Development of Sorghum Tolerant to Acid Soil Using Induced Mutation with Gamma Irradiation

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ARTICLE INFO

Article history:
Received 20 November 2009
Received in revised form 20 April 2010
Accepted 21 April 2010

Keywords:
Sorghum breeding
Induced mutations
Dryland
Acid soil tolerance
Bioethanol

ABSTRACT

Water scarcity still becomes a problem in some dryland agricultural areas in Indonesia. Development of dryland farming system may be focused on crops that are required less water such as sorghum. Sorghum is a cereal crop that is usually grown under hot and dry condition and it is ideal for Indonesia. Sorghum is a good source of food, animal feed and raw material for ethanol. Indonesia is currently looking for alternative renewable energy resources and sorghum is regarded as one of the promising source of bioethanol as bioenergy. Unfortunately, most agricultural land in western part of the country particularly in Sumatera and Kalimantan is dryland and dominated by acid soil. The main constraint of crop production in acid soil is deficiency and Al toxicity. Therefore, development of sorghum cultivation in dryland farming system requires a variety which is tolerant to such conditions. Sorghum breeding for acid soil tolerance had been conducted at PATIR-BATAN by using induced mutations with gamma irradiation. The breeding objective was to search for sorghum genotypes tolerant to acid soil condition and with regard to sorghum use for bioethanol production. A number of 66 breeding materials, including the mutants, had been screened for acid soil tolerance on land with soil pH of 4.2 and 39% Al saturation in Lampung Province. Ten sorghum genotypes had been identified as high yielding in the acid soil condition. The mutant lines GH-ZB-41-07, YT30-39-07, B-76 and B-92 had grain yield higher (>4.5 t/ha) than the control plants (Durra, Mandau and Numbu). Sorghum mutants ZH30-29-07, ZH30-30-07 and ZH30-35-07 were promising for grain-base bioethanol production with ethanol yield exceeded 2,000 l/ha. Meanwhile, the sweet sorghum mutants ZH30-35-07, ZH30-30-07 and ZH30-29-07 had brix content of 11.59, 11.95 and 10.50%, respectively. These mutant lines are promising to be developed further in sorghum breeding since they are highly tolerant to acid soils.

INTRODUCTION

For agricultural purposes, arable land in Indonesia is actually dominated by dryland farming areas. Meanwhile, wetland (paddy field) areas tend to decrease due to several reasons such as intensive expanding of land transformation to non-agricultural purposes happened recently. The fact that water scarcity still becomes a problem in some agricultural areas and it makes dryland farming system be more reliable for supporting sustainable agriculture development in the country. The development of dryland farming system may be focused on crops that are required less water such as sorghum.

Sorghum (*Sorghum bicolor*) is thought of being suitable crop for dry land farming system in Indonesia owing to its wide adaptability, drought tolerance, low-input crop and high yielding. Sorghum is a cereal crop that is usually grown under hot and dry conditions. In many countries sorghum is generally used as food source, animal feed, and raw materials for industry. Based on form of its spike and basic spikelet, sorghum is classified into 5 races namely *Bicolor*, *Guenia*, *Caudatum*, *Kafir*, and *Durra* [1]. Sweet sorghum having high sugar content in its stalk is the one commonly used for syrup, sugar (*jaggery*), and ethanol industry.

Sorghum has a high yield potential, comparable of rice, wheat, and maize. On a field basis, yields have exceeded 11 ton/ha, with above average yields ranging from 7-9 ton/ha where moisture is not a limiting factor. In those areas where sorghum is commonly grown, yields of 3-4 ton/ha are obtained under normal condition [2]. Sorghum is also known to have wide adaptability, ranging from lowland, medium and highland

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altitude. Highest yields are usually obtained from varieties maturing in 100-120 days. Late-maturing varieties tend to be appropriate for forage crop. Available sorghum genotypes have been introduced from abroad e.g. from the International Crop Research Institute for the Semi-Arid Tropics (ICRISAT). Through plant breeding programs, some local varieties have also been released by the Ministry of Agriculture. Further sorghum breeding, however, is needed especially to search for and to develop genotypes tolerant to adverse conditions such as soil acidity.

Indonesia is currently looking for alternative renewable energy resources. Sorghum is one of the source of bioethanol with high productivity. The productivity of bioethanol from sweet sorghum reached 8.419 liter/ha/year i.e. twice that of cassava (3.835 liter/ha/year) since sorghum can be planted twice a year in Indonesia. Sorghum is ideal to be developed in Indonsia because it is suitable for dryland areas and requires less irrigation. From a total of 99.50 million ha of dryland agricultural area in Indonesia, about 68.75 ha (69.1%) consisted of acid soil (*ultisols*) which is mainly found in Sumatera and Kalimantan [3]. The main constraint of crop production in acid soils is phosphor (P) deficiency and aluminium (Al) toxicity [4, 5, 6]. Therefore, development of sorghum cultivation in dryland farming system requires varietes which are tolerant to such conditions. Besides drought tolerance sorghum is believed to have wide adaptability to various types of soil including acid soils. The objective of this research was to search for sorghum genotypes tolerant to acid soil condition and especially with regard to sorghum use for bioethanol production.

EXPERIMENTAL METHODS

Sorghum Durra variety originated from ICRISAT in India was used as starting breeding material. This variety is widely grown in India owing to high yielding and its potential use for food and bioethanol industry. Genetic improvement of this sorghum variety was carried out at the Center for the Application of Isotopes and Radiation Technology, National Nuclear Energy Agency (BATAN). Induced plant genetic variability was made by mutation techniques using gamma irradiation. The schematic procedure of inducing mutations is shown in Fig. 1. Dry seeds of sorghum with water content of 12 % were irradiated with gamma rays emitted from Cobalt-60 source in the Gamma Chamber 4000A. The dose level of 300-500 Gy of gamma rays was applied as an optimal dose range for sorghum breeding [7].

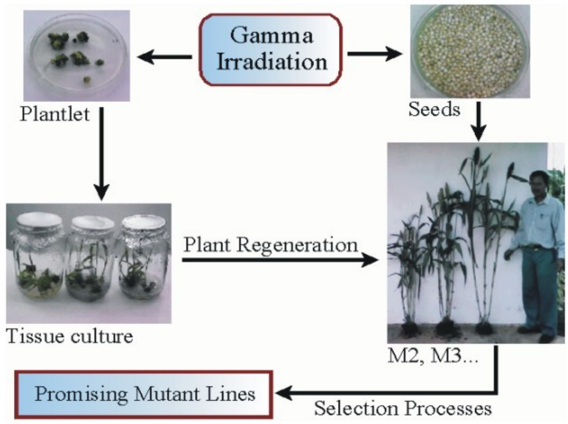

Fig. 1. Schematic procedure of inducing mutations in sorghum using gamma irradiation treatment on seeds or in-vitro plantlet.

All M1 plants were harvested to generate about 4000 M2 plants in the field. Individual plant selections based upon phenotypic variations were started in the segregating population of M2, focusing on agronomic and yield component characters. The seeds of selected M2 plants were multiplied in the M3 for direct screening of acid soil tolerance in the field.

A number of 66 selected breeding materials included in the screening experiment are listed in Table 1. The parental variety Durra and five national varieties i.e. Higari, Mandau, Kawali, Numbu and UPCA-S1 were included and used as control plants. Screening for acid soil tolerance was conducted in the M4 during dry season in Lampung Province and under natural irrigation. The plants were grown at Taman Bogo areas where soil condition was classified as very acid with soil pH of 4.2 and 39% Al saturation. Plant growth, grain yield, biomass, stem juice, brix content and ethanol production were measured and used as criteria of selection for acid soil tolerance. Analysis of bioethanol was conducted at the laboratory of Center for Starch Technology (B2TP-BPPT) at Sulusuban in Central Lampung. Ratooning ability was also studied with regard to bioethanol production. The experiment used randomized design with 10 samples taken from selected tolerant genotypes only.

Table 1. Breeding materials included in the screening of acid soil tolerance.

No	Genotype	No	Genotype	No	Genotype	No	Genotype
1	BR-ZH30-01-07	18	ZH30-19-07	35	ZH30-36-07	52	B-72
2	BR-ZH30-02-07	19	ZH30-20-07	36	ZH30-37-07	53	YT30-3P-06
3	BR-ZH30-03-07	20	ZH30-21-07	37	ZH-30	54	PSj-71-05
4	BR-ZH30-05-07	21	ZH30-22-07	38	YN30-38-07	55	PSj-72-05
5	BR-ZH30-06-07	22	ZH30-23-07	39	YT30-39-07	56	PSj-79-05
6	BR-ZH30-07-07	23	ZH30-24-07	40	YT30-40-07	57	PSj-80-05
7	BR-ZH30-08-07	24	ZH30-25-07	41	GH-ZB-41-07	58	PSj-81-05
8	ZH30-09-07	25	ZH30-26-07	42	GH-ZB-42-07	59	PSj-82-05
9	ZH30-10-07	26	ZH30-27-07	43	GH-ZB-43-07	60	Zhengzu
10	ZH30-11-07	27	ZH30-28-07	44	B-100	61	Higari
11	ZH30-12-07	28	ZH30-29-07	45	B-95	62	Mandau
12	ZH30-13-07	29	ZH30-30-07	46	B-92	63	Kawali
13	ZH30-14-07	30	ZH30-31-07	47	B-83	64	Numbu
14	ZH30-15-07	31	ZH30-32-07	48	B-76	65	Durra
15	ZH30-16-07	32	ZH30-33-07	49	B-75	66	UPCA-S1
16	ZH30-17-07	33	ZH30-34-07	50	B-69		
17	ZH30-18-07	34	ZH30-35-07	51	B-90		

RESULTS AND DISCUSSION

All breeding materials could grow but they gave various responses to the acid soil condition. Visually some genotypes showed highly tolerant in seedling stage but the other showed susceptible response (Fig. 2). Observation was particularly given to the tolerant genotypes by measuring data of their biomass, grain yield, stem juice, brix content and ethanol production. The grain yield data of the top ten tolerant genotypes are presented in Table 2. It was of interesting to show that some mutant lines i.e. GH-ZB-41-07, YT30-39-07, B-76 and B-92 had grain yield higher than the control plants (Durra, Mandau and Numbu). Even though still lower than Kawali, these mutant lines were promising to be developed further owing to highly tolerant to acid soils. Representing all of the mutant lines, the B-76 mutant was visually shown in Figur 3. In general all tolerant genotypes expressed more extensively fibrous root system in response to acid soils.

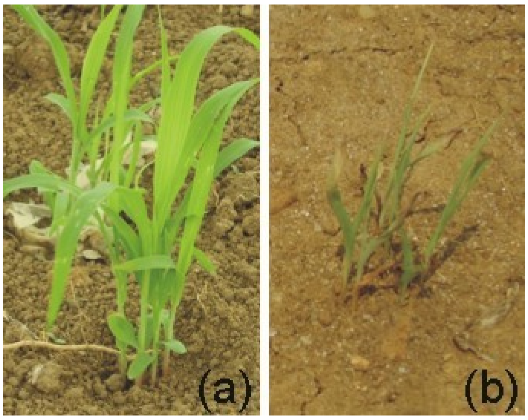

Fig. 2. Response of sorghum seedlings to acid soil with soil pH of 4.2 and 39% Al saturation in Lampung Province: (a) tolerant and (b) susceptible.

Table 2. Sorghum genotypes having high grain yield in acid soil.

Genotype	Grain Yield (t/ha)	Genotipe	Grain Yield (t/ha)
1. Kawali	6.14	6. B-92	4.58
2. GH-ZB-41-07	5.73	7. Durra	4.34
3. YT30-39-07	5.37	8. YT30-40-07	4.20
4. B-76	5.33	9. Mandau	4.20
5. ZH30-29-07	5.29	10. Numbu	4.20

Principally, bioethanol can be made from grain (carbohydrate) or stem juice (sugar) through fermentation process. Grain yield and carbohydrate and sugar content will determine bioethanol production. Table 3 shows the five genotypes having high bioethanol production made from grain (carbohydrate) base. Sorghum mutant ZH30-29-07, ZH30-30-07 and ZH30-35-07 were included in the promising lines for high grain-base bioethanol production exeeding 2,000 l/ha. The mutant line ZH30-29-07 had also good ratooning ability and produced grain yield of 2.08 t/ha in the second harvest (ratoon-1)

Table 3. Sorghum genotypes having high ethanol production from grain.

Genotype	Grain Yield (t/ha)		Total Grain Yield (t/ha)	Ethanol Production (l/ha)
	Seedling	Ratoon-1		
ZH30-29-07	5.29	2.08	7.37	3,169.1
Mandau	4.20	1.60	5.80	2,494.0
Numbu	4.20	1.57	5.77	2,481.1
ZH30-30-07	3.47	1.31	4.78	2,055.4
ZH30-35-07	3.81	1.04	4.85	2,085.5

Table 4. Sorghum genotypes having high ethanol production from sugar juice.

Genotype	Stem Production (t/ha)		Juice Production (l/ha)		Brix Content (%)	Ethanol Production (l/ha)
	Seedling	Ratoon-1	Seeling	Ratoon-1		
Numbu	39.30	12.40	7,562.5	1,493.8	14.75	694.65
ZH30-35-07	39.15	15.30	3,175.0	1,830.2	11.59	306.17
ZH30-30-07	33.04	13.21	3,462.5	935.4	11.95	275.83
ZH30-29-07	30.30	14.06	2,750.0	2,184.1	10.50	272.95
Mandau	20.75	11.33	1,168.8	1,039.1	9.25	106.86

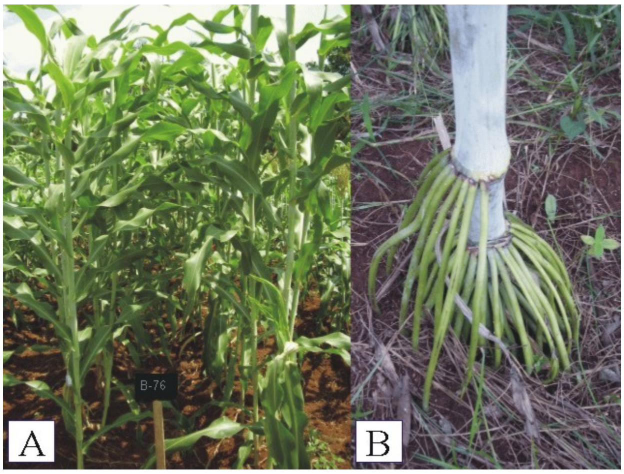

Fig. 3. One of acid soil tolerant sorghum mutants B-76 near flowering stage (A) and extensively fibrous root system in response to acid soils (B).

Numbu variety was the control of sweet sorghum included in this experiment. This variety had high brix (sugar) content of 14.75%, therefore, it could yield the highest ethanol production made from stem juice. The mutant lines of ZH30-35-07, ZH30-30-07 and ZH30-29-07 had brix content of 11.59, 11.95 and 10.50%, respectively. These mutant lines are also promising to be developed further since they are highly tolerant to acid soils. All the promising lines are now kept in sorghum germplasm collection at PATIR-BATAN for being used in further sorghum breeding program including hybridization or other related biotechnologies.

CONCLUSIONS

Acid soil is one of important problems in agricultural development especially on dryland farming system in Indonesia. Efforts to cope with this problem have been made by searching for adaptable crops to acid soil condition. Sorghum is believed to have wide adaptability to various types of soil including acid soils. Sorghum breeding using induced mutation with gamma irradiation has resulted a number mutant lines. From a total of 66 breeding materials screened for acid soil tolerance in Lampung Province, ten sorghum genotypes had been identified as high yielding in the acid soil condition. The mutants GH-ZB-41-07, YT30-39-07, B-76 and B-92 had grain yield higher than the control plants. It could be concluded that induced mutations with gamma irradiation had improved sorghum grain yield and increased plant genetic variability. Sorghum mutants ZH30-29-07, ZH30-30-07 and ZH30-35-07 were promising for grain-base bioethanol production. The sweet sorghum mutants ZH30-35-07, ZH30-30-07 and ZH30-29-07 had sugar content (brix) of 11.59, 11.95 and 10.50%, respectively. These mutant lines are promising to be developed further in sorghum breeding program since they are highly tolerant to acid soils.

ACKNOWLEDGEMENTS

We would like to thank the International Atomic Energy Agency (IAEA) for supporting this research through RAS/5/045 Project. Thanks also to Center for Starch Technology (B2TP-BPPT) in Lampung for helping us analyze the bioethanol produce of sorghum.

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