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Hans H. Ruthenberg Award for Graduates 2019

Birgit Bierschenk “Rice wild relatives under different iron stress regimes growth - parameters, yield and grain quality factors”, University of Bonn, 2018

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Summary

The Problem

Oryza sativa, the Asian rice, accounts for 23 % of the world's calorie supplies and it represents the major staple food for most people in several less developed countries of Asia, Africa and South America. This is why an improvement in the yield quantity and quality of this crop in small-scale production and under adverse conditions would contribute greatly to the fulfillment of one of the major points of the UN Sustainable Development Goals, the food security.

The most common nutritional disorder in the production of lowland rice is iron toxicity. Especially in tropical regions, iron concentrations in the old and weathered soils tend to be high: In flooded fields, the mineral is reduced to Fe^{2+} which is easily taken up by plants and leads from huge yield losses up to complete failures. It is a great threat especially for poor farmers who depend on their harvest to feed their families but lack knowledge, funds and manpower to undertake alleviating measures. A lot of research on this topic has already been undertaken in the past, but no universal solution could be concluded upon. Providing the farmers with seeds of iron tolerant rice plants has often been stated as a feasible solution, but up to date no plants with sufficient iron tolerance have been obtained.

Meanwhile, to alleviate hunger and poverty, not only food quantity should be borne in mind, also food quality needs to be considered. Nutrient deficiency -the hidden hunger -has multiple detrimental effects on human body development and performance, to the extent that its effects can be perceived on major scales like the national socioeconomic development. The Copenhagen Consensus listed especially deficiencies of iron and zinc as a top priority in 2008 in order to improve the health of poor people and as an aside enhance the labor force in less developed countries. In prepared rice dishes the concentration of these minerals is quite low. Increasing the mineral content of rice grains would therefore be a huge nutritional improvement, especially for those whose diet consists of the cereal to a great extent. Another problem in this context is the anti-nutrient phytate, which is present in rice in considerably high quantities and impairs the uptake of the already scarce minerals in the food. Up to date

the breeding of low phytate varieties is still a challenge, because the molecule is important for several physiological processes in the plant that have not fully been understood yet. A natural plant with a low phytate content would be of use for a better understanding of this complex topic.

Both the problem of iron toxicity as well as the nutritional quality in the case of *Oryza sativa* have been addressed by means of field management or advanced genetic engineering. But these approaches, apart from the in many cases only marginal positive effect, are often not feasible for poor farmers as they are cost or labor intensive and also often not well accepted.

The Objective

The present study seeks to target these two distinct but related problems at the same time. The aim was to find varieties with beneficial traits regarding iron toxicity tolerance and grain mineral content for further breeding. This approach is far from new, but up to date no considerable screenings of the here presented extend have been undertaken among the wild relatives of rice. Even though the high potential for promising findings should be evident -the at least 23 relatives of the cultivated rice in the *Oryza* genus are distributed pantropical in very different environments and as a result the probability is high that their genomes harbor adaptations to several biotic and abiotic stresses. And this is even more pertinent when considering that the cultivated species *Oryza sativa* went through a severe bottleneck during domestication and therefore has relatively little genetic variation to offer.

Methodological Approach

For the experimental setup, a semi-natural system with soil plots in a greenhouse was chosen. It is closer to real field conditions than a hydroponic system, while influencing factors like weather conditions are easy to control. Fifty-eight wild rice accessions of 20 species and 17 cultivated varieties were grown in 6 soil plots of 2 x 6 m under greenhouse conditions leading to two polders each for the three treatments chronic iron stress, acute iron stress and control. Four semi-randomized replicates were planted in each polder. Iron was applied in the form of iron(II) sulfate heptahydrate ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) in such amounts that an approximate concentration of 1500 mg/l of reduced iron in the upper soil solution was reached in acute treatment, while for chronic treatment a concentration of 200 to 300 mg/l was aspired. The control polders received a corresponding amount of sulfate in form of 5 kg calcium sulfate (CaSO_4).

Temperature and air humidity were measured constantly. The plants were provided with adequate amounts of fertilizer and plant protection measures where necessary. Heading times of the plants were noted to get more information on development characteristics of the varieties on the one hand and to have another parameter for iron stress reactions on the other. After harvest plant height, tiller number, dry weight of straw biomass and grain yield were determined as parameters for iron stress tolerance. Iron, zinc and phytate grain concentrations were determined in brown rice of wild relatives and selected *O. sativa* varieties to characterize their nutritional quality.

Results

Plants under Iron treatment showed typical iron toxicity symptoms (further investigated in another master thesis). Growth parameters were increased under acute stress compared to control, while plants under chronic stress only showed an increase in height. Different

reasons for these findings are discussed in the main work. Iron stress affected the onset of reproductive stage in some genotypes, which might be of interest for basic stress research. Grain yield was elevated under acute stress but reduced under chronic stress compared to control. Yield losses were mainly caused by increased spikelet sterility and decreased thousand kernel weights under iron stress, which were compensated in acute treatment by an elevated panicle number. The *Oryza rufipogon* variety 105491 and *O. alta* 100161 showed signs of iron tolerance along with some varieties for which iron tolerance was already known, such as *O. glaberrima* CG 14 and *O. sativa* Dom Sofid. Especially the first mentioned should be considered for further research, because it produced the highest grain yields of all accessions including cultivated *O. sativa* varieties and it exhibited a high harvest index.

Grain analysis revealed average Fe values between 19.1 and 51.9 mg/kg per genotype but with a high variation among samples. Zn values on average ranged from 21.3 to 34.1 mg/kg and phytate concentrations from 6.8 to 12.8 g/kg. One *O. sativa* var. *spontanea* showed significantly less phytate compared to nearly all other varieties. Therefore it should be an interesting organism for further research. One *O. barthii*, one *O. alta* and *O. sativa* Dom Sofid could be of use for Zn biofortification through breeding. Grain Zn concentration was significantly elevated under chronic treatment and phytate concentration was higher in acute treatment compared to control while no effect was measured for Fe.

This pioneering work provided some interesting hints and new insights for further research regarding the use of wild relatives for rice breeding. The here mentioned varieties with favorable traits regarding iron toxicity tolerance or grain quality parameters should be investigated for the underlying mechanisms and also considered for breeding programs in the Future