# Study on Drought Resistance Caused by Ion Beam-mediated Transformation of Exogenous DNA of Wheat Variant

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**Abstract:** Wheat variation materials with great economic value were obtained by ion beams and they were used for drought resistance relevant research. Through the analysis of 12 variation materials at germination period, the relative germination rate and relative germination energy of variation material 5504, 5606 and 5626 were higher than that of wild materials, which indicated that drought resistance of variation materials at the seeding stage was stronger than wild ones. Several agronomic traits of variation materials such as height, tiller number and 1000-grain weight were analyzed by the means of variance analysis and multiple comparisons on the condition of whole-growth-stage water stress and normal irrigation respectively. The key drought index of variation material 5402 and 5486 were higher than 1, which showed their drought resistance had been improved. Among 780 bands identified by RAPD analysis, 328 ones were polymorphic and the accumulated frequency was 42.05%, therefore, it was concluded that abundant variations, which was induced by the transformation mediated by ion beams, occurred in variation materials at the level of DNA.

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**Key words:** Transformation mediated by ion beam implantation； wheat； drought resistance

## 1. Introduction

Wheat is an important crop of the world (Hischenhuber C, 2006). As an abiotic stress factor, drought limits its production and quality. Carrying out the genetic mechanism of drought resistance of wheat is vital to Chinese food security and sustainable development (Zhaoling Qi, 2010). In order to improve drought resistance of wheat and use water efficiently, self-transgenic approach is one of the important methods to improve the drought resistance of wheat. Transgenic technology is widely used in crop quality improvement. How to get some high resistance crops? It is an issue concerned by geneticists and plant breeders. Some achievements have been made on the transgenic technology and it has the widespread application prospect (Nikals Holmberg et al, 1998 and Qiang Liu et al, 2000).

Transgenic technology mediated by ion beam (Zengliang Yu, 1998) has been widely used in wheat (Zhanglei su, 2009 and Lei Ji, 2005), rice (Qunce Huang, 2007), soybean and microbe breeding (Jun Wei, 2006 and Peirui Li, 2008). This technology can transform target DNA fraction and genome DNA (Shengdong Ji, 2004 and Guowei Huang, 2008). Exogenous DNA is introduced into wheat by ion beam mediated in our laboratory. The donors are hexaploid triticale and oats which have prominent drought resistance characteristic. We get some special variation materials through several years of screening. This paper studies the drought resistance of the variation materials in each growth stage and reveals its variation characteristic. We hope to provide important resource for breeding drought-resistance wheat varieties.

## 2. Materials and methods

### 2.1 Materials

In this paper the materials were wheat variation offspring. The exogenous DNA was introduced into wheat material by $N^+$ mediated. The receptor material is Yuwheat 49. In 2004, the Yuwheat 49's seeds were implanted with 30 keV N ions at the normal temperature. The ion implant dose was $3\times10^{17}$ ions/cm$^2$. The seeds were dipped into oat (concentration of 150μg/ml), hexaploid tritical (concentration of 200μg/ml) s' whole genome DNA about 24 hours after the ion implant. We got the variation materials which name was 5402, 5443, 5486, 5504, 5533, 5563, 5564, 5606, 5626, 5629 and 5647, after several years field observation. As shown in Table 1.

### 2.2 Methods

#### 2.2.1 The PEG stress experiment in germination stage

Each species selected 300 seeds which must be grain plumpness, good uniformity, the same size and no damage. The seeds were placed in the baking oven at 35℃. After about 10 hours, we disinfected the seeds with 0.1% HgCl and washed it with distilled water. Last we divided the seeds into six parts and placed it in glass culture dish.

Seeds were dipped in distilled water and cultured under light for 24 hours. Then we set up comparing experiments which was the PEG Stress experiment and hydroponic culture experiment. Every day 19.2% PEG-6000 solution was added to the petri dish in order to keep the filter paper moist .The hydroponic culture experiment only added to equal amounts distilled water. We were doing three replications respectively.

Table 1. Variation of material information

| number | Receptor seeds | Donor genomic DNA source | Transformation time(years) |
|---|---|---|---|
| 5402 | yuwheat49（5401） | oat（5024） | 2004 |
| 5443 | yuwheat49 | oat | 2004 |
| 5486 | yuwheat 49 | oat | 2004 |
| 5504 | yuwheat 49 | oat | 2004 |
| 5533 | yuwheat 49 | oat | 2004 |
| 5563 | yuwheat 49 | oat | 2004 |
| 5564 | yuwheat 49 | oat | 2004 |
| 5606 | yuwheat 49 | oat | 2004 |
| 5626 | yuwheat 49 | oat | 2004 |
| 5629 | yuwheat 49 | oat | 2004 |
| 5647 | yuwheat 49 | hexaploid tritical（5037） | 2004 |

**2.2.2 The field management methods of whole growth period**

In 2009-2010, we did the drought stress experiments with the automatic folding drought shed in Luoyang Academy of Agricultural Sciences. The length of test cell is 2 m, the width is 1m, 4-line area. The drought stress was set up in the shed .The supplementary irrigation treatment in adjacent land of the Shed. By using randomized-block design of two factors, we were doing twice repeat respectively. Before the next wheat sowing, by moving the dry shed, we controlled the natural precipitation of experiment field, in order to make the 0-150cm soil water storage capacity of about 150mm. Through controls of the different irrigation treatment during the whole growth stages of wheat, the field contrast experiment is carried out. On the total condition change of this experiments, all soil nutrient content, soil texture and soil thickness are consistent with the dry shed except moisture content of the soil.

**2.3 The survey on relevant index of variation materials**

**2.3.1 The study of indicators of germination period**

The number of seeds which germinated was recorded in the first 4 days and 7 days. In the seventh day, 15 Germinating seeds were randomly selected to evaluate the length of coleoptiles, the length of main radical and bud length for each treatment, take the average.

**2.3.2 The study for index of whole growth period**

10 single plants were randomly selected in each strain to test the agronomic traits, including plant height, tiller number, fertile spikelet number, sterile spikelet number, panicle length, panicle number and 1000-grain weight, during the harvesting time.

**2.4 Data processing methods**

**2.4.1Data processing of germination period**

Relative germination rate：

$$RGer = Ger_T \cdot Ger_{ck}^{-1} \cdot 100\% \quad \text{…………（1.1）}$$

Relative germination energy：

$$RGerE = Ger_{T1} \cdot Ger_{ck1}^{-1} \cdot 100\% \quad \text{………..（1.2）}$$

In the above formula（1.1）and (1.2) :

$Ger_{ck}$ --- the germination rate of control materials；

$Ger_{ck1}$ --- the germination energy of control materials；

$Ger_T$ --- the germination rate by PEG-6000 stress cultivation；

$Ger_{T1}$ --- the germination energy by PEG-6000 stress cultivation；

The formula of germinating energy：

$$Ger_{T1} = \overline{X}_{GerE.T} \cdot \overline{X}_{TS.T}^{-1} \cdot 100\% \quad \text{................（1.3）}$$

$$Ger_{ck1} = \overline{X}_{GerE.CK} \cdot \overline{X}_{TS.CK}^{-1} \cdot 100\% \quad \text{……..（1.4）}$$

In the above formula（1.3）and (1.4) :

$Ger_{T1}$ --- the germination energy by PEG-6000 stress cultivation；

$\overline{X}_{GerE.T}$ --- in 96 hours, the average of triplicate germinated seeds under PEG-6000 stress cultivation；

$\overline{X}_{TS.T}$ --- the average of triplicate seeds under PEG-6000 stress cultivation；

$Ger_{ck1}$ --- the germination energy of control materials；

$\overline{X}_{GerE.CK}$ --- the average of triplicate germinated seeds were cultivated for 96 hours by normally;

$\overline{X}_{TS.CK}$ --- the average of triplicate seeds were cultivated by normally;

The formula of Germinating rate:

$$Ger_T = \overline{X}_{Ger.T} \cdot \overline{X}_{TS.T}^{-1} \cdot 100\% \quad (1.5)$$

$$Ger_{ck} = \overline{X}_{Ger.CK} \cdot \overline{X}_{TS.CK}^{-1} \cdot 100\% \quad (1.6)$$

In the above formula (1.5) and (1.6) :

$Ger_T$ --- the germination rate by PEG-6000 stress cultivation;

$\overline{X}_{Ger.T}$ --- in 168 hours, the average of triplicate germinated seeds under PEG-6000 stress cultivation;

$\overline{X}_{TS.T}$ --- the average rage of triplicate seeds were cultivated under PEG-6000stress;

$Ger_{ck}$ --- the germination rate of control materials;

$\overline{X}_{Ger.CK}$ --- the average of triplicate germinated seeds were cultivated for 168 hours by normally;

$\overline{X}_{TS.CK}$ --- the average of triplicate seeds were cultivated by normally;

The study results of agronomic traits were analyzed by SPSS 17.0.

#### 2.4.2 Data processing for agronomic trait of whole growth period

According to the drought index was proposed by Mr. Li Xian-bin, we had revised the drought resistance coefficient and drought resistance index. The following formula:

$$\text{Drought coefficient} = \frac{\text{characteristics of material in shed}}{\text{characteristics of material out of shed}} \quad (1.7)$$

$$\text{Index of drought resistance} = \frac{\text{characteristics of material in shed}}{\text{characteristics of control breed in shed}} \times \text{drought coefficient / drought coefficient of control breed} \quad (1.8)$$

### 2.5 RAPD basic reaction system

RAPD basic reaction system was 25 μL, we could reference to the literature of the Junwei Wang (2004) and Changhong Guo (1999). The RAPD primer was provided by the sangon biotech company.

| Material | | volume |
|---|---|---|
| 10×PCR Buffer | | 2.5μL |
| dNTP Mixture | | 2μL |
| RAPD primer | | 1.25μL |
| DNA template | | 1μL |
| rTaq-DNA (5U/μL) | | 0.25μL |
| sterile water | | 18μL |
| Total volume | | 25μL |

The reaction mixture was mixed in the low speed by centrifuge, and then it was amplified in the PCR machine.

RAPD amplification procedure:

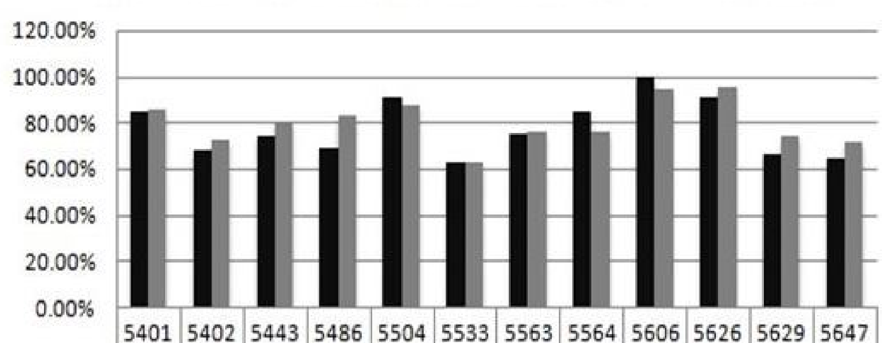
(1)94℃ - 2min (2) 94℃-15s
(3)39℃ - 1min (4)72℃ - 30s
(5) 72℃ - 10min (6)4℃ - 30min

The RAPD was amplified for 45 cycles. The product was store in 4 ℃.

## 3 Results and Analysis

### 3.1 Relative germination potential and rate of the variation materials in the germination stage

As could be seen from Figure 1, the No. 5401 belonged to the control material, that was Yu wheat 49 (Wen 6), the other numbers were the variant materials. It showed that the relative germination energy and relative germination rate of the variant materials 5504, 5605 and 5625 were higher than the control material. And the relative germination energy and relative germination rate of the variant material 5606 had already reached 100% and 94.3% respectively.




| | 5401 | 5402 | 5443 | 5486 | 5504 | 5533 | 5563 | 5564 | 5606 | 5626 | 5629 | 5647 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Relative germination energy | 85.10 | 67.80 | 74.50 | 68.70 | 91.20 | 63.00 | 75.40 | 84.40 | 100.0 | 91.10 | 66.30 | 64.90 |
| Relative germination rate | 85.60 | 72.40 | 80.60 | 83.30 | 87.40 | 62.80 | 76.10 | 76.00 | 94.30 | 95.10 | 73.90 | 71.60 |

Figure 1. Data analysis of relative germination energy and germination rate

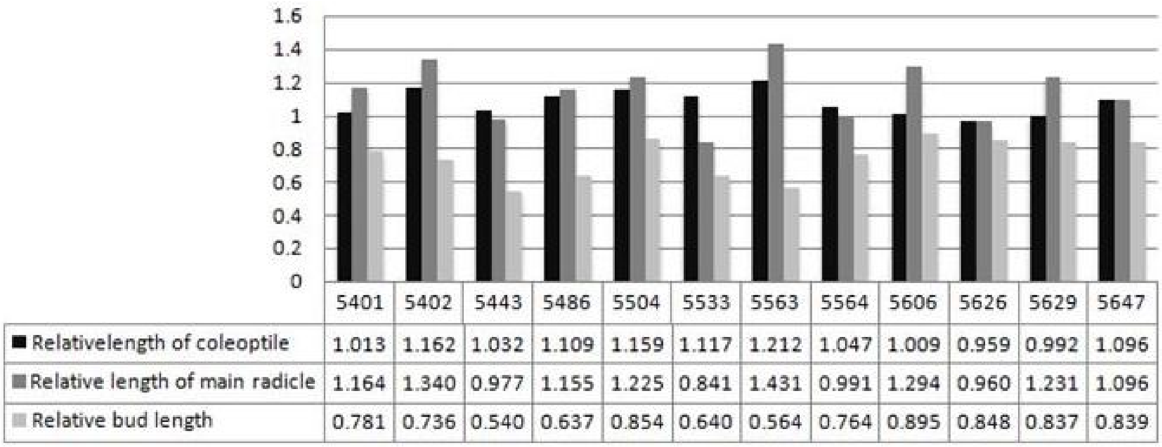



Figure 2. Analysis in main agronomic trait's date of variation materials in the germination stage

**3.2 Analysis main agronomic traits of variation materials in the germination stage**

As the diagram in Figure 2 illustrates, the relative coleoptile length = coleoptile length under PEG stress / coleoptile length of hydroponics, the relative main radicle length, the relative shoot length and the relative coleoptile length had the same calculation method.

The data in Figure 2 showed that the variant materials 5606, 5626 and 5629 were lower than the control material in the coleoptile length, but other variant materials were higher than the control material. The relative coleoptile length of the variant materials 5626 and 5629 were less than 1, the other variant materials were greater than 1, which indicated that the coleoptile length index of the variant material 5626 and 5629 were more sensitive to moisture, while the coleoptile length index of other variant materials were not sensitive to moisture.

In the main radicle length, the relative value of the variant materials 5443, 5486, 5533, 5564, 5626, 5647 were lower than the control material. The relative main radicle length of variant materials 5443, 5533, 5564 and 5626 were less than 1, which indicated that these variation materials were sensitive to moisture.

The variant materials 5443, 5486, 5533 and 5563 were lower than the contrast material in the bud length, other variant materials were higher than the contrast material on relative value. At the same time, the bud length of all the variant materials were less than 1, which indicated that the shoot length index of all variant materials were more sensitive to moisture.

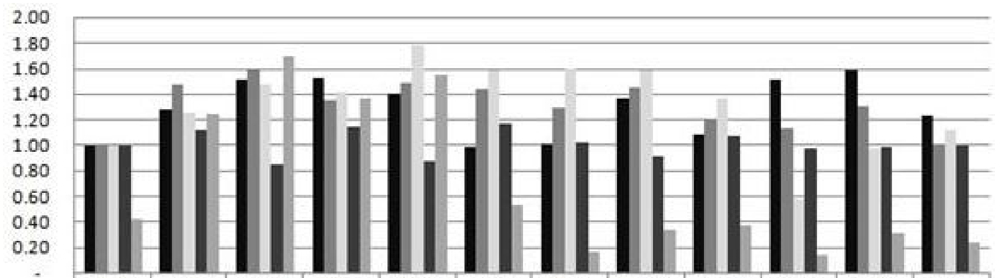
Table 2. Remarkable analysis in agronomic traits of variation materials under water stress during whole-growth-stage (Mean±SD)

| number | Plant Height（cm） | Tiller number | Fertile spikelet number | Sterile spikelet number | panicle lengt（cm） | Panicle Number | 1000-grain-weigt（g） |
|---|---|---|---|---|---|---|---|
| 5401 | 58.75±3.69 | 3.90±0.88 | 14.20±1.87 | 4.80±1.48 | 7.89±0.76 | 29.20±4.02 | 48.00±1.00 |
| 5402 | 71.35±2.87** | 4.60±0.97 | 16.30±2.50* | 5.40±0.97 | 7.72±0.42 | 37.10±4.41** | 52.00±1.00** |
| 5443 | 70.25±2.78** | 5.00±1.05* | 16.30±2.26* | 5.80±0.92 | 7.48±0.56 | 37.30±6.93** | 45.33±0.58* |
| 5486 | 70.53±2.72** | 5.20±1.03** | 16.20±1.40* | 4.00±0.82 | 7.42±0.75 | 31.20±2.90 | 51.00±1.00* |
| 5504 | 68.90±2.77** | 4.50±0.85 | 14.90±4.15 | 6.20±0.79* | 7.20±0.48* | 30.80±6.76 | 45.00±1.00* |
| 5533 | 62.75±3.55* | 3.80±0.42 | 16.20±3.05 | 5.80±0.92 | 8.14±0.61 | 33.40±5.17 | 55.00±1.00** |
| 5563 | 76.95±7.30** | 3.50±1.78 | 14.60±4.09 | 5.20±1.87 | 9.65±1.02** | 32.30±1.23 | 49.00±1.00 |
| 5564 | 53.05±6.10* | 3.80±1.23 | 16.10±2.56 | 4.60±1.35 | 9.07±0.79** | 38.10±6.45** | 46.00±1.00 |
| 5606 | 67.43±3.22** | 4.40±0.70 | 14.00±1.41 | 3.60±0.52* | 7.40±0.63 | 30.00±4.40 | 50.00±1.00 |
| 5626 | 54.31±8.97 | 6.20±1.40** | 14.20±3.39 | 4.50±1.51 | 7.01±1.67 | 14.40±2.01** | 47.00±1.00 |
| 5629 | 60.70±3.38 | 5.00±0.82** | 15.30±1.57 | 4.50±0.85 | 7.38±0.75 | 28.60±2.80 | 45.33±0.58** |
| 5647 | 60.80±8.72 | 4.40±1.51 | 16.60±3.50 | 4.70±2.71 | 8.31±1.51 | 34.00±1.19 | 47.00±1.00 |

Note: The different * and ** means significant at 5% and 1% levels, respectively

**3.3 Analysis agronomic traits of the variation materials under water stress in the whole period**

Table 2 showed that the plant height of the variant material 5564 was much lower than the contrast material 5401, however, the plant height of 5402, 5443, 5486, 5504, 5563 and 5606 were significantly higher than the contrast material. The tiller number of the variant materials 5486, 5626 and 5629 were significantly higher than the contrast material. The fertile spikelet number of the variant materials 5402, 5443 and 5486 were much higher than the contrast material. There was a significant correlation between the variant materials 5606, 5504 and the contrast material in the sterile spikelet number. The panicle length of 5504 was much shorter than the contrast material, but the variant materials 5563 and 5564 were much higher. The panicle number of the variant materials 5402, 5443, 5626 and 5564 were extremely significantly differences than the contrast material. The 1000-grain weights of the variant materials 5402, 5533, 5629 were significantly differences than the contrast material.




| | 5401 | 5402 | 5443 | 5486 | 5504 | 5533 | 5563 | 5564 | 5606 | 5626 | 5629 | 5647 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| tiller number | 1.00 | 1.28 | 1.51 | 1.52 | 1.40 | 0.99 | 1.01 | 1.36 | 1.08 | 1.51 | 1.58 | 1.23 |
| fertile spikelet number | 1.00 | 1.47 | 1.60 | 1.35 | 1.49 | 1.44 | 1.29 | 1.45 | 1.19 | 1.14 | 1.30 | 1.00 |
| panicle Number | 1.00 | 1.26 | 1.47 | 1.41 | 1.78 | 1.59 | 1.60 | 1.59 | 1.36 | 0.59 | 0.97 | 1.12 |
| 1000-grain weight | 1.00 | 1.12 | 0.85 | 1.15 | 0.88 | 1.17 | 1.03 | 0.91 | 1.07 | 0.98 | 0.99 | 1.00 |
| yield | 0.431 | 1.245 | 1.695 | 1.360 | 1.551 | 0.534 | 0.170 | 0.343 | 0.381 | 0.147 | 0.314 | 0.238 |

Figure 3. Analysis in drought resistance factor of main agronomic trait factors during whole-growth-stage

**3.4 Analysis the agronomic traits of variation materials in whole-growth-stage**

According to analysis index of drought resistance of the tiller number in Figure 3, we could know that all the indexes of drought resistance of the variant materials were close to 1, which indicated that the variant materials had a better drought resistance in seedlings period. Among them, the index of drought resistance of the variant material 5629 reached to 1.58, which indicated that this material had the best drought resistance in seedlings period.

According to analysis the index of drought resistance of fertile spikelet number, we could know that all the indexes of drought resistance of variant materials were more than 1, which indicated that the variant materials had a better drought resistance in the beginning period. Among them, the index of drought resistance of the variant material 5443 reached up to 1.60, which indicated that this material had the best drought resistance in seedlings period.

According to analysis index of drought resistance of panicle number, we could know that the index of drought resistance of the variant material 5626 was less than 1 and the index of drought resistance of the other variant materials were close to 1, which indicated that the other variant materials had a better drought resistance except 5626 in jointing stage.

According analysis index of drought resistance of 1000-grain weight, we could know that the indexes of drought resistance of the variant material 5443 and 5504 were less than 1 and the indexes of drought resistance of the other variant materials were close to 1, which indicated that the other variant materials had a better drought resistance except 5443 and 5504 in filling stage.

According analytical index of drought resistance of the outputs, we could know that the indexes of drought resistance of the variant materials 5402，5443，5486 and 5504 were all more than 1, which indicated that the outputs of these four materials were larger than the other materials.

**3.5 Analysis RAPD bands of variation hexaploid triticale offspring**

The variation material (5647)'s RAPD bands were Analyzed in Figure 4 and Figure 5. Each of the three bands were divided into a group. The first lane was the donor material 5037, the second lane was the control material 5401, the third lane was the variation material 5647. "➚ " was the variation bands of the variation material.

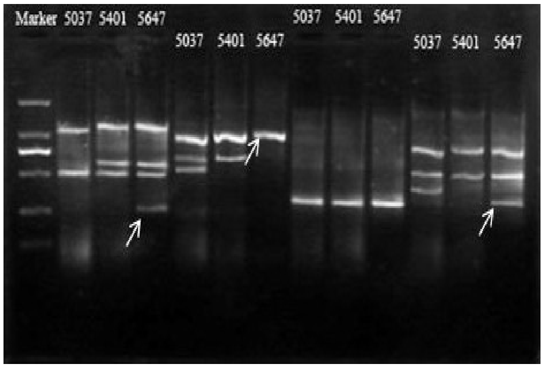



Figure4.Reaction RAPD bands of Primer s17, s2114,s2036,s2046

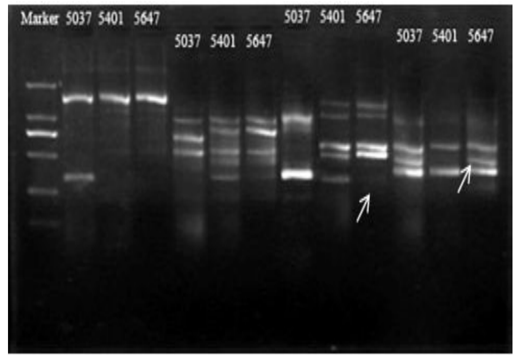



Figure5.Reaction RAPD bands of Primer s88, s2075, s11, s2020

Table 3. Analysis RAPD bands of variation hexaploid triticale offspring

| Primer | 5037 | 5401 | 5647 | increased bands | reduced bands |
|---|---|---|---|---|---|
| S2 | 4 | 3 | 4 | 1 | 0 |
| S3 | 2 | 4 | 4 | 1 | 1 |
| S11 | 2 | 5 | 4 | 0 | 1 |
| S17 | 2 | 3 | 4 | 1 | 0 |
| S28 | 0 | 3 | 2 | 2 | 0 |
| S86 | 5 | 5 | 4 | 0 | 1 |
| S128 | 3 | 4 | 3 | 2 | 0 |
| S130 | 2 | 2 | 1 | 0 | 1 |
| S490 | 0 | 6 | 4 | 0 | 2 |
| S498 | 2 | 3 | 2 | 0 | 1 |
| S2011 | 3 | 4 | 3 | 0 | 1 |
| S2013 | 1 | 2 | 1 | 0 | 1 |
| S2019 | 3 | 3 | 2 | 0 | 1 |
| S2020 | 3 | 2 | 3 | 1 | 0 |
| S2038 | 2 | 3 | 2 | 0 | 1 |
| S2041 | 3 | 0 | 1 | 1 | 0 |
| S2046 | 3 | 2 | 3 | 1 | 0 |
| S2047 | 0 | 2 | 3 | 1 | 0 |
| S2053 | 3 | 2 | 2 | 0 | 0 |
| S2054 | 4 | 2 | 2 | 1 | 0 |
| S2056 | 1 | 1 | 2 | 2 | 0 |
| S2070 | 3 | 3 | 2 | 1 | 1 |
| S2075 | 3 | 5 | 3 | 0 | 2 |
| S2077 | 2 | 2 | 2 | 0 | 1 |
| S2078 | 2 | 4 | 4 | 1 | 0 |
| S2086 | 2 | 1 | 2 | 1 | 0 |
| S2096 | 4 | 4 | 4 | 0 | 0 |
| S2098 | 3 | 2 | 1 | 0 | 1 |
| S2109 | 4 | 2 | 3 | 1 | 0 |
| S2114 | 3 | 2 | 1 | 0 | 1 |
| S2115 | 3 | 4 | 3 | 0 | 1 |
| Other Primer | 46 | 52 | 43 | 0 | 0 |
| Total bands | 123 | 142 | 124 | 18 | 18 |
| Variation rate(%)[b] | 25.35 | | | | |

The hexaploid triticale 5037 was amplified a total of 123 bands in Table 3，the control material 5401 was amplified 142 bands, the variation material 5647 was amplified 124 bands. The variation bands were 36, and the variation rate was 25.35%.

**3.6 Analysis RAPD bands of variation oat offspring**

The variation oat offspring's RAPD bands were indicated in Figure 6 and Figure 7. Table 4 showed that the donor oats 5024 was amplified 55 bands, the receptor 5401 was amplified 66 bands, and 11 variation materials were amplified a total of 656 bands. 292 of them showed polymorphism, accounting for 44.51% of the total number of bands. However, variation rates of variation materials 5504 and 5533 reached the lowest level, which were 27.27%; variation rates of variation material 5663 reached the highest level, which were 74.24%.

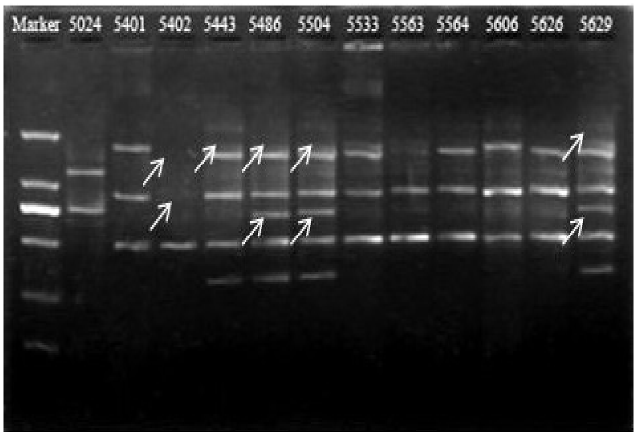



Figure6.Reaction RAPD bands of Primer s2019

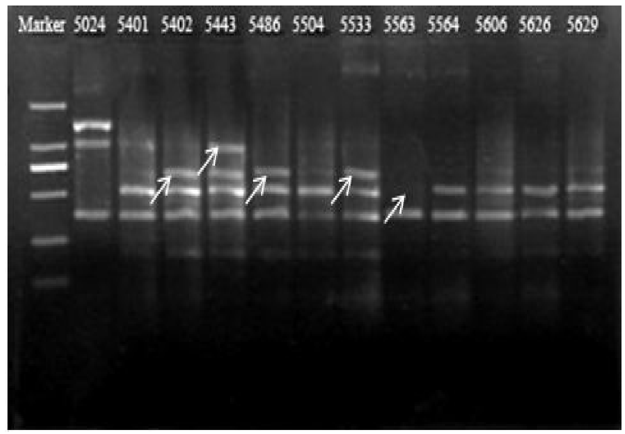



Figure7.Reaction RAPD bands of Primer s2070

Table 4. **A**nalysis RAPD bands of variation oat offspring

| Primer | 5024 | 5401 | 5402 | 5443 | 5486 | 5504 | 5533 | 5563 | 5564 | 5606 | 5626 | 5629 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| S3 | 1 | 4 | 5(1,0) | 5(1,0) | 5(1,0) | 6(1,0) | 0(2,0) | 0(0,4) | 0(0,4) | 3(0,2) | 7(3,0) | 6(2,0) |
| S9 | 3 | 3 | 3(0,0) | 4(1,0) | 4(1,0) | 4(1,0) | 3(0,0) | 2(0,1) | 3(1,1) | 2(0,1) | 3(1,1) | 4(1,0) |
| S11 | 0 | 1 | 1(0,0) | 2(1,0) | 2(1,0) | 0(0,0) | 2(1,0) | 0(0,1) | 0(0,1) | 2(1,0) | 2(1,0) | 4(3,0) |
| S17 | 0 | 1 | 1(0,0) | 1(0,0) | 1(0,0) | 1(0,0) | 1(0,0) | 0(0,1) | 0(0,1) | 1(0,0) | 0(0,1) | 1(0,0) |
| S28 | 0 | 2 | 1(0,1) | 1(0,1) | 1(0,1) | 1(0,1) | 2(0,0) | 1(0,1) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) |
| S39 | 2 | 4 | 4(0,0) | 3(0,1) | 1(0,3) | 3(0,1) | 3(0,1) | 0(0,4) | 2(0,2) | 4(0,0) | 3(0,1) | 3(0,1) |
| S88 | 4 | 2 | 0(0,2) | 1(0,1) | 0(0,2) | 2(1,1) | 2(1,1) | 2(1,1) | 2(1,1) | 4(2,1) | 2(1,1) | 2(1,1) |
| S130 | 0 | 1 | 2(1,0) | 1(0,0) | 2(1,0) | 2(1,0) | 0(0,1) | 2(1,0) | 2(1,0) | 2(1,0) | 0(0,1) | 2(1,0) |
| S133 | 2 | 4 | 5(2,1) | 6(2,0) | 5(2,0) | 6(2,0) | 6(2,0) | 4(2,1) | 3(2,2) | 6(3,1) | 3(1,2) | 4(1,0) |
| S180 | 2 | 1 | 1(0,0) | 1(0,0) | 1(0,0) | 1(0,0) | 1(0,0) | 3(2,0) | 3(2,0) | 2(1,0) | 3(2,1) | 2(1,0) |
| S485 | 2 | 4 | 4(0,0) | 2(0,2) | 5(1,0) | 5(1,0) | 4(0,0) | 1(0,3) | 4(0,0) | 0(0,4) | 5(1,0) | 5(1,0) |
| S490 | 2 | 6 | 6(0,0) | 7(1,0) | 7(1,0) | 6(0,0) | 8(2,0) | 6(1,1) | 6(1,1) | 8(2,0) | 7(1,0) | 4(1,2) |
| S495 | 4 | 2 | 2(0,0) | 4(2,0) | 2(0,0) | 1(0,1) | 2(0,0) | 2(0,0) | 2(0,0) | 3(1,0) | 2(0,0) | 3(1,0) |
| S498 | 3 | 1 | 2(1,0) | 1(1,1) | 1(1,1) | 1(1,1) | 2(2,1) | 1(1,1) | 2(1,0) | 1(1,1) | 2(1,0) | 2(1,0) |
| S2011 | 2 | 4 | 3(0,1) | 3(0,1) | 4(0,0) | 4(0,0) | 4(0,0) | 2(0,2) | 3(0,1) | 3(0,1) | 4(0,0) | 3(0,1) |
| S2013 | 1 | 2 | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(2,2) | 3(2,1) | 4(2,0) | 2(0,0) | 2(0,0) |
| S2019 | 2 | 3 | 1(0,2) | 4(1,0) | 5(2,0) | 5(2,0) | 3(0,0) | 2(0,1) | 3(0,0) | 3(0,0) | 3(0,0) | 5(2,0) |
| S2020 | 2 | 2 | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 1(0,1) | 2(1,1) | 1(0,1) |
| S2037 | 3 | 3 | 3(0,0) | 3(0,0) | 3(0,0) | 3(0,0) | 3(0,0) | 4(2,1) | 4(2,1) | 4(2,1) | 2(0,1) | 3(0,0) |
| S2047 | 1 | 3 | 2(2,3) | 3(2,2) | 0(0,3) | 2(0,1) | 2(0,1) | 2(0,1) | 2(0,1) | 4(2,1) | 2(0,1) | 1(0,2) |
| S2053 | 3 | 2 | 0(0,2) | 3(1,0) | 2(0,0) | 2(0,0) | 2(0,0) | 1(0,1) | 2(0,0) | 2(0,0) | 3(1,0) | 2(0,0) |
| S2058 | 4 | 2 | 2(1,1) | 1(0,1) | 1(0,1) | 2(0,0) | 2(1,1) | 3(2,1) | 3(2,1) | 2(1,1) | 2(1,1) | 1(0,1) |
| S2070 | 3 | 2 | 3(0,1) | 4(2,0) | 3(1,0) | 2(0,0) | 3(1,0) | 1(0,1) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) |
| S2086 | 3 | 2 | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 4(2,0) | 2(0,0) | 1(0,1) | 1(0,1) | 2(0,0) |
| S2098 | 3 | 2 | 0(0,2) | 2(0,0) | 2(0,0) | 0(0,2) | 2(0,0) | 2(0,0) | 2(0,0) | 2(0,0) | 1(0,1) | 2(0,0) |
| S2115 | 3 | 3 | 3(1,1) | 0(0,3) | 2(0,1) | 3(0,0) | 3(0,0) | 1(1,3) | 2(0,1) | 2(1,2) | 2(1,2) | 3(0,0) |
| Variation bands | | | (9,17) | (15,13) | (12,12) | (10,8) | (12,6) | (17,32) | (15,19) | (20,18) | (16,16) | (16,9) |
| Total bands | 55 | 66 | 60 | 68 | 65 | 68 | 66 | 50 | 61 | 70 | 77 | 71 |
| Variation rate (%)[c] | | | 39.39 | 42.42 | 36.36 | 27.27 | 27.27 | 74.24 | 51.51 | 57.57 | 48.48 | 37.89 |

c: Variation rate = Variation bands /total bands， in the parentheses, the first was the increased band, the second was the decreased band.

**4 Discussions**

In this paper, we did some experimental investigation and data analysis on the main agronomic traits and drought resistance of the variation material in germination period and growth period. And the offspring of variation wheat, which was transformed by ion beam, was analyzed by RAPD method.

First of all, we can simulate the drought environment by using PEG-6000 Stress in order to check the drought resistance in germination stage, whose results show that relative germination rate and the relative germination energy of the variation materials 5504, 5606 and 5626 are higher than control materials. The coleoptiles of the variation materials 5626 and 5629 are longer, the main radicle indices of the variation materials 5533, 5564 5443 and 5626 are sensitive to moisture variation, and all materials are more sensitive to moisture. Among them, all the agronomic indicators of the variation material 5626 are sensitive to moisture on germination period.

Then, through the investigation of the plant height, tiller number, fertile spikelet number, sterile spikelet number, grain number per ear, weight and other agronomic characters of the variation materials in different water stress and water condition, it can be found that some agronomic traits of the variation material and the contrast material are different (Table 2). At the same time, each agronomic trait of variation materials are analyzed by the analysis of variance and multiple comparisons, it can be known that the agronomic traits of variation materials are different (Table 2).

Finally, through the drought resistance index analysis of the main agronomic traits in the whole growth period, we can get the result that the main drought resistance index of the variation materials 5402 and 5486 are greater than 1, indicating that the overall drought resistance of the material has been greatly improved. We analyze the coefficient of variation on each index. The results show that the drought resistances of some varieties have been significantly improved in some important periods such as germination period and wheat growth period. This can provide the reference for the further study.

At the same time the variation material are analyzed by RAPD. In total of 780 bands are identified, which 328 bands are polymorphic, it have an accumulated frequency of 42.05%, showing that abundant variations occurred in the level of DNA of variation materials. We conclude that the ion beam-mediated transformation is an effective method in wheat breeding.

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