Breeding and Seed Reproduction Techniques of a New Tetraploid Common Buckwheat Variety 'Pintianqiao 3'

Xinghai SHI[1], Jiandong ZHAO[2*], Xiulian LI[1], Wenliang CHEN[1], Xiuzhen QIN[1], Jun REN[1], Xiaohuan YANG[1], Jinhu MA[1]

1. College of Agriculture, Shanxi Agricultural University, Taiyuan 030031, China; 2. Agricultural Genetic Resources Center, Shanxi Agricultural University, Taiyuan 030031, China

Abstract [**Objectives**] This study was conducted to actively carry out the breeding of new tetraploid common buckwheat varieties and its supporting breeding techniques. [**Methods**] Pintianqiao 3 is a new tetraploid common buckwheat variety developed by College of Agriculture of Shanxi Agricultural University and Agricultural Genetic Resources Center of Shanxi Agricultural University, using 'Pintianqiao 1' as the parent, through mutation treatment with 0.2% colchicine aqueous solution, grain selection, plant selection, isolation and identification, variety comparison, regional test and field investigation. The variety has chromosomes $2n = 4X = 32$, and shows a spring sowing period of 101 d and a summer sowing period of 80 d, large flowers and seeds (with a 1 000-grain weight of 41.4 g), and good resistance to lodging. [**Results**] From 2021 to 2022, Pintianqiao 3 participated in the independent joint regional test of common buckwheat varieties in Shanxi Province, and the average yield in 10 test positions was 1.8 kg, equivalent to 1 800 kg/hm^2, which was 8.4% higher than the control. It passed the field investigation conducted by Shanxi provincial expert group for identification of non-major crop varieties in Dongyang and Kelan experimental sites on September 2 -3, 2022. On January 4, 2024, it passed the preliminary examination of Shanxi Provincial Crop Variety Approval Committee. The seed reproduction technique of Pintianqiao 3 including land selection, preparation before sowing, sowing, field management and timely harvesting has been developed. [**Conclusions**] This study provides technical support for the demonstration and popularization of this new variety.

Key words Tetraploid; Pintianqiao 3; Breeding; Characteristic; Breeding technique

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Common buckwheat (*Fagopyrum esculentum* M.) is not only a kind of medicinal and edible plant, but also known as the star crop in the 21st century[1]. Moreover, its feed, honey, foreign trade export and other values should not be underestimated[2]. Common buckwheat is also a characteristic coarse cereal crop in Shanxi, with an annual planting area of about 20 000 hm^2. From the mid-1980s to the end of 2023, with the strong support of governments at all levels and the continuous efforts of scientific and technological personnel engaged in the research of new buckwheat varieties, eight common buckwheat varieties[3-4] were successively bred, including Ribentianqiao, Jinqiaomai 1, Jinqiaomai 3, Pintianqiao 1, Jinqiaomai 7, Jinqiaomai 8, Pintianqiao 2 and Youtianqiao 1, among which Jinqiaomai 7 is currently the only tetraploid common buckwheat variety in China that has been applied in production[5-7]. The lack of high milled rice rate, high flour yield[8] and high-grade and first-class export common buckwheat varieties[9] remains a technological bottleneck that restricts the development of the common buckwheat industry. Therefore, it is particularly important to actively carry out the breeding of new tetraploid common buckwheat varieties and the research on its supporting breeding techniques[10].

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Xinghai SHI (1971 -), male, P. R. China, associate professor, devoted to research about breeding and cultivation techniques of new buckwheat varieties.

* Corresponding author.

Parent Sources and Breeding Process

Parents

'Pintianqiao 1' was bred by the Agricultural Genetic Resources Center of Shanxi Agricultural University (formerly Institute of Crop Variety Resources of Shanxi Academy of Agricultural Sciences) in 2004 from the common buckwheat resource F326 stored in the National Agricultural Germplasm Resource Bank (Taiyuan) for isolation and propagation. In specific, three excellent individual plants with tall plants, lush growth, many flower clusters at the top and high seed-setting rate were selected and mixed to form a group, which was then developed to the final variety through many times of line group selection for many years. The variety has a growth period of 81 d. Its roots are robust and developed, and the plants grow neatly and consistently, with strong growth potential. The seedlings have green leaves and young stems, and the plant type is compact. The height of the main stem is 95 -148 cm. The stems are green at the upper part and light red at the lower part, and the main stem has 17.4 main stem nodes and 4.1 primary branches. The leaves are green and the flowers are white. The seeds are triangular and brown, and the number of grains per plant, grain weight per plant and 1 000-grain weight are 710 grains, 22.1 and 31.5 g, respectively. The seeds have a protein content of 9.16%, a fat content of 2.43%, a lysine content of 0.57%, a VE content of 0.78 mg/100 g, and a nicotinamide content of 5.82 mg/100 g. It was approved by the

Shanxi Provincial Crop Variety Approval Committee in 2014 and designated as Jinshenqiao (approval) 2014001.

Breeding process

In 2014, the growth points from Pintianqiao 1 seedlings were treated with colchicine aqueous solution, and hypocotyls, cotyledons, true leaves, flowers and the shape, size and color of grains of mutant (tetraploid) and control (diploid) plants were observed, compared, recorded, photographed and labeled at seedling stage, flowering stage and maturity stage, respectively. In the maturity stage, large seeds of different mutant plants were selected and mixed. Bunch planting was carried out in 2015, and 15 large-grain (tetraploid) plants were selected and threshed respectively. In 2016, a plant-to-row test was carried out, and four non-tetraploid plants were eliminated. In 2017, the plant-to-row test was continued, and six excellent plant rows were selected. In each excellent plant row, six excellent plant groups (lines) were selected, and five poor plant rows were eliminated. In 2018, the six lines were propagated in an isolated state. A comparison test was carried out in 2019 – 2020. The selected line participated in the comparative test of common buckwheat varieties in the Agricultural Genetic Resources Center of Shanxi Agricultural University (formerly Institute of Crop Variety Resources of Shanxi Academy of Agricultural Sciences) in 2019 – 2020, and it participated in the independent joint regional test of common buckwheat varieties in Shanxi Province in 2021 – 2022, and passed the field investigation conducted by Shanxi provincial expert group for identification of non-major crop varieties in Dongyang and Kelan experimental sites on September 2 – 3, 2022. On January 4, 2024, it passed the preliminary examination of Shanxi Provincial Crop Variety Approval Committee.

Characteristics

The growth period is 101 d for spring sowing and 80 d for summer sowing. The seedlings are green, contain no anthocyanins. The plants are 115.4 cm in height, and compact, and have green stems, each of which has 13.8 main stem nodes and 4.5 main stem branches. The leaves are dark green, and large, and the heart leaves are wrinkled. The leaf veins are green, and the flowers are white. The grain weight per plant is 4.4 g, and the 1 000-grain weight is 41.4 g. The seeds are brown-colored.

Yield Performance

Variety comparison test

In 2019 – 2020, Pintianqiao 3 participated in the comparison test of buckwheat varieties in Dongyang Base, Yuci, Agricultural Genetic Resources Center, Shanxi Agricultural University. Seven varieties (lines) were tested, with parental Pintianqiao 1 as the control. Each plot had an area of 10 m^2, and three replicates were set. The average plot yield of Pintianqiao 3 in 2019 was 1.87 kg, equivalent to 1 870.5 kg/hm^2, which was 8.8% higher than that of the control, ranking first. The average plot yield in 2020 was 1.73 kg, equivalent to 1 729.5 kg/hm^2, which was 8.1% higher than that of the control, still ranking first. The average plot yield in two years was 1.8 kg, equivalent to 1 800 kg/hm^2, which was 8.4% higher than that of the control.

Regional test

In 2021 – 2022, Pintianqiao 3 participated in the independent joint regional trial of common buckwheat varieties in Maozao of Huairen City, Xihui of Kelan County, Caijiahui of Xing County, Xincheng of Youyu County, Cuijiaping of Heshun County and Dongyang of Yuci District. Two varieties were tested. Randomized block design was adopted, and the plot area was 66 m^2. Jinqiaomai 8 (sweet) served as the control, and two replicates were set. In 2021, the average yield of five test sites (the test site of Caijiahui was abandoned) was 1 432.5 kg/hm^2, which was 7.1% higher than that of the control, ranking first. In the continued test of 2022, the average yield of the five pilot sites was 1 359 kg/hm^2, which was 9.2% higher than that of the control, and still ranked first. The average yield of 10 test positions in two years was 1 396.5 kg/hm^2, which was 8.1% higher than that of the control.

Field investigation

On September 2 – 3, 2022, a field investigation was conducted by Shanxi provincial expert group for identification of non-major crop varieties in Dongyang of Yuci District and Xihui of Kelan County. The experts agreed that Pintianqiao 3, with neat growth, stable genetic traits, stout stems, strong lodging resistance, no pests and diseases found in the field, and extremely large seeds, is suitable for planting in Shanxi sweet buckwheat producing areas.

Seed Reproduction Technique

Field selection

Common buckwheat is a cross-pollinated plant, and it is not only an entomophilous plant, but also an anemophilous plant. Buckwheat also has natural irresistible grain dropping. In order to maintain the specific characters and purity of varieties, it is necessary to adopt reproduction with isolation and crop rotation. Pintianqiao 3 is a tetraploid variety, which is incompatible with diploid common buckwheat varieties. It is advisable to choose fields with flat terrain, where buckwheat (tartary buckwheat and common buckwheat) has not been planted within 2 years, and the previous crops of leguminous crops or potato are appropriate, and other tetraploid varieties of common buckwheat should not be planted around it (within a spacing distance of 4 000 m).

Soil preparation and fertilization

After harvesting the previous crop, timely autumn plowing should be carried out at a depth of 35 to 40 cm, while ensuring consistent depth, uniform and tight ridges, and no missed tillage. The soil is harrowed to conserve moisture when soil is frozen and thawed alternately. Combined with deep ploughing in autumn, 15 000 – 22 500 kg of organic fertilizer or 150 kg of compound fertilizer is applied as base fertilizer every hectare.

Seed treatment

Seed treatment before sowing can improve the air permeability

and water permeability of seed coat, promote seed ripening and increase seed germination rate and germination potential. It can prevent and control underground pests such as mole crickets, cutworms, grubs and buckwheat diseases. It plays a great role in improving seed quality, whole seedlings and strong seedlings and laying a high-yield foundation of buckwheat. At present, buckwheat seeds are mainly treated by drying seeds and dressing seeds.

Drying seeds Buckwheat seeds are spread thinly on dry ground exposed to the sun on sunny days, 7 – 10 d before sowing, from 10:00 to 16:00, for 2 – 3 d in successive. The site must be cleaned up.

Dressing seeds (optional) Seeds are mixed evenly with 20% isofenphos methyl emulsifiable concentrate at an amount of 0.3% – 0.5% of the seed weight or quintozene powder of 0.05% – 0.1% of the seed weight. After standing for 3 – 4 h, the seeds are spread for air-drying.

Sowing

Sowing time The suitable sowing time is mid-May in spring sowing areas and mid-June in summer sowing areas.

Sowing mode and sowing depth Buckwheat is a dicotyledonous plant, and its cotyledons are unearthed, so sowing should not be too deep. It is difficult for buckwheat to emerge from deep sowing, but shallow sowing is prone to air drying. To master the sowing depth, first, soil moisture should be checked. Seeds should be sown slightly shallow with sufficient soil moisture, and slightly deep with deficient soil moisture. Second, soil quality should be checked. Seeds can be sown appropriately deep in sandy soil and dry land, but no deeper than 6 cm, while in clay, they can be sown slightly shallow. Generally, mechanical sowing in line is adopted, with a depth of 3 – 5 cm and a row spacing of about 35 cm, and the soil is suppressed in time after sowing. Attention must be paid to cleaning the sowing machinery before sowing to prevent mechanical mixing.

Seeding rate and density The sowing amount is 45 kg/hm^2, and the planting density is 0.75 – 1.05 million plants/hm^2.

Breaking soil hardening

In case of hardening due to heavy rain before emergence, the ground surface should be loosened with a rake in time to break hardening.

Intertillage weeding

Intertillage has the effects of loosening soil, increasing soil permeability, storing water and preserving moisture, increasing ground temperature and promoting seedling growth, as well as the effects of weeding and increasing fertility. Generally, intertillage weeding is performed once or twice. The first time is carried out at the stage of 4 – 6 leaves, and the second time is carried out before closure of ridges.

Control of diseases and pests

Sheath blight and brown spot disease When diseases occur, 40% chlorothalonil suspension 600 times dilution, 50% carbendazim wettable powder 800 times dilution or 50% procymidone wettable powder 1 000 times dilution is sprayed.

Northern armyworms and beet webworms The larvae are treated with 4.5% beta-cypermethrin emulsifiable concentrate 1 500 times dilution, 600 kg per hectare for spraying, or 450 g of 25% diflubenzuron No. 3 suspensoid in mixture with 600 kg of water for spraying.

Auxiliary pollination (choosing one of the three ways)

Common buckwheat is a cross-pollinated crop, and its seed setting rate is generally low, ranging from 5% to 15%, which limits the increase of yield. A better way to improve the seed setting rate of common buckwheat is to carry out auxiliary pollination.

Bee-assisted pollination In the full bloom stage, bees are put in the field for pollination, and one box of bees is placed every 3 × 666.7 m^2. The hive is close to the edge of corresponding buckwheat field.

Manual pollination In the full bloom stage, during 9:00 – 11:00 am on a sunny day, a cloth strip with a length of 20 – 25 m and a width of 0.5 m is tied to a rope at both ends. And two people hold one end each and gently pull the cloth strip along the top of a common buckwheat field, back and forth once every 3 d, for a total of 3 – 4 times.

UAV-assisted pollination In the full bloom stage, during 9:00 – 11:00 am on a sunny day, a rotary-wing UAV is controlled to fly around a plot back and forth for 2 – 3 times. The flight speed and height should be such that common buckwheat plants start to shake, once every 3 d, for 3 – 4 times in total.

Harvesting

Common buckwheat is a crop with indefinite inflorescence, which lasts for a long time from flowering to maturity, usually 30 – 40 d, and the longest can reach 90 d. Because the duration from flowering to maturity is very long, the maturity of seeds on the plant is very inconsistent. Those that blossom and bear fruit first mature first, and those that blossom and bear fruit later mature later. When 60% – 70% of the seeds in the whole plant mature, there are still many immature seeds and flowers that are still in bloom. If harvesting is carried out until all the seeds are ripe, large and full seeds that ripen first will naturally fall off, which will affect both the yield and the quality of the seeds. In the case of too-early harvesting, a large number of immature seeds will be mixed, which will reduce the quality and yield of buckwheat. According to the experiments, the best harvest time is when 70% of the seeds show the inherent color of the variety. Harvesting at this time, not only is the yield high, but the quality is also good. Therefore, when 70% of the seeds in a seed reproduction field turn dark brown, they can be harvested separately with a combine harvester on sunny days, and dried in the sun separately. The seeds are packaged and stored when the moisture content is not higher than 13.5%. The harvesting machinery, threshing ground and packaging must be cleaned.

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late growth period, thereby ensuring the yield-increasing physiological basis of dry matter accumulation and translocation. If the nitrogen supply exceeds a certain amount, the nitrogen supply will be excessive in the late growth period, resulting in remaining green when it should become yellow and ripe, and fertilizer waste and environmental pollution.

Considering the current market price and cost-saving space, 225 kg/hm^2 of slow-release urea could be used as a suitable fertilization model for summer maize, which has important theoretical and practical guiding significance to saving cost and increasing efficiency in summer maize cultivation in the soil area.

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In particular, if there is strong wind or frost before harvesting, the fruit stalk will wither and become brittle after freezing, and a large number of seeds will fall off. Therefore, the weather forecast before harvesting should be closely monitored, and harvest the seeds in a rush if there is strong wind or frost.

Seed quality inspection

It is required to invite the seed inspection department to reexamine the produced seeds. The seeds of Pintianqiao 3 which meet the standards and have a certificate of conformity are labeled inside and outside the package.

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