Breeding and characterization of the high cadmium-accumulating rice line ‘Akita 119’

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Cadmium (Cd) is a toxic heavy metal that is mainly accumulated through the consumption of foods produced in Cd-contaminated fields. Phytoremediation is one of the most effective methods to reduce the soil Cd concentration. In this study, we bred a new rice line, ‘Akita 119’, for Cd phytoremediation. ‘Akita 119’ was obtained by a soft X-ray mutation of ‘Cho-ko-koku’, a naturally high-Cd-accumulating rice cultivar. The heading date of ‘Akita 119’ was about 2 weeks later than that of ‘Akitakomachi’, which is the leading cultivar in Akita Prefecture, Japan. ‘Akita 119’ has a short culm length and many panicles. The shattering resistance and lodging resistance of ‘Akita 119’ were improved compared to ‘Cho-ko-koku’. The thousand-grain weight of ‘Akita 119’ was much smaller than that of ‘Akitakomachi’, and grains of ‘Akita 119’ could be easily distinguished from general japonica cultivars. When ‘Akita 119’ was grown in Cd-contaminated fields, the shoot dry weight and Cd concentration were similar to those of ‘Cho-ko-koku’. These results demonstrate that ‘Akita 119’ has improved agronomic characteristics compared to ‘Cho-ko-koku’ while retaining the ability to extract Cd. Therefore, it should be considered a promising candidate for Cd phytoremediation in paddy fields in northern parts of Japan.

Key Words: ‘Akita 119’, cadmium, phytoremediation, rice breeding, rice line.

Introduction

Cadmium (Cd) is a heavy metal that is toxic for humans and causes various health problems (Horiguchi et al. 2013, Trzcinka-Ochocka et al. 2010). Cadmium accumulates in the human body following the consumption of contaminated foods. A Cd limit in some agricultural products was established by the Codex Alimentarius Commission to provide food safety and human health (CODEX 2006). Some agricultural products that are harvested from Cd-contaminated fields exceed the Cd limit value, which is becoming a serious human health problem. In Japan, Cd limits are exceeded on 2.7 km² of farmland, requiring remediation measures (Ministry of the Environment 2017). Techniques to reduce Cd concentrations in soil are therefore urgently needed.

Phytoremediation, which is the removal of soil contaminants using plants, is one of the most effective methods to reduce soil Cd concentrations. Rice has been used effectively for Cd phytoremediation of paddy fields due to its large biomass yield and well-established cultivation and harvest methods (Ishikawa et al. 2006). Various Cd levels have been measured in the shoots and grains of different rice cultivars (Arao and Ae 2003, Ishikawa et al. 2005, Takahashi et al. 2011, Uraguchi et al. 2009). ‘Cho-ko-koku’, ‘Jarjan’, and ‘Anjana Dhan’ accumulate significantly higher levels of Cd in their shoots and grains than do other cultivars grown worldwide (Uraguchi et al. 2009). After 2–4 years’ cultivation of ‘Cho-ko-koku’, the Cd concentration in the soil and grains of subsequent cultivars were reported to be lower than the levels in a control field (Ibaraki et al. 2009, 2014, Murakami et al. 2009). These results indicate that high-Cd-accumulating rice cultivars are effective for Cd phytoremediation.

Heavy metal ATPase (HMA) is a metal transporter, and some of the HMA in rice can transport Cd (Takahashi et al. 2012, Uraguchi and Fujiwara 2012, 2013). OsHMA3 functions to enable the sequestration of Cd into vacuoles in the root cells (Miyadate et al. 2011, Ueno et al. 2010). The responsive gene of quantitative trait loci (QTL) for the Cd concentration in the shoots of ‘Cho-ko-koku’, ‘Jarjan’, and ‘Anjana Dhan’ was identified as OsHMA3 (Ueno et al. 2010, 2011, Tezuka et al. 2010). OsHMA3 from these cultivars cannot transport Cd into vacuoles, resulting in the direct translocation of absorbed Cd from roots to shoots.
(Ishikawa et al. 2011). Backcross inbred lines (BILs) containing the QTL for the Cd concentration from the ‘Jarjan’ allele reduced the Cd concentration in the soil and the shoots of subsequently grown rice (Abe et al. 2011). The loss of function of OsHMA3 causes high Cd accumulation in the shoots and grains (Yan et al. 2016). This indicates that OsHMA3 plays an important role in Cd accumulation in the shoots of rice.

OsNRAMP5 is a metal transporter involved in Cd uptake by roots (Ishimaru et al. 2012, Sasaki et al. 2012). OsNRAMP5 knockdown plants have been reported to accumulate higher levels of Cd in shoots than wild-type plants due to enhanced Cd translocation to shoots; this process is associated with the expression of genes related to metal transport, such as OsNRAMP1 and OsIRT2 (Ishimaru et al. 2012). Higher Cd accumulation levels have been observed in the OsNRAMP5 knockdown mutant of ‘Anjana Dhan’ than in wild-type ‘Anjana Dhan’, even in a Cd-contaminated field (Takahashi et al. 2014). However, high Cd-accumulating rice cultivars such as ‘Cho-ko-koku’, ‘Jarjan’, and ‘Anjana Dhan’ are difficult to cultivate because they tend to shatter and are easily lodged.

Abe et al. (2013, 2017) produced the rice line ‘MJ3’, ‘MA22’, and ‘TJTT8’ for Cd phytoremediation. ‘MJ3’ and ‘MA22’ were obtained by a gamma ray mutation of ‘Jarjan’ and ‘Anjana Dhan’, respectively. ‘TJTT8’ was obtained by backcrosses of ‘Tachisugata’, a rice variety used as a livestock feed after it is crossed with ‘Jarjan’. ‘MJ3’ and ‘TJTT8’ accumulate high levels of Cd, with both varieties having a non-shattering habit and lodging resistance (Abe et al. 2013, 2017). However, the heading and maturing day of ‘MJ3’ are too late for it to be grown in Akita Prefecture, and it is difficult to obtain ‘TJTT8’ seeds due to sterility. Therefore, it is difficult to cultivate ‘MJ3’ and ‘TJTT8’ in northern regions of Japan. Previously, we released ‘Akita 110’ for Cd phytoremediation in northern regions of Japan (Takahashi et al. 2016). However, Cd accumulation in ‘Akita 110’ was lower than that in ‘Cho-ko-koku’ depending on field conditions. To improve the stability of Cd accumulation in the shoots, we produced a new rice line, ‘Akita 119’, for Cd phytoremediation.

Materials and Methods

Plant growth conditions

Seeds of ‘Akita 119’, ‘Cho-ko-koku’, ‘Akitakomachi’, and ‘Akita 110’ were germinated and grown as described previously (Takahashi et al. 2016). ‘Akitakomachi’ is a general japonica cultivar and is the leading variety grown in Akita Prefecture, Japan. Seedlings were grown in plastic greenhouses in Akita Prefectural Agricultural Experiment Station, Akita Prefecture, Japan from mid-April to May. For line selection, seedlings were transplanted one by one into planting lines in a paddy field at Akita Prefectural Agricultural Experiment Station. For DNA marker selection, a technique reported previously was applied (Miyadate et al. 2011). To determine agronomic traits, yield, and Cd extraction, four seedlings were transplanted as one hill. About 120 hills were planted per experimental plot, with two replicates. In each experiment, seedlings were transplanted into 22 × 22-cm areas, with a planting density of 20.7 hills m⁻². Experimental plots were established in Cd-contaminated paddy fields located in Akita Prefecture. The soil type of the paddy fields was classified as gray lowland soil, and the soil pH was around 5.5. The amount of base dressing fertilizer applied and the soil Cd concentration (before planting, extraction with 0.1 M HCl) of each experimental field are indicated in Table 1. No additional fertilizer was supplied. Flood irrigation was conducted until the heading stage. When the plants entered the heading stage, irrigation was stopped, and drainage was maintained until harvest.

Yield and agronomic trait tests

Yield and agronomic trait tests were performed based on the Rice Cultivation Guidebook for Akita Prefecture (Akita Prefecture 2010). For yield tests, harvesting was performed on 64 hills in two experimental plots for each cultivar. Culm length, panicle length, and panicle number were measured at the maturing stage for 10 hills per experimental plot. Shoot dry weight, unhulled rice grain weight, and thousand-grain weight were measured as described previously (Takahashi et al. 2016). Heading date and lodging were estimated from observations of experimental plots within the growing fields. Days to heading was calculated as the period from transplanting day to heading day. Lodging was evaluated on an ordinal scale from 0 (not lodged) to 5 (completely lodged). The shattering habit was evaluated by shedding rate after grasping panicles in the maturing stage.

Measurement of Cd concentration

To measure the Cd concentration of test plants, two hills were harvested from three locations in the experimental plots. Harvested plants were separated into shoots and panicles and dried at 80°C for 2 days. Sample grinding and

| Field A | Field B | Field C | Field D | Field E | Field F |
|---------|---------|---------|---------|---------|---------|
| Year    | 2012    | 2013    | 2014    | 2015    | 2016    |
| Base dressing fertilizers (gN m⁻²) | 11.0 | 8.0 | 4.0 | 4.0 | 6.0 | 6.0 |
| Cd concentration (mg kg⁻¹) | 0.55 ± 0.09 | 1.47 ± 0.07 | 0.73 ± 0.13 | 0.96 ± 0.08 | 0.75 ± 0.09 | 0.55 ± 0.08 |
digestion were performed as described previously (Takahashi et al. 2016). To measure the soil Cd concentration, soil was collected from five or six locations in each field (fields A–F). The Cd concentration was measured using inductively coupled plasma optical emission spectrometry (ICP-OES: Vista-PRO; Varian, Australia). The Cd extraction was calculated as Cd concentration in the shoots × shoots dry weight per area.

Results

Breeding process and agronomic characteristics

‘Akita 119’ is a rice line developed at the Akita Prefectural Agricultural Experiment Station. We used soft X-rays to irradiate seeds of a naturally high-Cd-accumulating rice cultivar, ‘Cho-ko-koku’, in 2005 (Table 2). Forty-five M2 plants were identified from 35,000 plants in the initial screening based on their non-shattering habit and suitable heading date for Akita Prefecture. After the M3 generation, we selected rice lines based on the Cd concentration in the aerial parts of the plants. In the M6 generation, we identified plants that possessed the OsHMA3 allele of ‘Cho-ko-koku’. In the M6 generation (2011), one of the promising breeding lines was designated ‘Akikei 697’, and we determined agronomic traits, yield, and Cd extraction until the M11 generation (2016). ‘Akikei 697’ was eventually evaluated as a promising candidate rice line for Cd phytoremediation and was redesignated ‘Akita 119’. The heading date of ‘Akita 119’ was around 2 weeks later than that of ‘Akita-komachi’ (Table 3). ‘Akita 119’ had a short culm length and large panicle numbers (Fig. 1A). The culm of ‘Akita 119’ was around 30 cm shorter than that of ‘Cho-ko-koku’. The shattering resistance of ‘Akita 119’ was strong, and the lodging resistance of ‘Akita 119’ was improved compared to ‘Cho-ko-koku’. The thousand-grain weight of ‘Akita 119’ was slightly lower than that of ‘Cho-ko-koku’ and much lower than that of ‘Akitakomachi’. The grain size and shape of ‘Akita 119’ were different from those of ‘Akitakomachi’, and its grains could be easily distinguished from general japonica cultivars (Table 4, Fig. 1B, 1C). In the M12 generation (2017), the plant type of ‘Akita 119’ was investigated using 30 plants from six lines to determine the genetic fixation. Culm length in most ‘Akita 119’ plants

| Year | Generation | Process |
|------|------------|---------|
| 2005 | M1         | Mutation processing to seed of ‘Cho-ko-koku’ by soft X-ray |
| 2006 | M2         | Initial screening by non-shattering habit and heading date (35,000 plants→45 plants) |
| 2007 | M3         | Line selection |
| 2008 | M4         | Marker check (OsHMA3) |
| 2009 | M5         | Named ‘Akikei 697’ |
| 2011 | M6         | Agronomic traits tests |
| 2012 | M7         | Yield tests, Cd extraction tests |
| 2013 | M8         | Named ‘Akita 119’ |
| 2014 | M9         | Check genetic fixation |
| 2015 | M10        | |
| 2016 | M11        | |
| 2017 | M12        | |

Table 2. The ‘Akita 119’ breeding process

| Year | Generation | Process |
|------|------------|---------|
| 2005 | M1         | Mutation processing to seed of ‘Cho-ko-koku’ by soft X-ray |
| 2006 | M2         | Initial screening by non-shattering habit and heading date (35,000 plants→45 plants) |
| 2007 | M3         | Line selection |
| 2008 | M4         | Marker check (OsHMA3) |
| 2009 | M5         | Named ‘Akikei 697’ |
| 2011 | M6         | Agronomic traits tests |
| 2012 | M7         | Yield tests, Cd extraction tests |
| 2013 | M8         | Named ‘Akita 119’ |
| 2014 | M9         | Check genetic fixation |
| 2015 | M10        | |
| 2016 | M11        | |
| 2017 | M12        | |

Table 3. Characteristics of ‘Akita 119’. The data shown are averages calculated for the period 2011–2016. Lodging was evaluated on an ordinal scale from 0 (not lodged) to 5 (completely lodged)

| Year | Generation | Process |
|------|------------|---------|
| 2005 | M1         | Mutation processing to seed of ‘Cho-ko-koku’ by soft X-ray |
| 2006 | M2         | Initial screening by non-shattering habit and heading date (35,000 plants→45 plants) |
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| 2012 | M7         | Yield tests, Cd extraction tests |
| 2013 | M8         | Named ‘Akita 119’ |
| 2014 | M9         | Check genetic fixation |
| 2015 | M10        | |
| 2016 | M11        | |
| 2017 | M12        | |

Fig. 1. (A) ‘Akita 119’ in the maturing stage. White bars indicate 30 cm. (B) ‘Akita 119’ grains. (C) ‘Akita 119’ decorticated grains.

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|------|------------|---------|
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| 2006 | M2         | Initial screening by non-shattering habit and heading date (35,000 plants→45 plants) |
| 2007 | M3         | Line selection |
| 2008 | M4         | Marker check (OsHMA3) |
| 2009 | M5         | Named ‘Akikei 697’ |
| 2011 | M6         | Agronomic traits tests |
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| 2014 | M9         | Check genetic fixation |
| 2015 | M10        | |
| 2016 | M11        | |
| 2017 | M12        | |

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| 2005 | M1         | Mutation processing to seed of ‘Cho-ko-koku’ by soft X-ray |
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| 2013 | M8         | Named ‘Akita 119’ |
| 2014 | M9         | Check genetic fixation |
| 2015 | M10        | |
| 2016 | M11        | |
| 2017 | M12        | |

Table 3. Characteristics of ‘Akita 119’. The data shown are averages calculated for the period 2011–2016. Lodging was evaluated on an ordinal scale from 0 (not lodged) to 5 (completely lodged)
was between 45 and 55 cm, which was short compared with ‘Cho-ko-koku’ and ‘Akitakomachi’ (Table 5). The panicle length of ‘Akita 119’ was slightly shorter than that of ‘Cho-ko-koku’. The panicle number was almost the same as that of ‘Cho-ko-koku’, although much higher panicle numbers were observed in the agronomic trait and yield tests (Table 3).

### Cadmium extraction capacity of ‘Akita 119’

To investigate the Cd extraction capacity, ‘Akita 119’ was grown in a Cd-contaminated field located in Akita Prefecture from 2011 to 2016. Both the shoot dry weight and Cd concentration of ‘Akita 119’ were similar to those of ‘Cho-ko-koku’ in each field (Table 6). Although the ‘Akita 119’ culm was much shorter than that of ‘Cho-ko-koku’, shoot dry weight did not differ due to the many tillers of

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### Table 4. Size and shape of decorticated grains of ‘Akita 119’. Twenty grains were investigated per experimental plot, with two replicates in 2017

| Length (mm) | Width (mm) | Thickness (mm) | Length/Width | Shape              |
|-------------|------------|----------------|--------------|--------------------|
| Akita 119   | 5.80       | 1.80           | 1.47         | 3.23              |
| Cho-ko-koku | 6.06       | 1.78           | 1.58         | 3.41              |
| Akita 110   | 5.67       | 2.17           | 1.52         | 2.61              |
| Akitakomachi| 4.90       | 2.87           | 2.08         | 1.71              |

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### Table 5. Culm length (A), panicle length (B), and panicle number (C) of ‘Akita 119’. Thirty plants of six lines were investigated in 2017.

#### (A) Culm length

| Line No. | Average ± SD | CV (%) | Culm length (cm) |
|----------|--------------|--------|------------------|
|          | ~40 ~45 ~50 ~55 ~60 ~65 ~70 ~75 ~80 ~85 ~90 ~95 Total |
| Akita 119| 1 50.1 ± 4.8 | 9.6    | 1 4 12 10 3 0 0 0 0 0 0 0 30 |
|          | 2 48.8 ± 4.9 | 10.0   | 0 11 8 9 1 1 0 0 0 0 0 0 30 |
|          | 3 51.0 ± 5.2 | 10.2   | 2 5 9 9 7 0 0 0 0 0 0 0 30 |
|          | 4 50.6 ± 4.0 | 7.9    | 3 11 13 2 1 0 0 0 0 0 0 0 30 |
|          | 5 53.4 ± 3.9 | 7.3    | 0 0 5 19 5 1 0 0 0 0 0 0 30 |
|          | 6 52.4 ± 4.5 | 8.6    | 1 10 11 7 1 0 0 0 0 0 0 0 30 |
| Cho-ko-koku| 82.9 ± 8.6 | 5.5    | 0 0 0 0 0 0 0 0 5 3 14 8 1 30 |
| Akitakomachi| 69.6 ± 4.2 | 6.0    | 0 0 0 0 0 8 10 8 4 0 0 0 30 |

#### (B) Panicle length

| Line No. | Average ± SD | CV (%) | Panicle length (cm) |
|----------|--------------|--------|---------------------|
|          | ~16 ~18 ~20 ~22 ~24 ~26 Total |
| Akita 119| 1 19.8 ± 1.0 | 5.1    | 0 2 17 10 1 0 30 |
|          | 2 19.7 ± 1.0 | 5.1    | 0 0 18 12 0 0 30 |
|          | 3 20.0 ± 0.9 | 4.5    | 0 0 14 16 0 0 30 |
|          | 4 19.9 ± 1.1 | 5.5    | 1 14 14 1 1 0 30 |
|          | 5 19.9 ± 0.9 | 4.5    | 0 0 17 12 1 0 30 |
|          | 6 19.5 ± 1.1 | 5.6    | 0 5 14 11 0 0 30 |
| Cho-ko-koku| 20.6 ± 1.0 | 4.9    | 0 0 13 15 2 2 30 |
| Akitakomachi| 17.7 ± 1.1 | 6.2    | 2 19 9 0 0 30 |

#### (C) Panicle number

| Line No. | Average ± SD | CV (%) | Panicle number per hill |
|----------|--------------|--------|-------------------------|
|          | ~6 ~9 ~12 ~15 ~18 ~21 Total |
| Akita 119| 1 13.0 ± 2.8 | 21.5   | 0 2 13 10 3 2 30 |
|          | 2 11.5 ± 2.5 | 21.7   | 0 4 17 8 0 1 30 |
|          | 3 12.6 ± 2.4 | 19.0   | 0 1 15 9 5 0 30 |
|          | 4 12.5 ± 2.5 | 20.0   | 0 3 12 11 4 0 30 |
|          | 5 13.2 ± 2.7 | 20.5   | 0 1 15 6 7 1 30 |
|          | 6 12.8 ± 2.9 | 22.7   | 0 3 10 12 4 1 30 |
| Cho-ko-koku| 12.7 ± 2.4 | 18.9   | 0 2 14 11 2 1 30 |
| Akitakomachi| 8.6 ± 1.5 | 17.4   | 1 20 9 0 0 30 |
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Table 6. Shoot dry weight (DW), cadmium (Cd) concentration in the shoots, and Cd extraction from ‘Akita 119’. Different letters indicate statistically significant differences between cultivars (Tukey–Kramer test, \( P < 0.05 \))

| Field A | Field B | Field C |
|---------|---------|---------|
|         |         |         |
| Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) | Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) | Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) |
| ‘Akita 119’ | 726.6* | 16.6a | 11.9* | 830.1* | 84.4a | 70.9* | 786.6* | 15.2a | 14.9* |
| ‘Cho-ko-koku’ | 647.9* | 17.0b | 10.8* | 894.2* | 76.4b | 68.5* | 666.5* | 14.5b | 12.2* |
| ‘Akita 110’ | 883.9* | 14.2a | 12.7* | 830.1* | 59.8b | 48.7* | 561.0* | 11.1b | 7.6b |
| Akitakomachi | 689.3* | 4.1b | 2.8b | 660.3* | 18.0b | 11.9b | 472.0* | 2.9b | 1.6b |

| Field D | Field E | Field F |
|---------|---------|---------|
|         |         |         |
| Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) | Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) | Shoots DW (g m\(^{-2}\)) | Cd conc. (mg kg\(^{-1}\)) | Cd extraction (mg m\(^{-2}\)) |
| ‘Akita 119’ | 672.8* | 17.6a | 13.9* | 768.0* | 15.2a | 15.2a | 534.1* | 11.0a | 8.2a |
| ‘Cho-ko-koku’ | 643.8* | 19.7b | 15.1a | 610.7* | 16.1a | 13.1a | 469.9b | 12.8b | 8.2b |
| ‘Akita 110’ | 751.4* | 18.7a | 15.7a | 621.0* | 11.1b | 8.5b | 451.3ab | 14.5a | 8.1a |
| Akitakomachi | 641.7* | 4.7b | 3.4b | 623.1* | 3.0a | 2.2a | 368.5b | 2.5b | 1.1b |

‘Akita 119’. As a result, Cd extraction using ‘Akita 119’ was similar to that achieved by ‘Cho-ko-koku’ in all fields, whereas Cd extraction using ‘Akita 110’ was lower than that achieved by ‘Cho-ko-koku’ in fields C and E.

**Discussion**

Rice is an effective plant for practical Cd phytoremediation of paddy fields (Ishikawa et al. 2006). A rice variety used for Cd phytoremediation is required to have excellent agronomic characteristics and high Cd-extraction capacity. The lodging resistance of ‘Akita 119’ was stronger than that of ‘Cho-ko-koku’ because of the shorter culm length, and ‘Akita 119’ also had high shattering resistance (Table 3). ‘Akita 119’ had high tiller numbers, and its biomass was as great as that of ‘Cho-ko-koku’ (Table 6). These results demonstrated that ‘Akita 119’ exhibited improved agronomic characteristics compared to ‘Cho-ko-koku’.

Cd extraction rates depend on shoot Cd concentration and shoot dry weight. ‘Akita 119’ attained the same Cd concentration and shoot dry weight as ‘Cho-ko-koku’ in all fields (Table 6). As a result, their Cd extraction rates were similar. However, less Cd was sometimes extracted from ‘Akita 110’ than from ‘Cho-ko-koku’ (Table 6). The Cd concentration of ‘Akita 110’ was lower than that of ‘Cho-ko-koku’ in fields B and E, and the shoot dry weight of ‘Akita 110’ was lower than that of ‘Cho-ko-koku’ in field C. Cadmium absorption from the soil to rice roots is dependent on soil conditions after the heading stage, and it is maximized under drained and oxidative soil conditions (Ibaraki et al. 2009, Murakami et al. 2009). This result suggested that Cd phytoremediation by ‘Akita 119’ was less susceptible to soil conditions than ‘Akita 110’ would be. As observed for ‘Cho-ko-koku’, the gene responsible for Cd concentration in ‘Akita 119’ and ‘Akita 110’ shoots is OsHMA3, which suggests that they may have very similar Cd translocation ability from root to shoot. It is possible that the much higher panicle numbers observed in ‘Akita 119’ cause stably high Cd extraction ability compared to ‘Akita 110’. ‘Akita 119’ panicle numbers were found to be nearly identical to those of ‘Cho-ko-koku’ in a genetic fixation test (Table 5), whereas far higher panicle numbers were observed in agronomic trait and yield tests (Table 3). In 2017, it was not possible to increase the tiller number in the early stage because of the low temperature and sunshine duration in June in Akita (data not shown). It is possible that the tiller number of ‘Akita 119’ was affected by weather conditions in the early stage.

In conclusion, compared to ‘Cho-ko-koku’, ‘Akita 119’ exhibited improved agronomic characteristics and improved stability of Cd accumulation in shoots. ‘Akita 119’ also exhibited improved stability of Cd accumulation in shoots compared to ‘Akita 110’. The small grain size of ‘Akita 119’ requires only a short maturing period, and seed production can be stable even in northern parts of Japan. Therefore, ‘Akita 119’ is a promising candidate rice line for Cd phytoremediation of paddy fields in northern parts of Japan.

**Author Contribution Statement**

Breeding ‘Akita 119’ including yield and agronomic traits tests was performed by RT, KK, IK, SS, KS, RT, SM and TK. Measurement of Cd concentrations was performed by MI.

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