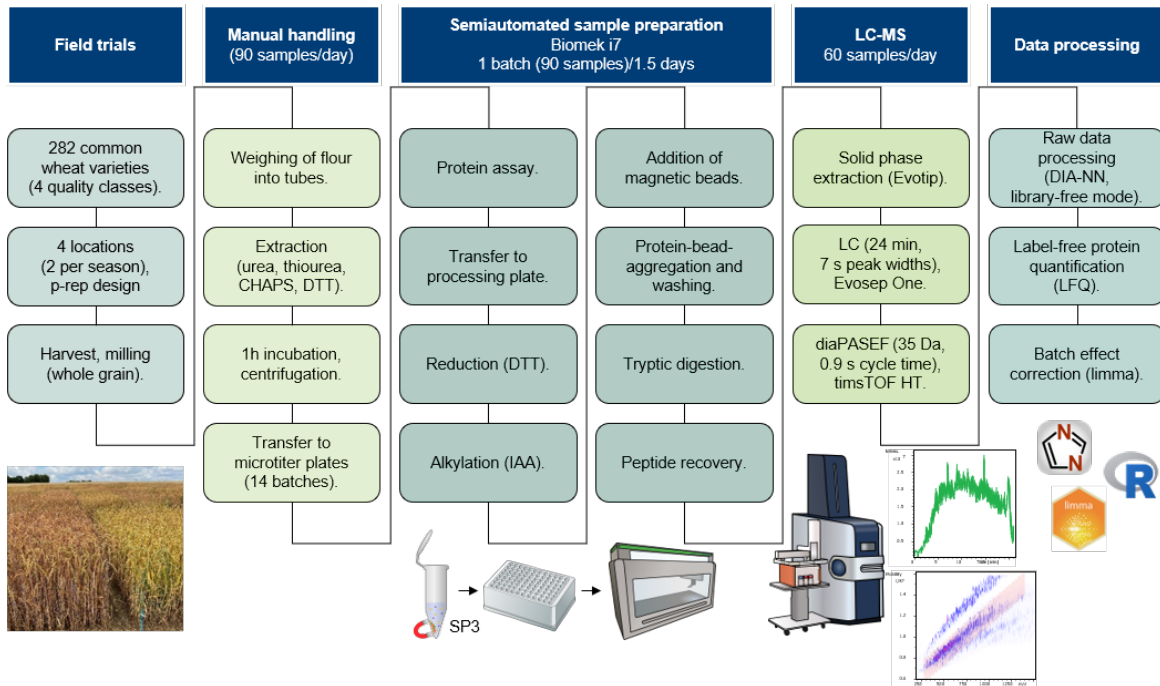
#Betterwheat results 7: One joint genomic region (major QTL) was found influencing protein and mineral content but with opposite trait effects while few small QTL for protein content were found in close vicinity to large QTL for mineral content. For Zn and yield, however, 2 out of 4 major QTLs found were in close genomic vicinity with opposite trait effects explaining partly the negative correlation of yield and Zn content. The QTL on chromosome 4A seems to influence beside Zn and yield also plant height and Ca content.

Joint major QTL Zn and yield or Zn and protein content				
Trait	**Chromosome**	**Position**	**% of variance**	**effect on trait**
Zinc	2B	~120Mbp	23,80	pos
Grain yield	2B	~120Mbp	13,50	neg
Protein content	2D	~700Mbp	9,16	neg
Zinc	2D	~700Mbp	10,55	pos
Zinc	4A	~700Mbp	32,70	pos
Grain yield	4A	~700Mbp	8,30	neg
Calcium	4A	~700Mbp	13,20	pos
Plant height	4A	~700Mbp	8,20	neg

All major QTL for Zn, yield and protein content				
Trait	**Chromosome**	**Position**	**% of variance**	**effect on trait**
Protein content	2A	~100Mbp	9,37	pos
Zinc	2B	~120Mbp	23,77	pos
Grain yield	2B	~120Mbp	13,51	neg
Protein content	2D	~700Mbp	9,16	neg
Zinc	2D	~600Mbp	10,55	pos
Zinc	3B	~800Mbp	8,20	pos
Zinc	4A	~700Mbp	32,72	pos
Grain yield	4A	~700Mbp	8,33	neg
Grain yield	5A	~600Mbp	7,09	neg
Grain yield	5B	~600Mbp	14,88	neg
Protein content	7B	~130Mbp	17,18	pos

#Betterwheat results 8: High-throughput LC-MS proteomics was established @tenzerlab delivering after intensive extraction and cleaning steps >6.000 proteins in 1.200 samples (282 wheat cultivars grown at 4 locations) with data completeness >90%. After strict filters for heritability >0.5 and environment-stable expression across locations, ~2.600 proteins were left for in-depth analyses regarding correlation to agronomic, quality and nutritional traits or genome-wide association mapping. This represents to our knowledge the first "pan-proteome" reported for wheat grain.

#Betterwheat results 9: Protein content and grain yield were strongly negatively correlated, which was partly influenced by genes located in similar genomic regions (Pleitotropic QTLs). Similarly, 11 proteins influenced both grain yield and protein content in parallel with 5 of them having QTLs in above reported genomic regions → more in depth research warranted ☺

Trait	Chromosome	Protein
Yield	2B	IPN_3465
Protein content	2B	IPN_3465
Yield	3A	IPN_1147, IPN_3465
Protein content	3A	IPN_1147, IPN_3465
Yield	5B	IPN_2248
Protein content	5B	IPN_2248
Yield	6A	IPN_3465
Protein content	6A	IPN_3465