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Genetic diversity for grain Zn concentration in finger millet genotypes: Potential for improving human Zn nutrition

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ABSTRACT

Nearly half of the world population suffers from micronutrient malnutrition, particularly Zn deficiency. It is important to understand genetic variation for uptake and translocation behaviors of Zn in relevant crop species to increase Zn concentration in edible parts. In the present study, genetic variation in grain Zn concentration of 319 finger millet genotypes was assessed. Large genetic variation was found among the genotypes, with concentrations ranging from 10 to 86 $\mu\text{g g}^{-1}$ grain. Uptake and translocation studies with Zn/⁶⁵Zn application in 12 selected low-Zn genotypes showed wide variation in root uptake and shoot translocation, with genotypes GEC331 and GEC164 showing greater uptake and translocation. Genotypes GEC164 and GEC543 showed increased grain Zn concentration. Genotypes GEC331 and GEC164 also showed improved yield under Zn treatment. Appreciable variation in grain Zn concentration among finger millet genotypes found in this study offers opportunities to improve Zn nutrition through breeding.

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1. Introduction

Micronutrients play vital roles in the biochemical and physiological functions of biological systems [1]. It is reported that the diets of over two thirds of the world's population lack one or more essential micronutrients [2]. Among micronutrients, zinc (Zn) deficiency accounts for many severe health complications [3]. Zn deficiency is one of the major risk factors in human health and cause of death globally. Plants play a vital role in human nutrition by providing all essential nutrients required

for human health [4]. Zn deficiency in soils and plants is a global micronutrient deficiency problem reported in many countries. Notably, 50% of cultivated soils in India and Turkey, a third of cultivated soils in China, and most soils in Western Australia are classified as Zn-deficient [5].

Conventional approaches such as Zn supplementation or fortification and dietary diversification adapted to ameliorate Zn deficiency in humans are neither practical nor cost-effective in the developing world. Genetic strategies for Zn biofortification are more practical, sustainable, and cost-effective [6,7]. It is possible

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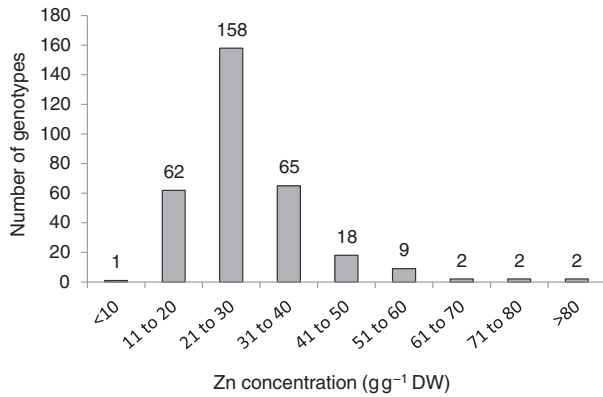


Fig. 1 – Frequency distribution of grain Zn concentration in 319 finger millet genotypes. The grain of 319 finger millet genotypes grown in three different seasons on red soil was analyzed for Zn concentration using Atomic absorption spectroscopy (AAS).

to improve Zn concentration in crops grown on deficient soils by exploiting genotypic differences in Zn uptake and tissue-use efficiency that are present in crop species [8,9]. Strategies to increase Zn concentrations in edible portions seek to exploit genetic variation in acquisition of Zn from the soil, accumulation in edible portions, and tolerance of high tissue Zn concentrations [2]. There is considerable species and varietal variation in Zn uptake, translocation, and storage in edible parts of crop plants, despite the very low Zn concentrations in edible parts of all food crops [10]. It is important to identify staple food crops specific to the Zn deficient regions of the world and address Zn deficiency issues. This activity requires a comprehensive exploration of potential genetic resources in regional crops and an in-depth understanding of the physiological and genetic basis of nutrient accumulation processes in seeds. In this study, we assessed the diversity in grain Zn concentration of 319 genotypes of finger millet [*Eleusine coracana* (L.) Gaertn.], the predominant millet food crop of India and Africa. We also measured uptake and translocation differences in 12 selected low-Zn genotypes.

2. Materials and methods

2.1. Finger millet grain material

Grain of 319 finger millet genotypes, grown in three different seasons on red soil with Zn concentration of 7 g kg⁻¹ soil, was procured from All India Coordinated Research Project on millets (AICRP), Gandhi Krishi Vignana Kendra, University of Agricultural Sciences, Bangalore, India.

2.2. Zn treatment and Zn estimation

Twelve finger millet genotypes (GEC265, 460, 331, 164, 543, 392, 329, 440, 61, 236, 403, and 202) were grown in pots (21 cm height and 21 cm diameter) filled with red soil. Plants were provided with recommended levels of fertilizers (NPK 60:30:30 kg ha⁻¹). Fifteen days after sowing, plants were supplied with two concentrations of Zn in the form of

ZnSO₄. Three sets of plants were maintained at different Zn fertilization: a control (normal red soil containing 7 mg Zn kg⁻¹ soil), T1 (moderate Zn fertilization with 5 mg kg⁻¹ soil in addition to 7 mg kg⁻¹ present in the soil sample) and T2 (high Zn fertilization of 10 mg kg⁻¹ in addition to 7 mg kg⁻¹ present in the soil sample). One set of plants were harvested after 20 days of treatment to determine Zn concentrations in root and shoot, and grain was harvested from the second set of plants at physiological maturity. Zinc was estimated as described [11] on dry weight (DW) basis.

⁶⁵Zn treatment was given to 12 selected low-Zn genotypes, which were raised in perforated Styrofoam cups (10 cm long and 5 cm diameter) filled with soil mixture (soil, sand and farmyard manure at the ratio 5:4:1). Cups were placed on a sand bed to facilitate growth of roots. Twenty-one-day old seedlings in the cups were transferred to plastic trays containing 2.5 L of half-strength Hoagland's medium [12], such that hanging roots were completely immersed in the nutrient medium. After 4 days of acclimation, the regular Hoagland's solution was exchanged with 2.5 L of Hoagland's solution containing ⁶⁵Zn (obtained from Board of Radiation and Isotope Technology, Department of Atomic Energy, Mumbai, India; 40 mL of ⁶⁵Zn stock with specific activity of 196 μCi diluted with 60 L of Hoagland's solution). The experiment was performed in a radioisotope containment facility for 48 h. Plants were harvested and washed in calcium sulfate solution. ⁶⁵Zn activity in the fresh samples was measured with a liquid scintillation counter (WALLAC 1409, Perkin Elmer, California, USA). Radioactive disintegrations per minute were converted into pmol g⁻¹ fresh weight.

2.3. Statistical analysis

Data were analyzed for significant differences by ANOVA (generalized linear model procedure) using SAS software version 9.3 (SAS Institute Inc., Cary, NC, USA). Differences in short-term uptake of ⁶⁵Zn, transportation of ⁶⁵Zn, and grain Zn concentration under external Zn application in finger millet genotypes were tested by one-way ANOVA at $P < 0.05$. Differences in grain yield in finger millet genotypes under Zn application was tested by two-way ANOVA at $P < 0.05$.

3. Results

3.1. Genetic diversity for grain Zn concentration among finger millet genotypes

Genetic diversity for grain Zn concentration was studied among a large collection of 319 finger millet genotypes including core germplasm, locally adapted cultivars, and released varieties of India. Frequency distributions of grain Zn concentration are presented in Fig. 1 (for detailed data see Table S1). There was large variation among the genotypes, with Zn concentrations ranging from 10 to 86 μg g⁻¹. In nearly 50% of the genotypes (155) the Zn concentration was 21–30 μg g⁻¹ and only six genotypes showed >63 μg Zn g⁻¹ (Table S1). Genotypes with extreme Zn concentrations were called low and high grain Zn types and the 12 genotypes with less than 20 μg Zn g⁻¹ grain were selected for study of their uptake and translocation characteristics. The grain Zn concentrations in these genotypes ranged from 10 to 17 μg g⁻¹.

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