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Identification of univalent chromosomes in monosomic lines of cotton (*Gossypium hirsutum* L.) by means of cytogenetic markers

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The lack of clear morphological markers of cotton chromosomes contributed to the development of an unconventional method for marking chromosomes using translocations. Today, tester translocation cotton lines represent the most complete set of cytological markers. The results of cytogenetic analysis of F_1 hybrids obtained from crosses of monosomic cotton lines with translocation lines with identified chromosomes are presented. Cytogenetic identification and numbering of univalent chromosomes in 25 monosomic lines of the cytogenetic collection of the National University of Uzbekistan allowed us to establish the following univalent occurrences: chromosome 2 in four monosomic lines, chromosome 4 in 15 lines, chromosome 6 in four lines, chromosome 7 of the A_t-subgenome in one line and chromosome 18 of the D_t-subgenome in one line. The remaining 21 lines were duplicates of three non-homologous chromosomes. All monosomic lines identified were characterized by differences in univalent sizes, meiotic index, number of tetrads with micronuclei, pollen fertility, frequency of monosomy in the progeny, and a complex of morphological characters associated with the monosomy of the chromosome identified. Despite differences in the genotypic environment and methods for producing monosomics in the two cotton collections, there is a surprising coincidence of data suggesting a higher frequency of chromosomes 2, 4 and 6 occurring as monosomics, while the other chromosomes of the set occur as monosomics at a much lower frequency, and eight nonhomologous chromosomes (5, 8, 13 of the A_t-subgenome and 14, 15, 19, 22 and 24 of the D_t-subgenome of cotton) never do.

Key words: cotton; *Gossypium hirsutum* L.; cytogenetic analysis; cotton cytogenetics; monosomic lines; chromosome identification.

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Идентификация унивалентных хромосом у моносомных линий хлопчатника *Gossypium hirsutum* L. с помощью цитогенетических маркеров

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Отсутствие четких морфологических маркеров хромосом хлопчатника способствовало разработке нетрадиционного метода маркировки хромосом с помощью транслокаций. Тестерные транслокационные линии хлопчатника на сегодняшний день представляют собой наиболее полный набор цитологических маркеров. Приведены результаты цитогенетического анализа гибридов F_1, полученных от скрещиваний моносомных линий хлопчатника с транслокационными линиями с идентифицированными хромосомами. Цитогенетическая идентификация и нумерация унивалентных хромосом у 25 моносомных линий цитогенетической коллекции Национального университета Узбекистана позволила установить, что четыре моносомные линии имеют унивалентные хромосомы по хромосоме 2, 15 линий – по хромосоме 4, четыре линии – по хромосоме 6, одна линия – по хромосоме 7 A_t-субгенома и одна линия – по хромосоме 18 D_t-субгенома хлопчатника. Остальная 21 линия была дубликатом трех негомологичных хромосом. Все идентифицированные моносомные линии характеризовались различиями в размерах унивалентов, величине мейотического индекса, числа тетрад с микроядрами, фертильности пыльцы, частоты воспроизводства моносомного состояния в потомстве и комплекса морфологических признаков, ассоциированных с моносомией по идентифицированной хромосоме. Несмотря на различия в генотипической среде и методах получения моносомиков в двух коллекциях хлопчатника, наблюдается удивительное совпадение дан-

ных по большей частоте появления моносомиков по хромосомам 2, 4 и 6, тогда как моносомики по другим хромосомам набора появляются с куда меньшей частотой, а по восьми негомологичным хромосомам (5, 8, 13 A_t-субгенома и 14, 15, 19, 22 и 24 D_t-субгенома) вообще никогда не выявляются.
Ключевые слова: хлопчатник; *Gossypium hirsutum* L.; цитогенетический анализ; транслокационные линии; моносомные линии; идентификация хромосом.

Introduction

The need for a karyological study of cultures with weakly morphologically differentiated chromosomes has contributed to the development of non-traditional methods for marking chromosomes using translocations. Work on the creation of translocation tester sets that were obtained in five plant species was widespread: maise (Burnham, 1954), barley (Burnham et al., 1954), pea (Lamm, Miravalle, 1959), rye (Sybenga, Wolters, 1972) and tomato (Gill et al., 1980). The problem of identifying individual chromosomes of a set in species such as horse beans, beans, and soybeans was partially solved by obtaining translocation lines for some non-homologous chromosomes of the genome (Sjodin, 1971; Ashraf, Bassett, 1986; Mahama et al., 1999).

As is known, cultivated cotton species *Gossypium hirsutum* L. is an allotetraploid ($2n$ = 52) and includes two subgenomes (the A_t-subgenome and the D_t-subgenome). Brown M.S. et al. (1980) received 62 translocation lines of *G. hirsutum* cotton using X-, γ, Bikini radiation and fast neutron irradiation of seeds or pollen of various varieties, as well as several lines. In 58 of these lines, two non-homologous chromosomes were involved, in three – three chromosomes, and in one – four. To identify and number chromosomes, studies were carried out to classify translocated chromosomes as subgenomes. As a result of identification, it was found that the 26th chromosome was not involved in any of the translocations and was determined by the exclusion method, since for all years of research it was not possible to obtain a translocation involving this chromosome (Stelly, 1993). All these translocations include the most complete set of cytological markers for studying cotton genomes (Menzel, Brown, 1978).

In decision the problem of identifying small chromosomes, high hopes were associated with the use of the differential staining method, however, attempts to obtain differential chromosome banding sufficient to identify non-homologous chromosomes did not lead to the desired result (Turkov et al., 1980; Escalant, Schwendiman, 1984; Wang, 1985). The staining of prometaphase cotton chromosomes using BrdU-Hoechst-Giemsa and a special analysis system made it possible to detect from 2 to 9 main blocks on the chromosome that corresponded to early replicating DNA (Muravenko et al., 1998; Muravenko, Zelenin, 2009).

Due to the fact that cultivated cotton *G. hirsutum* is an allotetraploid and includes two subgenomes, it is tolerant to the loss of individual chromosomes. However, the creation of a series of monosomic lines in the United States characterized by the loss of individual chromosomes ($2n$ = 51) has been went on for many years (Endrizzi, Brown, 1964; Endrizzi et al., 1985). So, until 1985, only 15 of the 26 non-homologous chromosomes of *G. hirsutum* were isolated and identified in the United States. To date, the cotton cytogenetic collection created in the USA is characterized by the absence of any types of deficiencies for three chromosomes (13, 19 and 24), whereas for five non-homologous chromosomes (5, 8 A_t-subgenome and 14, 15, 22 D_t-subgenome), in collection have deficiencies only individual chromosome arms (Saha et al., 2012). However, this does not prevent their use for the chromosome localization of marker genes and the production of a series of substituted lines that are created simultaneously with the participation of three tetraploid species (Saha et al., 2004, 2006, 2013).

For many years, studies on the induction of cotton plants by chromosomal aberrations using various methods of induced mutagenesis have been conducted at the National University of Uzbekistan (Sanamyan, 2003, Samanyan, Rakhmatullina, 2003). As a result, a unique cytogenetic collection of cotton was created, including monosomic, monotelodisomic, and translocation lines, which place second in the world in terms of the number of lines after a similar collection created in the USA (Sanamyan et al., 2010, 2014).

The aim of the work is the unified identification of univalent chromosomes in previously obtained monosomic cotton lines using a set of cotton tester translocation lines with identified chromosomes.

Materials and methods

The research material was hybrid monosomic cotton plants obtained by crossing monosomic lines of different origin from the cytogenetic cotton collection *G. hirsutum* of the National University of Uzbekistan (NUUz) (Sanamyan et al., 2014), with translocation lines with identified chromosomes of the American cytogenetic collection (Stelly, 1993). Monosomic cotton lines grow year-round in the cellophane envelopy greenhouse of NUUz, which are monitored and all agricultural activities are carried out. Cytogenetic markers of a set of lines with identified translocations were kindly provided by Professor David Stelly (Department of Soil and Crop Sciences, Texas A&M University, College Station, TX, USA) through the ARS-USDA exchange program.

Cytological analyzes examined metaphase I (MI) meiosis in pollen mother cells (PMCs) by fixed 2–3 mm buds in alcohol-acetic acid (7:3). Then, PMCs were stained with iron-acetocarmine. The metaphases of the first division of meiosis were analyzed on temporary pressed preparaty under a light microscope and the nature of chromosome pairing, the number of uni-, bi-, tri- and multivalents were taken into account.

All cytological observations were carried out using microscopes Laboval, AxioScopeA1 (Carl Zeiss, Germany) and Biomed (Leica, Switzerland) with a magnification of 10×, 100×, binocular lens 1.6× and GF 12.5 × 120 and 10× eyepiece. Microphotography was performed using a Mikroskop kamera AxioCamERc5s digital camera. When exposure was

used green filter 3C-11-3. The plants and their parts were photographed using a CanonA-610 digital camera.

Results and discussion

The cytogenetic analysis of F_1 hybrids obtained from crosses of cotton monosomic lines with the translocation lines identified in accordance with the international nomenclature allowed identification of univalent chromosomes of monosomic lines of the cytogenetic collection of cotton of NUUz. It is known that the identification of pollen in mother cells in the metaphase I of meiosis in hybrid translocation monosomics of quadrivalent and univalent indicates the non-homology of the univalent and chromosomes involved in translocation (Endrizzi, Brown, 1964). If trivalents are found in hybrid translocation monosomics in the PMCs, this indicates the homology of the univalent and one of the chromosomes in the translocation. In this case, crosses of this monosomic plant are carried out with other translocation lines, in which one of their chromosomes in translocations was the same as that of the first line. Analysis of chromosome associations in monosomic hybrids allows us to identify the univalent chromosome as a specific chromosome of the set.

The analysis was carried out for all hybrid offspring obtained from crosses of cotton monosomic and translocation lines with identified chromosomes, however, only those variants that shared common chromosomes were included in the table for pairing of chromosomes (Table 1).

As a result of the analysis of cotton monosomic lines using a series of translocation lines with identified chromosomes, it was possible to identify univalent chromosomes in a number of lines. Thus, homology of univalent chromosomes was established in four monosomic lines (Mo11, Mo16, Mo19 and Mo93) and one of the translocated chromosomes in crosses with six translocation lines – TT2L-6R, TT2L-3Lb, TT2R-3La, TT2R- 8Ra, TT2R-8Rb, and TT2R-14R, since 24 bivalents plus one trivalent were observed in monosomic translocation hybrids in the meiosis metaphase I (Fig. 1, see Table 1). Since six translocation lines share one common chromosome 2, then the univalent chromosome lines in monosome Mo11, Mo16, Mo19, and Mo93 are chromosome 2 of the A_t subgenome of cotton and, as four monosomic lines are duplicates. Molecular genetic analysis of four monosomic interspecific F_1 hybrids with the participation of the lines Mo11, Mo16, Mo19 and Mo93 confirmed these data (Sanamyan et al., 2016).

Plants of the initial primary monosomics of all four monosomic cotton lines with deficiencies on chromosome 2 were obtained by pollination of irradiated pollen at doses of 10–25 Gy. All of them were characterized by an medium size of univalents, a high meiotic index (from 92.50 ± 0.31 to 98.25 ± 0.22), an increased number of tetrads with micronuclei (up to 3.00 ± 0.20 %) and high pollen fertility (from 91.54 ± 0.48 to 96.41 ± 0.42 %), as well as a reduced frequency of transmission of the monosomic state in the progenies (from 19.35 to 44.44 %) and a reduced transmission frequency of $n-1$ gametes. Moreover, once in the progeny of the monosomic line Mo19, the appearance of a monotelodisomic plant was noted, which indicated cases of incorrect (transverse) division of the centromere univalent in this monosomic line.

Table 1. Cytogenetic analysis of F_1 hybrids obtained from crosses of monosomic lines with translocation lines of the test set

Subgenome	Chromosome	Hybrids
A_t-subgenome	2	Mo11×TT 2L-3Lb
		Mo11×TT 2R-3La
		Mo11×TT 2R-8Ra
		Mo11×TT 2R-8Rb
		Mo11×TT 2R-14R
		Mo16×TT 2L-3Lb
		Mo16×TT 2R-8Ra
		Mo16×TT 2R-8Rb
		Mo16×TT 2R-14R
		Mo19×TT 2L-6R
		Mo19×TT 2L-3Lb
		Mo19×TT 2R-8Ra
		Mo19×TT 2R-8Rb
		Mo19×TT 2R-14R
		Mo93×TT 2R-8Ra
		Mo93×TT 2R-8Rb
		Mo93×TT 2R-14R
	4	Mo7×TT 4L-19R
		Mo31×TT 4L-19R
		Mo31×TT 4R-15L
		Mo38×TT 4L-19R
		Mo58×TT 4L-19R
		Mo59×TT 4L-19R
		Mo60×TT 4R-15L
		Mo69×TT 4L-19R
		Mo70×TT 4L-19R
		Mo70×TT 4R-15L
		Mo71×TT 4R-15L
		Mo72×TT 4L-19R
		Mo72×TT 4R-15L
		Mo73×TT 4L-19R
		Mo73×TT 4R-15L
		Mo75×TT 4L-19R
		Mo75×TT 4R-15L
		Mo76×TT 4L-19R
		Mo76×TT 4R-15L
		Mo81×TT 4L-19R
		Mo89×TT 4L-19R
		Mo89×TT 4R-15L
	6	Mo13×TT 3L-6L
		Mo13×TT 6L-7L
		Mo13×TT 6L-10R
		Mo34×TT 3L-6L
		Mo34×TT 6L-14L
		Mo67×TT 6L-7L
		Mo95×TT 3L-6L
		Mo95×TT 6L-10R
	7	Mo27×TT 1L-7L
		Mo27×TT 7L-12R
		Mo27×TT 7R-11R
		Mo27×TT 7R-21R
D_t-subgenome	18	Mo48×TT 7L-18R

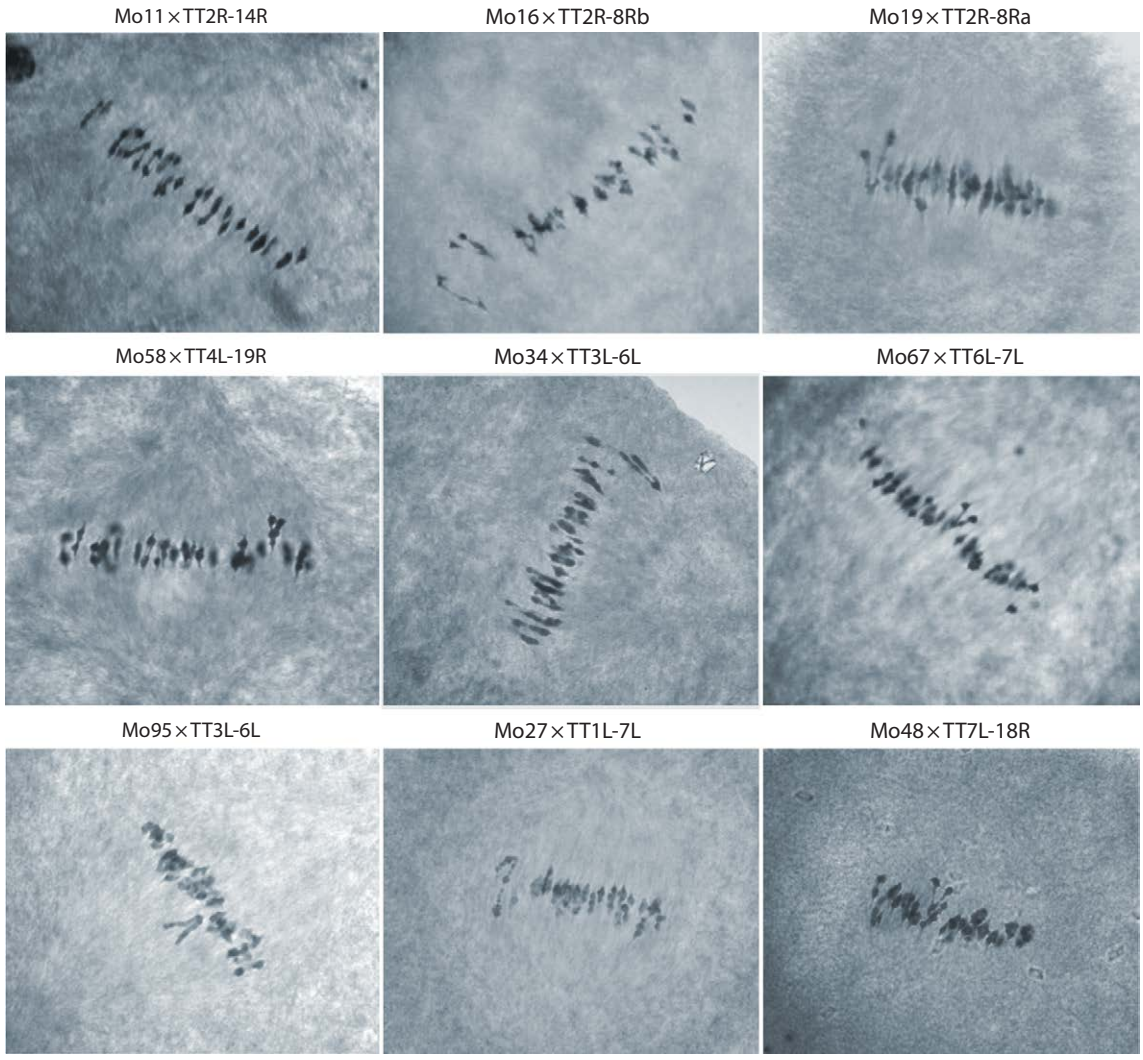

Fig. 1. Critical configuration of the chromosomes at the meiotic metaphase I in cotton monosome hybrids F_1, obtained from the crosses.

Monosomic lines on chromosome 2 were characterized by a similar set of characteristic phenotypic characters (Table 2, Fig. 2), low seed set (up to 54.84 %) compared with the original inbred line L-458 (89.81 %). This decrease in seed set occurred due to the presence of a large number of unfertilized ovules in the form of uluks in the monosomic bolls, the presence of which, along with a decrease in the number of seeds, led to a decrease in the size of the bolls.

When analyzing hybrids obtained by crossing seven monosomic lines of cotton (Mo31, Mo70, Mo72, Mo73, Mo75, Mo76, Mo89) with two TT4L-19R and TT4R-15L tester translocation lines, the homology of the univalents of these seven lines with one of the translocated chromosomes was established, since monosomic translocation hybrids in the metaphase I of meiosis showed 24 bivalents plus one trivalent (see Table 1, Fig. 1). In the TT4L-19R tester line, chromosomes 4 and 19 are involved in the translocation, and in the TT4R-15L line, chromosomes 4 and 15, therefore, one of these three chromosomes is homologous to the univalent chromosome in the monosomic lines Mo31, Mo70, Mo72, Mo73, Mo75, Mo76, Mo89. Since one common chromosome 4 is involved in both translocation lines, the mean univalent chromosomes of the monosomic lines Mo31, Mo70, Mo72, Mo73, Mo75, Mo76, Mo79 are chromosome 4 of the A_t-subgenome cotton, and these monosomic lines are duplicates. Molecular-genetic analysis of monosomic interspecific F_1 hybrids with the participation of the lines Mo31, Mo70, Mo72, Mo73, Mo75, Mo76, Mo89 confirmed these data (Sanamyan et al., 2016).

In the study of hybrids obtained by crossing eight monosomic cotton lines (Mo7, Mo38, Mo58, Mo59, Mo60, Mo69, Mo71 and Mo81) with one of two translocation lines – TT4L-19R or TT4R-15L, the homology of the univalents of these eight lines with one from translocated chromosomes was detected, since monosomic translocation hybrids in the meiotic metaphase I observed 24 bivalents plus one trivalent (see Table 1, Fig. 1). Since one common chromosome 4 is involved in both translocation lines, it can be assumed that the univalent chromosomes of the monosomic lines Mo7, Mo38, Mo58, Mo59, Mo60, Mo69, Mo71, and Mo81 are chromosome 4 of the A_t-subgenome cotton, and these monosomic lines are duplicates. The final cytological confirmation of this fact will be obtained after studying hybrids from the crossings of these eight monosomic lines with a different than the already studied translocation line involving chromosome 4. However, the lo-

Table 2. The origin and some characters of monosomic lines of cotton *G. hirsutum* L.

Monosomic line	Origin	Year of obtaining	Chromosome		Morphological characteristics
			Size	Identity	
Mo11	Pollen irradiation	1991	Medium	A 2	Small narrow leaf, shortened sympodial branches, small round bolls
Mo16		1991			
Mo19		1991			
Mo93		2007			
Mo7	Pollen irradiation	1990	Medium	A 4	Thick lush plant, elongated leaf blades, long bracts and pedicels, elongated ribbed bolls
Mo31		1993			
Mo38		1993			
Mo58	Desynapsis	1996			
Mo59		1996			
Mo60		1996			
Mo69		1997			
Mo70		1997			
Mo71		1997			
Mo72		1997			
Mo73		1997			
Mo75	Pollen irradiation	1999			
Mo76		2001			
Mo81		2003			
Mo89	Desynapsis	2003			
Mo13	Pollen irradiation	1991	Large	A 6	Sympodial branches, hard stem, small round bolls, late flowering
Mo34		1993			
Mo67	Heterozygous for translocation	1996			
Mo95	Pollen irradiation	2012			
Mo27	Pollen irradiation	1993	Medium	A 7	Short sympathies, thick bracts and leaves, small bolls
Mo48		1994	Small	D 18	Small leaves, long column and stigma, sympodial branches, round bolls

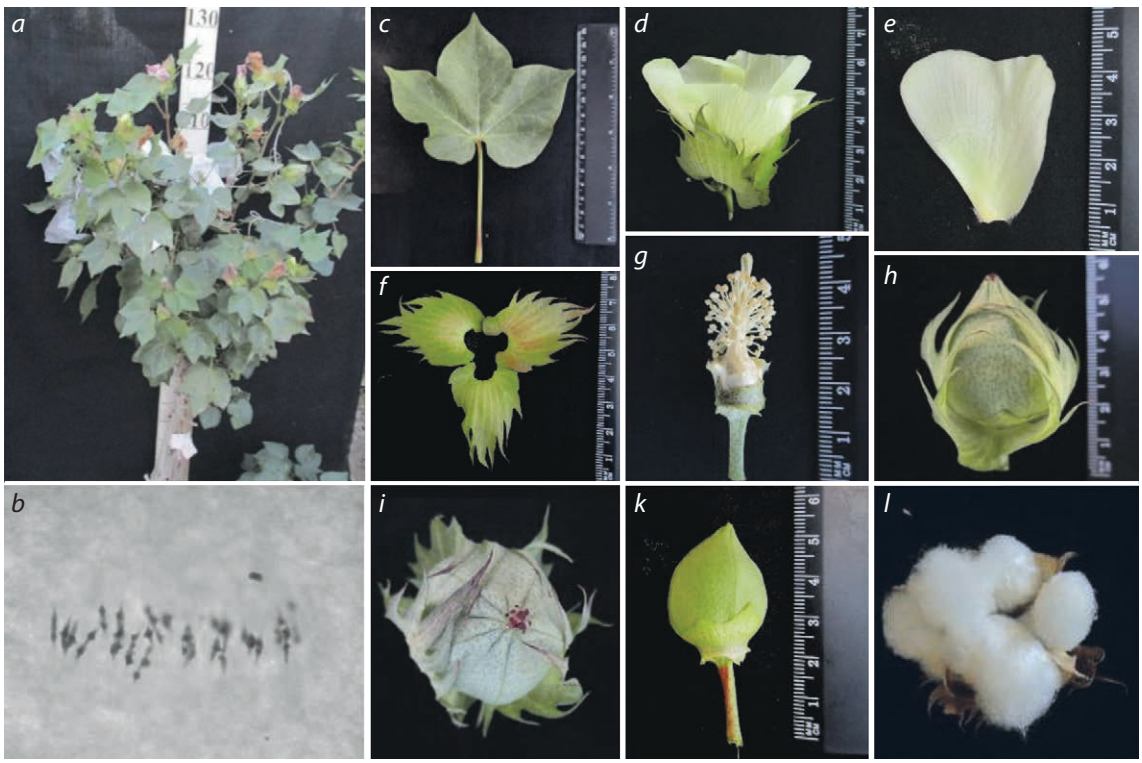

Fig. 2. Features of the cotton monosomic lines on chromosome 2:

a, bush; *b*, configurations of the chromosomes ($25^{II} + 1^{I}$); *c*, leaf; *d*, flower; *e*, petal; *f*, bract; *g*, staminate column; *h* and *i*, green bolls; *k*, boll with peduncle; *l*, open boll.

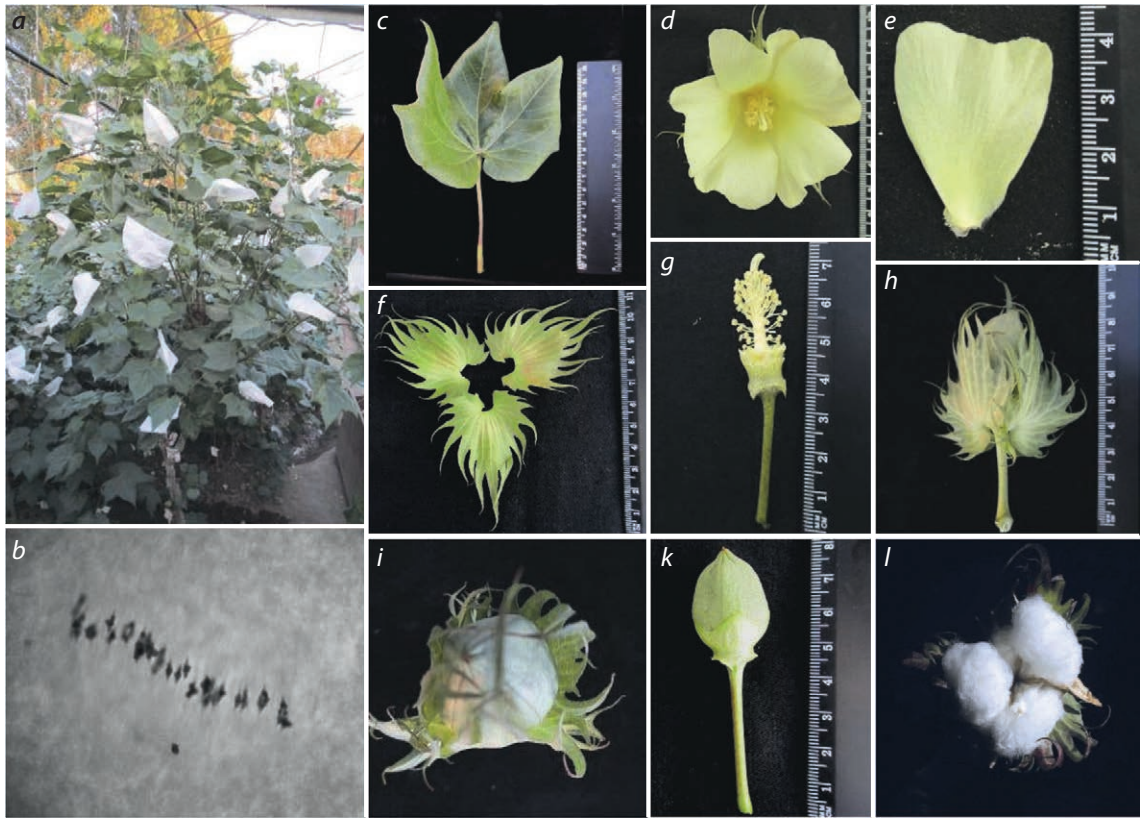

Fig. 3. Features of the cotton monosomic lines on chromosome 4:

a, bush; *b,* configurations of the chromosomes ($25^{II} + 1^{I}$); *c,* leaf; *d,* flower; *e,* petal; *f,* bract; *g,* staminate column; *h* and *i,* green bolls; *k,* boll with peduncle; *l,* open boll.

calization of chromosome-specific SSR markers on F_1 hybrids involving the lines Mo7, Mo38, Mo58, Mo59, Mo60, Mo69, Mo71, and Mo81 confirmed this data (Sanamyan et al., 2016).

Unfortunately, the third translocation line of the test set, TT4L-5, could not be used in a study to identify univalent chromosomes due to our discovery of two quadrivalents in "critical cells", apparently because of the homozygosity of both translocations simultaneously. Earlier M.S. Brown (1980) reported that two or more cytological aberrations were present in the initial plants of ten of the 62 translocation lines of cotton *G. hirsutum*, however only one translocation was obtained in a homozygous state later on their translocation lines.

The initial plants of six monosomic cotton lines (Mo7, Mo31, Mo38, Mo75, Mo76, Mo81) with deficiency of chromosome 4 were obtained as a result of pollination with irradiated pollen at doses of 10–25 Gy, while the original plants of nine monosomic lines (Mo58, Mo59, Mo60, Mo69, Mo70, Mo71, Mo72, Mo73, Mo89) were found in the progeny of desynaptic plants. All of the above lines were characterized by medium size of univalents, a high meiotic index (from 90.50 ± 0.72 to 98.46 ± 0.20), small number of tetrads with micronuclei (up to 2.07 ± 0.14 %) and high pollen fertility (from 90.67 ± 0.88 to 97.53 ± 0.35 %), as well as a reduced frequency of transmission of the monosomic state in the progeny (from 16.67 to 42.86 %), which led to a decrease in the frequency of transmission of haplo-deficient gametes.

Monosomic lines on chromosome 4 were characterized by similar phenotypic differences, which sharply distinguished them from other monosomic lines (Fig. 3, see Table 2), as well as a higher seed set (up to 72.22 %), with the exception of the Mo76 monosomic line (32.61 %).

When studying four monosomic cotton lines (Mo13, Mo34, Mo67, Mo95) using some of the four translocation lines – TT3L-6L, TT6L-7L, TT6L-10R, TT6L-14L, homology of univalent chromosomes was established for these four monosomic lines and one of the translocated chromosomes in the above translocations, since 24 bivalents plus one trivalent were observed in monosomic translocation hybrids in the meiosis metaphase I (see Table 1, Fig. 1). Since one common chromosome 6 is involved in four translocation lines, then the univalent chromosomes in monosomic lines Mo13, Mo34, Mo67, Mo95 are chromosome 6 A_t-subgenome cotton, and these monosomic lines are duplicates. Molecular-genetic analysis of four monosomic interspecific F_1 hybrids with the participation of the monosomic lines Mo13, Mo34, Mo67, Mo95 confirmed these data (Sanamyan et al., 2016).

The initial plants of three monosomic lines of cotton (Mo13, Mo34, Mo95) with a deficiency of chromosome 6 were obtained by pollination with irradiated pollen at doses of 20–25 Gy, while the original plant of the monosomic line Mo67 was found in the progeny of a plant heterozygous for translocation with a desynaptic effect. All these lines were characterized by a large univalent size, high meiotic index (from 94.13 ± 0.38 to 96.82 ± 0.49), a small number of tetrads with micronuclei (up to 2.07 ± 0.23 %) and reduced pollen fertility (from 88.46 ± 1.28 to 94.34 ± 0.51 %), as well as a

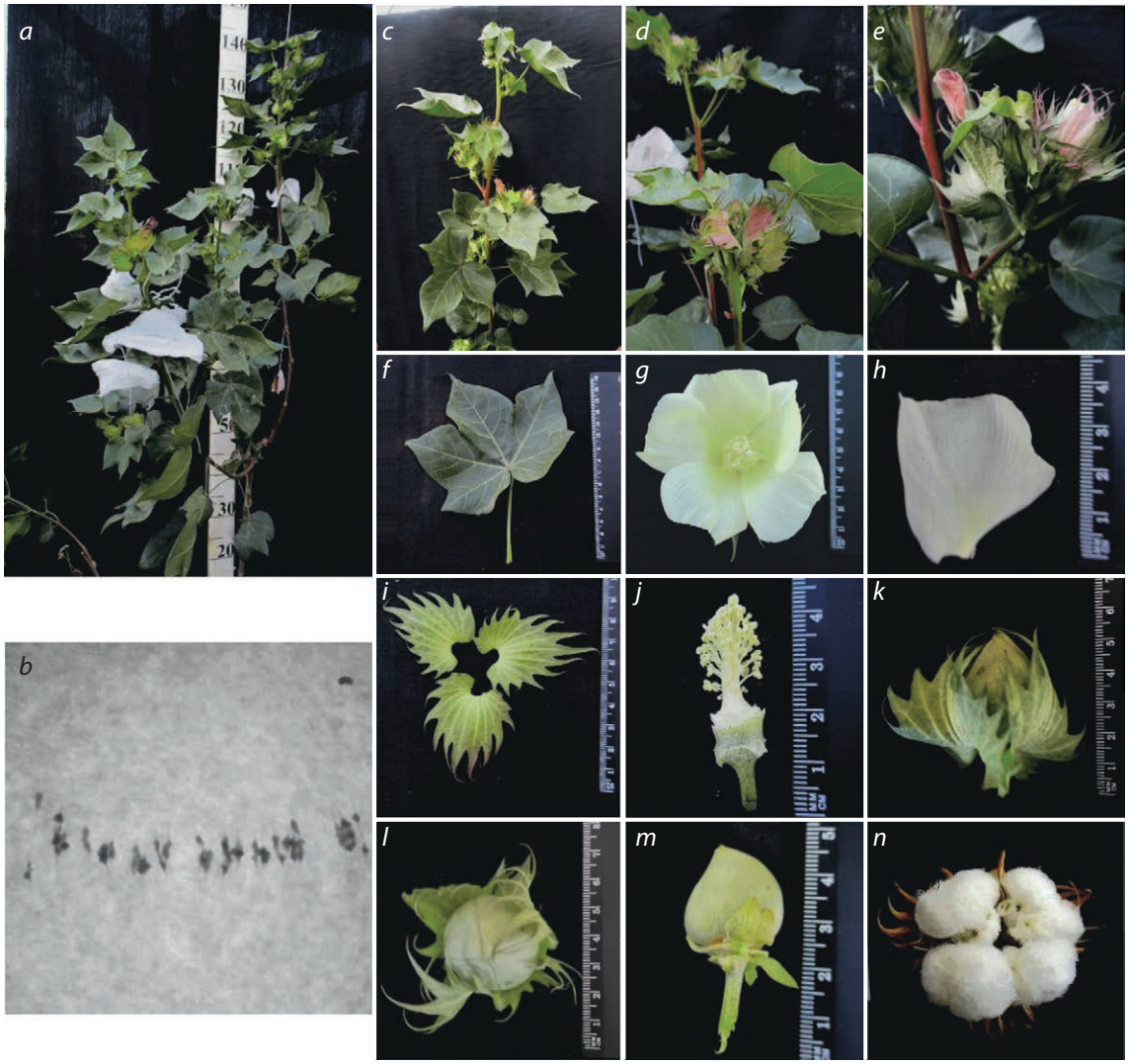

Fig. 4. Features of the cotton monosomic lines on chromosome 6:

a, bush; *b*, configurations of the chromosomes ($25^{II} + 1^{I}$); *c–e*, parts of the stem; *f*, leaf; *g*, flower; *h*, petal; *i*, bract; *j*, staminate column; *k* and *l*, green bolls; *m*, boll with peduncle; *n*, open boll.

low frequency of transmission of the monosomic state in the progeny (from 9.38 to 14.29 %), which significantly reduced the frequency of transmission of haplo-deficient gametes.

Monosomic lines with a deficiency of chromosome 6 were characterized by a whole complex of morphological characters associated with monosomy on this chromosome (Fig. 4, see Table 2), as well as lower seed set (from 38.78 to 57.45 %), compared with line L-458.

In the study of the Mo27 monosomic line in four variants of the crossings – Mo27×TT1L-7L, Mo27×TT7L-12R, Mo27×TT7R-11R and Mo27×TT7R-21R, homology of the univalent chromosome Mo27 and one of the translocated chromosomes was observed, since the monosome chromosome was observed 24 bivalents and one trivalent (see Fig. 1). In the TT1L-7L tester line, the involved chromosomes 1 and 7 are involved in translocation, in the TT7L-12R line, chromosomes 7 and 12, in the TT7R-11R line, chromosomes 7 and 11, and in the TT7R-21R line, chromosomes 7 and 21, therefore, one of these chromosomes is homologous to the univalent chromosome of the monosomic Mo27 line. Since one common chromosome 7 is involved in four-translocation lines, the univalent chromosome of the monosomic line Mo27 is chromosome 7 of the A_t-subgenome of cotton.

The initial plant of the monosomic cotton line Mo27 with a deficiency of chromosome 7 was obtained by pollination with irradiated pollen at a dose of 20 Gy. This line was characterized by an medium univalent size, a high meiotic index (95.81 ± 0.38), a small number of tetrads with micronuclei (1.77 ± 0.25 %) and reduced pollen fertility (89.88 ± 0.83 %), as well as a low transmission rate of the monosomic state in the progeny (22.23 %), which significantly reduced the frequency of transmission of haplo-deficient gametes. The monosomic line with a lack of chromosome 7 was also characterized by a complex of morphological characters associated with monosomy (Fig. 5, see Table 2), as well as reduced seed binding (65.10 %), compared with the L-458 line.

When studying the Mo48 monosomic line in one variant of crossing with the TT7L-18R translocation line in the meiosis metaphase I, 24 bivalents plus trivalent were found (see Fig. 1), which testified to the homology of the uni-

Fig. 5. Features of the cotton monosomic line on chromosome 7:

a, bush; *b*, configurations of the chromosomes ($25^{II}+1^{I}$); *c*, part of the stem; *d*, leaf; *e*, flower; *f*, bract; *g*, staminate column; *h* and *i*, green bolls; *k*, boll with peduncle; *l*, open boll.

valent chromosome in Mo48 and one of the translocated chromosomes in the translocation line. Since chromosomes 7 and 18 are involved in the translocation of the TT7L-18R test line, it can be assumed that the univalent chromosome in the monosomic line of Mo48 is homologous to one of the two chromosomes. Unfortunately, in the test set of lines with identified chromosomes, there is no second translocation line involving chromosome 18. Therefore, to determine which of the two chromosomes of this translocation, the homologous univalent chromosome of the Mo48 monosomic line was used earlier, we used chromosome-specific microsatellite SSR markers that were amplified by standard PCR. Molecular-genetic analysis of the monosomic interspecific hybrid F_1 (Mo48×Pima 3-79) revealed the presence of polymorphic alleles only from the species *Gossypium barbadense* L., which indicated the localization of the specific SSR marker BNL3280 chromosomes in the aforementioned hybrid (Sanamyan et al., 2016). Since this marker was previously located on the chromosome of the 18 D_t-subgenome of cotton, we can assume that the monosomic line – Mo48 of the collection of NUUs has a monosomy along the chromosome of the 18 D_t-subgenome.

The initial plant of the monosomic cotton line Mo48 with a deficiency of chromosome 18 was obtained by pollination with irradiated pollen at a dose of 25 Gy. This line was characterized by a small univalent size, a high meiotic index (95.68 ± 0.50), a small number of tetrads with micronuclei (0.86 ± 0.26 %) and a high pollen fertility (95.23 ± 0.74 %), and also low transmission rate of the monosomic state in the progeny (18.19 %), which significantly reduced the frequency of transmission of haplo-deficient gametes. The monosomic line with a deficiency of chromosome 18 was characterized by a set of morphological characters associated with monosomy (Fig. 6, see Table 2), as well as by high of seed set (85.64 %).

Conclusion

Thus, the use of translocation lines with identified chromosomes allowed us to bring the numbering of univalent chromosomes in the monosomic lines of our collection in accordance with the generally recognized nomenclature. Cytogenetic identification and numbering of univalent chromosomes in 25 monosomic lines of the cytogenetic collection of NUUs made it possible to establish that the four monosomic lines have univalent chromosomes on the chromosome 2, 15 lines on chromosome 4, the four lines on chromosome 6, the one line on chromosome 7 of A_t-subgenome and one line on chromosome 18 of D_t-subgenome of cotton. The predominant majority of monosomic lines were detected by the most frequently recorded cotton monosomics – chromosomes 2, 4, and 6.

A comparative analysis of the first 20 identified cotton monosomics obtained in the USA revealed similar trends, since the study revealed seven monosomics on chromosome 2, seven on chromosome 4, three on chromosome 6, and one on chromosomes 1, 17 and 18 (Brown, Endrizzi, 1964). The similarity of the data obtained in the study of different collections indicates that, despite differences in the genotypic environment and methods for producing monosomics, cotton has an amazing coincidence of data on the higher frequency of monosomics on chromosomes 2, 4 and 6, while monosomics on other the

Fig. 6. Features of the cotton monosomic line on chromosome 18:

a, bush; *b*, configurations of the chromosomes ($25^{II}+1^{I}$); *c*, leaf; *d*, flower; *e*, petal; *f*, bract; *g*, staminate column; *h*, green boll; *i*, boll with peduncle; *k* and *l*, open boll.

chromosomes of the set appear with a much lower frequency, and on eight non-homologous chromosomes (5, 8, 13 A_t-subgenome and 14, 15, 19, 22, and 24 D_t-subgenome of cotton) they were never detected at all (Saha et al., 2012). Apparently, the centromere regions of certain chromosomes are more prone to breakage and the genome as a whole remains tolerant of the loss of large A_t-subgenomic chromosomes without a large effect on viability and fertility, while the chromosomes of some small D_t-subgenomic chromosomes are not subject to any changes due to incompatibility with vitality.

A comparative analysis of the cytogenetic features of cotton monosomics from the two collections is not possible, since the literature contains only fragmentary information regarding misdivision of univalents and the frequency of transmission rate of the monosomic state in some monosomics of the American collection. However, the difficulties of creating a series of monosomic lines in tetraploid cotton and the obvious fact that certain chromosomes of the A_t-subgenome are more common in the monosomic state than the chromosomes of the D_t-subgenome do not diminish the value of the studies in view of the need for further development of molecular genetic studies and the creation of substituted cotton lines.

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