SSR diversity analysis of *Citrullus* landraces from India and their genetic inter relationship with continental watermelons

Dilbag Singh^{a*}, Rajinder Singh^b, Jagdeep Singh Sandhu^c, Parveen Chunneja^c

^a *Department of Vegetable Science, Punjab Agricultural University, Ludhiana, Punjab 141004, India*

^b *Office of Director Seeds, Punjab Agricultural University, Ludhiana, Punjab 141004, India*

^c *School of Agricultural Biotechnology, Punjab Agricultural University, Ludhiana, Punjab 141004, India*

*Corresponding author: *E-mail address:* dilbag.khullar@pau.edu

ABSTRACT

In a first report, watermelon (*Citrullus lanatus*) landraces from diversity rich Indo-Gangetic plains and arid regions of India were characterised using watermelon specific SSR markers. Thirty eight landraces were compared with germplasm of diverse origin comprising of cultivars, landraces and wild types. Significant differences were observed for days to first fruit maturity, average fruit weight, fruit yield/vine, TSS and Lycopene content. Thirty three polymorphic SSRs generated 120 bands with a mean of 3.63 per marker. Average PIC value was 0.46. Observed heterozygosity (0.50) was more than expected heterozygosity (0.45). Neighbour joining tree based on SSR analysis grouped accessions into seven clusters. Wild types formed a distinct cluster. Landraces showed genetic intermediateness with both the wild and cultivated watermelons. Combined morphological and molecular analysis revealed modern cultivars from USA, China, Japan and South Korea genetically resembled with each other. Hybridisation between landraces and exotic cultivars can be exploited to expand the genetic variability and introduce new traits in the breeding programs.

Key words: Citrullus; Breeding potential; Genetic diversity; Landraces; SSR

1. Introduction

Watermelon (*Citrullus lanatus*.), an ancient cucurbit belongs to family *Cucurbitaceae*. According to Maggs-Colling and Christiansen (2003), all *Citrullus spp* originated in Kalahari region of Africa. Paris (2015) in his work on ethno-botany of watermelon, articulated multiple origin hypothesis for dessert watermelon, but settled for north-eastern Africa as origin of sweet watermelon. However, there was no consensus on its putative progenitor. Cultivated watermelon has three species *Citrullus lanatus* var. *lanatus*, *C. lanatus* var. *citroides* and *C. mucospermus* (Fursa, 1972). *Citrullus colocynthis* and *Citrullus eccirrhosus* are two perennial wild species. *Praecitrullus fistulosus* (Stocks) Pangalo commonly called ‘Tinda’ from India and Pakistan is a related species.

Watermelon production in India was 1825 thousand tonnes in 2015, a significant six fold jump from 375 thousand tonnes in year 2011 ([www. nhb.gov.in](http://www.nhb.gov.in)). Due to increasing disposable incomes, more population is including fruits and vegetables in their daily diet, which earlier used to be mostly cereal based. Affordability and nutritive value of watermelon makes it preferred choice for the consumers. Various marker assisted diversity studies have reported narrow genetic base in cultivated watermelon (Levi *et al*, 2001 (a); Levi *et al* 2001 (b); Kwon *et al*, 2010) though species variation exists. Diversity analysis of indigenous watermelon genetic resources from Namibia (Maggs-Kolling and Christiansen, 2003), Sudan (Goda, 2007), Mozambique (Munisse *et al*, 2011), Zimbabwe (Mujaju *et al*, 2010), elucidated the role land races play in economy and nutrition of native communities. Besides, landraces are carriers of distinctive traits and can redress narrow genetic base, agronomic problems and conservation of genetic resources.

SSR markers are widely used for germplasm characterization, DNA finger printing for varietal identification and the construction of genetic linkage maps for tagging important traits (Levi et al, 2011; Kwon *et al.*, 2010; Levi *et al.*, 2006). Sequence specific SSR markers have been developed for genetic characterization (Guerra-Sanz, 2002; Joobeur et al, 2006; Levi et al, 2006 and Zhang et al, 2012) and DNA finger printing (Sheng et al, 2014) in watermelon.

Domestication of watermelon began about 4000 years ago (Gichimu, *et al.* 2009). Predominant species of watermelon found in India are *Citrullus lanatus* and *C. colocynthis*. Since its introduction in 8th century in India, watermelon has been in cultivation for multiple purposes. Farmer developed landraces are grown under irrigated or conserved moisture conditions in river beds in the Indo-Gangetic plains of Uttar Pradesh, Punjab and Haryana, for desert fruits commonly called *tarbuj* (dialect synonym). In arid and semi-arid regions of Thar Desert of Rajasthan watermelon is grown in mixed cropping systems, mainly for seed purpose. Here the fruit is called *mateera*. Landraces form a vital part of subsistence farming and family nourishment in these areas (Mahla and Chaudhary, 2013). From India it was taken to China, Central Asia and onwards to old world in 13th century and to North America in 17th century (Jeffery, 1975). There have only been sporadic reference of Indian origin watermelons in various diversity and taxonomic studies, although Indian genetic stocks have contributed to the development of cultivars elsewhere e.g. cultivars ‘Mickylee’ and ‘Bush Sugarbaby’ from USA owe their origin to Indian genetic pool (Hwang et al, 2011). Systematic study on genotyping of watermelon resources endemic to India could provide meaningful insight into genetic makeup of these landraces and their genetic relationship with global watermelons and also, in understanding the domestication and evolutionary processes in watermelon.

In India, key watermelon production areas fall in north-western plains of Indo-Gangetic region (sub-tropical with mild winters) and coastal states of Tamil Nadu, Karnataka

and Orissa (tropical). These regions also harbour maximum watermelon diversity in India (Wehner, 2009). Therefore, it becomes imperative to understand the population structure of the well adapted but so far neglected landraces from these regions for their inclusion in national watermelon breeding program.

The objectives of this study were aimed at: SSR based genetic diversity within and among watermelon landraces from India, inter relationship with global watermelons resources; conservation and future exploitation of the variation.

2. Materials and methods

2.1 Plant material

Eighty eight accessions belonging to different *Citrullus* species were used for molecular characterisation. Thirty eight landraces were collected from Uttar Pradesh (Indo-Gangetic plain region) and Rajasthan (Arid and semi-arid region). Germplasm from diverse geographic origin was included to give wider representation to the study material (Table 1).

2.2 Morphological evaluation

Seed sowing was done in polythene bags of size 6 x 4 inches filled with a mixture of soil and FYM in the ratio 1:1. Four week old seedlings were transplanted in the field on furrows at a spacing of 3.5 m (RR) x 0.5 m (PP). The accessions were evaluated in a Randomized Block Design replicated thrice. Each entry consisted of 10 plants in each replication. Recommended agronomic and plant protection practices (Anonymous, 2014) were followed to raise healthy crop. Five randomly selected plants from each replication were chosen for data collection and mean for each observation was used for statistical analysis. Data were recorded for days to first fruit maturity, average fruit weight (kg), fruit yield per vine (kg), total soluble solids (°Brix) and lycopene content (µg/g of fruit flesh). Fully mature fruits were selected

for biochemical analysis. Total soluble solids were measured using a hand held refractometer. Lycopene content was measured as prescribed by (Srivastava and Kumar, 2006).

2.3 Isolation and purification of genomic DNA

Genomic DNA was extracted from 5g of well ground tissue using the protocol described by Gusmini et al (2004). DNA was extracted from young leaves (3 weeks old) of five randomly selected plants in each accession and then bulked for subsequent analysis. Air dried DNA pellets were dissolved in 50 µl of 1X TE buffer (Tris-EDTA buffer- 10 mM Tris-HCl, 1 mM EDTA, pH 8.0. Quantity and quality of DNA was determined by Nanodrop 1000 instrument (Thermo Scientific, USA) using 2 µl of genomic DNA. Absorbance was recorded at 260/280 nm and readings were taken for both the quantity (ng/µl) and quality (Absorbance). Only the samples having Absorbance value from 1.90-2.00 were taken for DNA analysis.

2.4 Selection of primers

SSR primers (synthesized by IDT, USA) were selected from previously conducted mapping and diversity studies in watermelon. The in vitro DNA amplification through polymerase chain reaction (PCR) taking 30 ng DNA template were performed in Eppendorf thermal cycler using a final reaction volume of 12.5 µl. The DNA amplification conditions were as: initial cycle at 94 °C for 3 minutes followed by 30 cycles of: denaturation at 94 °C for 1 min, annealing at 44.0-55.0 °C (depending upon the primer), extension at 72 °C for 2 minutes and final extension at 72 °C for 15 minutes. The amplified product was resolved on 6% polyacrylamide gel electrophoresis (PAGE) at 300 V for 75 minutes. The gels stained with 100 µl of ethidium bromide (Promega, USA) were visualized under UV gel documentation (Thermo Scientific, USA). The SSR amplicons were recorded in a binary matrix as 1 (band present) and 0 (band absent). Total number of alleles for each primer were then scored in all the accessions under study.

2.5 Statistical analysis

Dissimilarity matrix was constructed using DICE's dissimilarity coefficient to measure the genetic inter relationship among the genotypes. The data was also subjected to Unweighted Pair Group Method with Arithmetic Mean (UPGMA) analysis to generate Neighbour Joining (NJ) tree dendogram using the software DARwin 6.0 (Perrier and Jacquemoud-Collet, 2006). Factorial correspondence analysis (FCA) was performed using the same software. PIC value for each primer were calculated using the formula given by Nei (1983) to determine the extent of polymorphism shown by each SSR marker. Observed heterozygosity (H_o) and expected heterozygosity (H_e) were also calculated (Marshall et al., 1998) Where; H_o = number of heterozygous individuals per number scored.

Expected heterozygosity (H_e) was calculated by the following formula: $H_e = 1 - \sum p_i^2$

3. Results

3.1 Horticultural performance comparison

Analysis of variance showed significant differences (data not shown) among the genotypes for first fruit maturity, fruit weight, fruit yield per vine, TSS and lycopene content. Pooled data (2 years) for each trait each accession for are presented in Table 1. All *Citrullus colocynthis* accessions except EC-829819 were poor in field performance. They were marked by lateness in fruit maturity, low in yield and poor fruit quality. On purely horticultural performance basis landrace WM-20 collected from Uttar Pradesh and EC-829878 were earliest (82 days) to first fruit maturity. Overall, average fruit weight (7.15 kg) and yield per vine (8.48 kg) was highest in EC-829840. IC-374802 recorded highest fruit weight (4.62 kg) and IC-611630 the highest fruit yield per vine (7.52 kg) among genotypes of Indian origin. Over-

all, TSS (10.85 °Brix) and lycopene content (93.37 µg/g) was highest in EC-829858 whereas among Indian genotypes maximum TSS (10.08 °Brix) and lycopene content (67.03 µg/g) was measured in Arka Manik. Range and mean for different metric traits for accessions constituting three groups viz. *colocynthis*, landraces and exotic germplasm are presented in Table 2. Exotic genotypes fared significantly better over landraces for mean fruit weight, fruit yield per vine, TSS and lycopene content.

Broad elliptic (80%) was chief fruit shape among landraces while (15%) has spherical fruits. Flesh colour in landraces varied from pinkish red (47.5%), red (30%), yellow (17.5%), white and orange (5%) each. Rind colour of fruit was grey (37.5%), green (40%) and light green (22.5%). Stripes were absent in 22.5% fruits whereas they were diffused in 17.5%, narrow (27.5%), medium (15%) and broad (17.5%) of fruits. Exotic genotypes had broad elliptic shape in 47.6% fruits, elliptic (23.8%), narrow elliptic and spherical (14.28% each). Red was dominant flesh colour (54.7%), followed by pinkish red (38.09%). Rind colour and stripe pattern was quite varied too. Green rind colour in varied degree was dominant (90%), whereas one genotype EC-829828 had yellow rind. Stripe width was narrow, medium and broad in 19%, 33% and 23% fruits.

3.2 SSR marker analysis

Fifty SSR primer combinations were used to discriminate the genotypes. Thirty three polymorphic SSRs amplified 120 bands with an average of 3.63 bands per marker. SSR primer pair (MCPI 16) amplified largest number of polymorphic bands i.e. 9 and primers MCPI 20, MCPI 29, WMA 6, BVWS 333, BVWS 441, BVWS 948, CLG 7996, EST 00674 and EST 00667 amplified lowest number of bands i.e. 2 each. The average polymorphic information content (PIC) was (0.46), with a range as low as 0.11 (CLG 7996) to as high as he 0.84 (MCPI 16). The number of SSR amplicons generated by each primer combination and their respective PIC values are presented in Table (3). Seventeen alleles (7.05%) were exclu-

sive to *C. colocynthis*, while 14 (8.57%) were found to be present only in *C. lanatus*. Observed heterozygosity (H_o) ranged from 0.15 in CLG 7996 to 0.88 in WMA 15, with mean heterozygosity at 0.50.

According to NJ tree (Fig. 1) dendrogram, all the eighty eight accessions were divided into seven distinct clusters viz. I, II, III, IV, V, VI and VII. Geographical affiliations in grouping were observed in some clusters. Clusters III, V and VII contained the landraces only. All landraces collected from Rajasthan were grouped in Cluster V, delineating them from those originated from other parts of India. Exotic genotypes occupied two clusters viz. II and IV, whereas wild watermelons along with one *C. amarus* accession were grouped in Cluster VI. USA genotypes were inter mixed with those of Indian sub continental and Asian accessions. Cluster IV had all genotypes from China, Japan, USSR and South Korea. The FCA plot (Fig. 2) is consistent with NJ tree dendrogram and showed differentiation between different *Citrullus spp.* genotyped in this study. Maximum dissimilarity was observed in Group VI and IV and was minimum in case of Group VI consisting of *colocynth* and Group I largely occupied by landraces from India.

4. Discussion

4.1 Morphological aspects

Variation in fruit shape, stripe pattern and flesh colour was relatively more in exotic genotypes than in landraces which could be attributed to the composition of exotic material included in our study, since in different countries consumer preferences vary for fruit size, shape and rind pattern. Scamosi et al (2009) found significant variation in fruit and flesh appearance in Turkish and Hungarian watermelons. Landraces from Uttar Pradesh (*tarbuj*) mostly had grey or dark green rind with very thin stripes or lack of them, whereas those from Rajasthan (*mateera*) had light green to green rind colour with medium to broad dark green stripes. Cultivars from China, USA, Japan and South Korea resembled each other in fruit

quality aspects, as most of them had red vibrant flesh generally associated with high TSS and lycopene content (Perkins-Veazie et al, 2001). (Hakimi and El Madidi (2015) and Elbekkay et al (2009) reported low performance in terms of fruit yield and quality of landraces in comparison to cultivars. Among *colocynthis* accessions, EC-829819 was found to be distinct from others in having red flesh, larger fruit weight and higher TSS and lycopene content uncharacteristic of *C. colocynthis*. All *colocynthis* accessions except EC-829819, were similar in fruit appearance at earlier fruit development stages and became yellow when fully mature. Our description of wild watermelon is in accordance with Yaniv et al (1999). EC-829810, lone *Citrullus amarus* Shrad, (termed *C. citroides*, Paris, 2015) accession was closer to *C. colocynthis* than *C. lanatus*. Levi et al (2013) observed two Egyptian accessions PI 525081 and PI 525083 previously registered as colocynthis and citron, respectively were found to be closer to dessert watermelons. Therefore, classification of watermelons on purely phenotypic basis could be deceptive and erroneous.

4.2 SSR analysis and genetic inter-relationships

Taxonomic classification in genus *Citrullus* is contradictory and concerted efforts are in progress to bring in clarity in classification of different species, sub species and botanical varieties in watermelon (Paris, 2015). Previously, citron, *egusi* and dessert watermelons were recognised as sub species or botanical varieties of *C. lanatus* (Hwang et al, 2011, Levi 2001). Fresh phylogenetic analysis has proposed separate species allocation for cultivated watermelon based on differences in genome assembly among cultivated watermelons (Paris, 2015).

Natural hybrids between *C. lanatus* and *C. colocynthis* and their intermediary forms called *khar* and *tatumba* have been testified in India where these two species are grown in close vicinity (Mahla et al, 2014). Free crossability and gene flow between these two species might have imparted genetic variation in Indian watermelon landraces (Sain et al, 2002). Higher H_o over H_e pointed towards mixing of populations, since landraces are populations

219 instead of improved cultivars. Anecdotal evidence in Arabic names for watermelon *battikh*
 220 *hindi* and *batikh sindi* indicate the emergence of sweet watermelons in north-western India,
 221 and its' spread to other parts of Asia during the Islamic reign (Watson, 1983). According to
 222 SSR marker analysis and NJ dendrogram Indian landraces exhibited intercessor genetic rela-
 223 tionship to both, *lanatus* and *colocynthis* accessions. This is reflected in the lighter flesh col-
 224 our and moderate fruit quality traits possessed by the landraces. Landraces from Rajasthan
 225 viz. Jaipur (code 19), Jaiselmer (code 20), Bikaner (code 21) and Barmer (code 22) from
 226 Cluster V showed high genetic similarity with *C. lanatus* cultivars from Japan (EC-829852,
 227 code 68; Asahi Yamato, code 69), China (EC-829870; code 82), South Korea (EC-828874,
 228 code 86) grouped in Cluster IV. Whereas, landraces from Uttar Pradesh viz. WM-6 (SSR
 229 code 2), WM-10 (3), WM-11 (4), WM-20 (6), WM-21 (7), WM-35 (12), WM-36 (13) and
 230 WM-38 (14) were closer to *C. colocynthis* genetic similarity range from 10% to 25%. Aver-
 231 age fruit weight for all these accessions from Uttar Pradesh was significantly lower than
 232 mean weight of Indian landraces. Grouping of some of the USA cultivars with accessions
 233 from Indian subcontinent and Central Asia (Clusters I and II), point toward common ancestry
 234 and development of these cultivars from breeding material of similar genetic background.
 235 Cultivars from China, South Korea and Japan and USA, possessing high fruit quality coa-
 236 lesced together in Cluster IV, accentuating narrow genetic base among modern cultivars aris-
 237 ing through concerted selection for horticulturally acceptable traits.
 238 In nutshell, our study revealed unique structure of watermelon landraces from India. High
 239 polymorphism in SSRs showed their efficiency in differentiating various watermelon types.
 240 EC genotypes possessed high fruit quality and yield. However, watermelon breeding in India
 241 has been inconsistent. For example, first seedless hybrid 'Pusa Bedana' was developed in
 242 1972 at IARI, New Delhi and since then there is no tangible progress on this aspect. Hybridi-
 243 sation between EC- accessions and well adapted landraces would help breeders to create

more variability, genetic conservation and improvement of existing landraces. Landraces need to be probed further for a wider range of agronomic traits for exploiting their full potential.

Acknowledgements

The authors are thankful to Tiffany Fields, ARS-USDA, Griffin, USA for supplying the germplasm. We also thank Department of Vegetable Science, PAU, Ludhiana for financial and material support to conduct the study.

References

- Anonymous., 2015. www.nhb.gov.in/area-production.
- Chomicki, G., Renner, S. S., 2015. Watermelon origin solved with molecular phylogenetics including Linnaean material: another example of museomics. *New Phytologist* 205, 526–532.
- Fursa, T. B., 1972. K sistematike roda *Citrullus* Schrad. [On the taxonomy of genus *Citrullus* Schrad.]. *Bot. Z*, 57, 31-41.
- Gichimu, B. M., Owuor, B. O., Mwai, G. N., Dida, M. M., 2009. Morphological characterization of some wild and cultivated watermelon (*Citrullus sp.*) accessions in Kenya. *ARPJ. Agric. Biol. Sci.*, 4 (2), 10-18.
- Goda, M., 2007. Diversity of local genetic resources of watermelon [*Citrullus lanatus* (Thunb.) Matsum and Nakai] in Sudan. M Sc Thesis, Swedish Biodiversity Centre, Uppsala, Sweden.
- Guerra-Sanz, M., 2002. *Citrullus* Simple Sequence Repeats Markers from Sequence Databases. *Mol.Ecol. Notes* 2, 223-225.

267 Gusmini, G., Wehner, T. C., Joobeur, T., Dean, R. A., 2004. Protocol for DNA extraction
 268 from watermelon leaves for SSR marker studies. *Cucurbit. Genet. Coop. Rep.* 27, 25-
 269 29.

270 Hwang, J., Kang, J., Son, B., Kim, K., Park, Y., 2011. Genetic diversity in watermelon culti-
 271 vars and related species based on AFLP and EST-SSRs. *Not. Bot. Horti. Agrobi.* 39
 272 (2), 285-292.

273 Jeffrey, C., 1975. Further notes on *Cucurbitaceae*. 3rd edition. Some African taxa, *Kew Bul-*
 274 *letin* 30, 475-493.

275 Joobeur, T. G., Gusmini, G., Zhang, X., Levi, A., Xu, Y., Wehner, T. C., Oliver, M., Dean,
 276 R. A., 2006. Construction of a watermelon BAC library and identification of SSRs
 277 anchored to melon or Arabidopsis genomes. *Theor. Appl. Genet.* 112, 553-562.

278 Kwon, Y. S., Oh, Y. H., Yi, S. I., Kim, H. Y., An, J. M., Yang, S. G., Ok, S. H., Shin, J. S.,
 279 2010. Informative SSR markers for commercial variety discrimination in watermelon
 280 (*Citrullus lanatus*). *Genes and Genomics* 32, 115-122.

281 Levi, A., Thomas, C. E., Keinath, A. P., Wehner, T. C., 2001a. Genetic diversity among
 282 watermelon (*Citrullus lanatus* and *Citrullus colocynthis*) accessions. *Genet. Resour.*
 283 *Crop Evol.* 48, 559-566.

284 Levi, A., Thomas, C. E., Wehner, T. C., Zhang, X. P., 2001b. Low genetic diversity indicates
 285 the need to broaden the genetic base of cultivated watermelon. *Hort. Sci.* 36, 1096-
 286 1101.

287 Levi, A., Thomas, C. E., Trebitsh, T., Salman, A., King, J., Karalius, J., Newman, M.,
 288 Reddy, O. U. K., Xu, Y., Zhang, X., 2006. An extended linkage map for watermelon
 289 based on SRAP, AFLP, SSR, ISSR and RAPD markers. *J. Am. Soc. Hort. Sci.* 131,
 290 393-402.

291 Levi, A., Wetcher, P., Massey, L., Carter, L., Hopkins, D., 2011. An extended genetic linkage
 292 map for watermelon based on a testcross and a BC₂F₂ popluation. Am. J. Plant Sci. 2,
 293 93-110.

294 Levi, A., Thies, J. A., Wechter, W. P., Harrison, H. F., Simmons, A. M., Reddy, U. K.,
 295 Nimmakayala, P. Zhangjun, F., 2013. High-frequency oligonucleotides: targeting ac-
 296 tive gene (HFO-TAG) markers revealed wide genetic diversity among *Citrullus* spp.
 297 accessions used for enhancing disease or pest resistance in watermelon cultivars.
 298 Genet. Resour. Crop Evol. 60, 427–440.

299 Maggs-Kolling, G. L., Christiansen, J. L., 2003. Variability in Namibian landraces of water-
 300 melon. Euphytica 132, 251-258.

301 Mahla, H. R., Singh, J. P., Roy, M. M., 2014. Seed purpose watermelon in arid zone. Central
 302 Arid Zone Research Institute, Jodhpur, Rajasthan, India. pp 1-39.

303 Mahla, H. R., Choudhary, B. R., 2013. Genetic diversity in seed purpose watermelon (*Citrul-*
 304 *lus lanatus*) genotypes under rainfed situations of Thar Desert. Ind. J. Agric. Sci. 83,
 305 300-303.

306 Marshall, T.C., Slate, J., Kruuk, L. E. B., Pemberton, J. M., 1998. Statistical confidence for
 307 likelihood-based paternity inference in natural populations. Mol. Ecol. 7, 639-655.

308 Mujaju, C., Sehic, J., Werlemark, G., Garkava-Gustavsson, L., Fatih, M., Nybom, H., 2010.
 309 Genetic diversity in watermelon (*Citrullus lanatus*) landraces from Zimbabwe re-
 310 vealed by RAPD and SSR markers. Hereditas 147, 142–153.

311 Munisse, P., Jensen, B. D., Andersen, S. B., 2011. Genetic differentiation of watermelon
 312 landraces in Mozambique using microsatellite markers. African J. Biotechnol. 12,
 313 5513-5521.

314 Nei, M., Tajima, F., Tateno, Y., 1983. Accuracy of estimated phylogenetic trees from mo-
 315 lecular data. II. Gene frequency data. J. Mol. Evol. 19, 153-170.

316 Paris, H. S., 2015. Origin and emergence of the sweet dessert watermelon, *Citrullus lanatus*.
 317 Annals of Botany 116, 133-148.

318 Perkins-Veazie, P., Collins, J. K., Pair, S. D., Roberts, W., 2001. Lycopene content differs
 319 among red fleshed watermelon cultivars. J. Food Sci. Agri., 81, 983-987.

320 Perrier, X., Jacquemoud-Collet, J. P., 2006. DARwin software <http://darwin.cirad.fr/>

321 Sain, R.S., Joshi, P., Divakara Sastry, E. V., 2002. Cytogenetic analysis of interspecific hy-
 322 brids in genus *Citrullus* (*Cucurbitaceae*). Euphytica, 128 (2), 205-210.

323 Scamosi, C., Solmaz, I., Sari, N., Barsony, C., 2009. Morphological characterization of Hun-
 324 garian and Turkish watermelon (*Citrullus lanatus* (Thunb.) Matsum. et Nakai) genetic
 325 resources. Genet. Resour. Crop Evol. 56, 1091-1105.

326 Sheng, J. Z., Hong, J. Z., Xu, Q. L., Nan, H., Wen, G. L., 2014. Studies on DNA finger-
 327 printing and genetic diversity of seedless watermelon (*Citrullus lanatus*) varieties us-
 328 ing core simple sequence repeat (SSR) markers. J. Agri. Biotech. 22, 188-194.

329 Srivastava, R. P., Kumar, S., 2006. Fruit and Vegetable Preservation: Principles and Prac-
 330 tices. New Delhi, India: International Book Distribution Company, pp. 1-463.

331 Watson, A. M., 1983. Agricultural innovation in the early Islamic world. Cambridge: Cam-
 332 bridge University Press.

333 Wehner, T. C., 2009. Watermelon. In: Prohens J, Nuez F, editors. Vegetables I. Asteraceae,
 334 Brassicaceae, Chenopodiaceae, and Cucurbitaceae. New Delhi, India: Springer, pp. 381-
 335 418.

336 Yaniv, Z., Shabelsky, E., Schafferman, D., 1999. Colocynth: Potential Arid Land Oilseed
 337 from an Ancient Cucurbit. 1st edition In: J. Janick (editor); Perspectives on New Crops
 338 and New Uses. Alexandria, VA, USA: ASHS Press, 257-61.

339 Zhang, H., Wang, H., Guo, S., Ren, Y., Gong, G., Weng, Y., Xu, Y., 2012. Identification and
340 validation of a core set of microsatellite markers for genetic diversity analysis in watermelon,
341 *Citrullus lanatus* hunb. Matsum. & Nakai. Euphytica 186, 329–342.

342

343

344

Table 1: Origin, source, fruit appearance and metric traits of eighty eight watermelon genotypes

<i>Citrullus colocynthis</i>			Fruit characteristics			Metric traits					
Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to maturity	Fruit weight (kg)	Fruit number/ vine	Total yield (q/ha)	TSS (°Brix)	Lycopene content (µg/g)
42	Tumba 1	India	Spherical	White	Green/dark green broad	121.00	0.08	2.73	7.31	2.73	5.00
43	Tumba 5	India	Spherical	White	Green/ dark green broad	119.00	0.06	2.43	5.94	1.87	3.10
44	EC-829816	Iran	Spherical	Pale white	Green/ dark green broad	116.50	0.10	2.53	8.45	1.40	3.67
45	EC-829818	Cyprus	Spherical	White	Green/ dark green medium	112.83	0.21	3.07	14.92	1.93	4.20
46	EC-829819	Unknown	Broad elliptic	Pinkish red	Light green/ narrow stripes	106.33	1.41	2.27	72.98	7.10	23.43
<i>Citrullus amarus</i>			Fruit characteristics			Metric traits					
41	EC-829810	Unknown	Elliptic	White	Light green/ absent	113.33	1.65	1.90	61.58	3.00	14.40
<i>Citrullus lanatus</i> var. <i>lanatus</i>			Fruit characteristics			Metric traits					
1	WM-5	India	Broad elliptic	Red	Solid grey/absent	85.33	1.76	2.63	105.91	6.83	40.40
2	WM-6	India	Broad elliptic	Pinkish red	Solid grey/absent	87.67	1.71	2.62	102.21	6.95	37.57
3	WM-10	India	Broad elliptic	Yellow	Grey / green narrow stripes	88.33	2.13	2.38	116.48	7.78	16.90
4	WM-11	India	Spherical	Yellow	Solid grey/absent	92.33	1.40	2.82	90.20	6.63	18.87
5	WM-14	India	Broad elliptic	Red	Solid grey /absent	90.83	2.33	2.17	115.54	6.83	50.67
6	WM-20	India	Broad elliptic	Yellow	Light green/dark green broad	82.00	2.13	2.38	115.43	8.35	19.93
7	WM-21	India	Broad elliptic	Yellow	Grey/ dark green narrow	87.00	1.70	2.47	96.25	7.78	22.47
8	WM-22	India	Elliptic	Yellow	Grey/ dark green medium	89.00	2.69	1.82	114.06	8.62	16.13
9	WM-26	India	Broad elliptic	Yellow	Green/ dark green medium	85.50	2.34	1.88	100.33	7.47	21.77
10	WM-29	India	Broad elliptic	Orange	Grey green/ absent	87.50	2.58	1.85	108.78	7.93	32.53
11	WM-30	India	Spherical	White	Grey/dark green narrow	87.67	1.58	2.28	81.63	8.00	14.33
12	WM-35	India	Spherical	Red	Grey/ diffused stripes	89.50	1.57	2.62	93.68	8.33	38.70
13	WM-36	India	Spherical	Red	Grey/dark green narrow	85.00	1.29	2.90	85.72	6.28	57.70
14	WM-38	India	Spherical	Orange	Grey/dark green narrow	85.17	1.92	2.43	106.28	7.90	33.77
15	WM-41	India	Broad elliptic	Red	Solid grey/absent	92.33	2.10	2.60	124.91	6.43	51.33
16	WM-43	India	Broad elliptic	Pinkish red	Green/ diffused	97.67	2.77	1.98	125.59	7.22	37.60

Table 1: Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ pattern	Days to maturity	Fruit weight (kg)	Fruit number/ vine	Total Yield (q/ha)	TSS (°Brix)	Lycopene content (µg/g)
17	WM-46	India	Broad elliptic	Red	Solid grey/absent	95.33	3.82	1.55	135.00	7.30	65.73
18	WM-50	India	Broad elliptic	Pinkish red	Grey/ green diffused	90.50	3.83	1.60	139.99	7.60	31.83
19	Jaipur	India	Broad elliptic	Pinkish red	Green/ dark green medium	97.67	2.68	2.15	131.54	6.07	38.23
20	Jaiselmer	India	Broad elliptic	Red	Green/ dark green medium	93.00	3.49	1.78	143.34	6.27	50.67
21	Bikaner	India	Broad elliptic	Pinkish red	Light green/dark green narrow	97.67	2.89	2.13	139.02	7.30	45.50
22	Barmer	India	Broad elliptic	Red	Green/ dark green medium	97.83	3.22	2.07	150.80	8.38	53.30
23	Thar Manik	India	Broad elliptic	Pinkish red	Light green/ dark green broad	89.67	2.53	2.05	118.44	6.93	46.73
24	Arka Manik	India	Broad elliptic	Red	Green/ dark green broad	85.50	3.24	2.08	154.40	10.08	67.03
25	Arka Jyoti	India	Broad elliptic	Red	Green/ dark green broad	89.50	3.76	2.00	171.71	9.97	61.87
26	IC-374795	India	Broad elliptic	Yellow	Light green/dark green broad	96.67	1.76	2.27	92.01	6.15	14.73
27	IC-611625	India	Broad elliptic	Pinkish red	Green/ dark green very narrow	97.83	3.03	2.23	155.03	4.03	36.00
28	IC-611623	India	Broad elliptic	White	Green/absent	107.33	3.72	1.88	159.77	5.33	13.73
29	IC-611626	India	Spherical	Red	Light green/medium dark green	102.83	3.58	2.13	174.15	5.93	42.30
30	IC-324819	India	Broad elliptic	Pinkish red	Light green/green very narrow	106.17	2.53	2.45	142.06	6.15	32.33
31	IC-374820	India	Broad elliptic	Pinkish red	Light green/diffused	107.50	1.73	2.52	99.24	3.77	34.87
32	IC-315321	India	Broad elliptic	Pinkish red	Green/ dark green broad	94.83	3.18	2.03	148.06	5.77	29.07
33	IC-374802	India	Broad elliptic	Pinkish red	Green/ dark green narrow	101.17	4.62	1.55	163.74	6.63	27.50
34	IC-374803	India	Broad elliptic	Pinkish red	Green/diffused	105.17	2.83	2.02	130.39	5.48	41.67
35	IC-599377	India	Broad elliptic	Pinkish red	Green/ dark green narrow	107.33	4.32	1.60	158.06	7.75	50.50
36	IC-315324	India	Broad elliptic	Red	Green/dark green medium	97.67	2.30	2.20	115.20	5.48	45.47
37	IC-13773	India	Broad elliptic	Pinkish red	Light green/ green narrow	96.50	1.93	2.18	96.42	5.52	41.20
38	IC-611624	India	Elliptic	Pinkish red	Green/diffused	99.00	3.73	1.97	167.68	6.43	24.70
39	IC-315328	India	Broad elliptic	Pinkish red	Light green/ green broad	99.33	2.50	2.13	121.39	4.22	35.70
40	IC-325808	India	Broad elliptic	Pinkish red	Green/absent	101.67	3.39	1.97	149.89	6.08	30.73
62	EC-829844	Turkey	Broad elliptic	White	Green/ dark green medium	97.33	1.82	2.22	91.66	5.85	16.57

Table 1: Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to maturity	Fruit weight (kg)	Fruit number/vine	Total yield (q/acre)	TSS (°Brix)	Lycopene content (µg/g)
63	EC-829846	Nigeria	Broad elliptic	Pinkish red	Green/dark green medium	106.50	3.52	2.07	164.82	7.12	38.53
64	EC-829848	Nigeria	Broad elliptic	Pinkish red	Green/diffused	101.00	3.27	1.87	139.88	6.50	44.63
65	EC-829849	Iran	Broad elliptic	Pinkish red	Grey/dark green narrow	107.67	0.93	2.70	57.22	4.37	49.33
66	EC-829850	Iran	Broad elliptic	Pinkish red	Green/dark green medium	104.67	4.22	1.57	148.28	5.62	32.37
67	EC-829851	Japan	Broad elliptic	Red	Light green/dark green medium	91.00	2.29	2.57	134.85	9.64	70.87
68	EC-829852	Japan	Elliptic	Red	Light green/ narrow dark green	89.67	2.54	2.42	140.51	9.03	72.80
69	Asahi Yamato	Japan	Broad elliptic	Red	Light green/diffused	91.50	3.17	2.07	149.51	9.80	77.20
70	EC-829853	Belize	Spherical	Pinkish red	Grey/dark green medium	95.33	5.93	1.23	167.33	7.39	32.43
71	EC-829854	Greece	Narrow elliptic	Pinkish red	Light green/dark green narrow	98.00	5.68	1.23	160.02	6.35	28.20
72	EC-829858	China	Broad elliptic	Red	Light green/ diffused	86.83	3.67	2.13	178.19	10.85	93.37
73	EC-829860	Zimbabwe	Broad elliptic	Pinkish red	Green/ dark green narrow	106.50	2.99	2.03	138.66	4.27	29.97
74	EC-829861	Zimbabwe	Broad elliptic	Pinkish red	Green/dark green narrow	104.00	1.88	2.28	98.10	5.20	22.77
75	EC-829862	Zimbabwe	Elliptic	Pinkish red	Light green/ absent	105.17	1.34	2.38	74.03	4.15	26.70
76	EC-829863	USSR	Broad elliptic	Pinkish red	Green/ dark green broad	106.33	5.64	1.25	160.78	7.43	32.53
77	EC-829864	USSR	Broad elliptic	Red	Light green/dark green broad	104.33	3.58	1.92	157.07	7.49	35.47
78	EC-829865	Pakistan	Spherical	Pinkish red	Light green/ dark green broad	108.50	1.82	2.48	103.24	5.75	20.67
79	EC-829866	Pakistan	Broad elliptic	Pinkish red	Green/dark green very broad	108.67	1.69	2.33	89.72	5.12	37.67
80	EC-829867	Spain	Elliptic	Red	Dark green/ absent	100.83	1.88	2.20	93.26	6.99	53.17
81	EC-829868	Spain	Spherical	Pinkish red	Green/ dark green medium	87.00	4.07	1.60	148.60	7.59	21.60
82	EC-829870	China	Narrow elliptic	Orange	Green/ dark green very broad	84.17	3.60	2.00	164.93	7.95	25.83
83	EC-829871	China	Broad elliptic	Red	Light green/dark green medium	93.33	3.75	2.05	175.94	7.27	91.17
84	EC0829872	South Korea	Broad elliptic	Red	Light green/ dark green medium	91.67	2.35	2.03	109.37	9.75	56.93
85	EC-829873	South Korea	Broad elliptic	Red	Light green/diffused	88.83	2.64	2.00	120.94	6.80	55.63
86	EC-829874	South Korea	Spherical	Red	Light green/ dark green diffused	88.00	2.29	2.35	122.72	9.58	79.43

Table 1. Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to maturity	Fruit weight (kg)	Fruit number/vine	Total yield (q/acre)	TSS (°Brix)	Lycopene content (µg/g)
87	EC-829875	South Korea	Spherical	Red	Light green/absent	89.50	2.72	2.07	129.12	10.42	82.77
88	EC-829878	Canada	Spherical	Pale white	Light green/ dark green medium	82.00	3.39	2.22	172.09	9.91	12.40
<i>Citrullus lanatus</i>			Fruit characteristics			Metric traits					
	Accession number	Origin	Fruit shape	Flesh colour		Days to maturity	Fruit weight (kg)	Fruit number/vine	Total yield (q/acre)	TSS (°Brix)	Lycopene Content (µg/g)
47	EC-829820	USA	Broad elliptic	Red	Grey/green diffused	87.17	3.05	2.10	146.20	10.25	84.30
48	EC-829822	USA	Narrow elliptic	Red	Light green/dark green medium	90.83	3.93	1.90	171.07	9.10	68.73
49	EC-829823	USA	Elliptic	Red	Green/dark green narrow	85.83	3.49	2.12	168.90	9.45	73.17
50	EC-829824	USA	Narrow elliptic	Red	Green/dark green medium	92.50	4.69	1.65	176.95	8.94	60.63
51	EC-829825	USA	Elliptic	Red	Light green/dark green broad	90.33	3.40	2.13	166.12	8.53	63.87
52	EC-829826	USA	Broad elliptic	Red	Light green/ dark green broad	87.67	3.66	1.90	158.33	7.97	59.57
53	EC-829827	USA	Narrow elliptic	Pinkish red	Light green/ dark green broad	97.17	2.85	2.12	138.01	5.70	47.10
54	EC-829828	USA	Elliptic	Red	Yellow rind/ narrow stripes	86.00	2.01	2.42	111.10	9.70	69.63
55	EC-829829	USA	Elliptic	Red	Light green/dark green medium	97.67	3.65	2.15	178.23	8.63	54.37
56	EC-829832	USA	Broad elliptic	Red	Green/dark green narrow	89.67	2.86	1.93	126.50	9.62	71.53
57	EC-829833	USA	Elliptic	Red	Green/ dark green broad	91.33	3.13	2.12	151.72	6.78	56.43
58	EC-829836	USA	Elliptic	Pinkish red	Green/absent	97.33	1.32	2.32	70.18	6.60	54.03
59	EC-829838	USA	Narrow elliptic	Red	Light green/dark green broad	84.50	4.24	1.90	186.46	8.97	64.17
60	EC-829840	USA	Broad elliptic	Red	Green/dark green medium	97.50	7.15	1.18	193.73	8.53	58.07
61	EC-829841	Turkmenistan	Broad elliptic	Pinkish red	Green/diffused	105.50	3.23	1.93	143.23	5.65	35.33
L.S. D						2.86	0.31	0.16	10.92	0.71	4.58

F test was significant @ P< 0.001

*Accessions starting with IC and EC number are obtained from GRIN-USDA, Griffin, USA; WM- Accessions were collected from Uttar Pradesh, India; Jaipur, Jaiselmer, Barmer, Bikaner and Thar Manik from Rajasthan, India; Arka Manik and Arka Jyoti from Indian Institute of Horticultural Research, Bangalore, India

** 10q (quintals) = 1 ton

Table 2: Range and mean for different fruit traits for *Citrullus colocynthis*, Indian landraces and exotic germplasm

Group	Days to fruit maturity	Fruit weight (kg)	Fruit number/ vine	Total yield (q/acre)	Total soluble solids (°Brix)	Lycopene content (µg/g)
<i>Colocynthis</i>						
Range (low-high)	106 to 122	0.06-1.41	2.27-3.07	5.94-72.98	1.40-7.10	3.10-23.43
Mean	133	0.36		0.86		
Landraces						
Range (low-high)	82-107.5	1.29-4.62	1.55-2.90	81.00-174.15	3.77-10.08	14.33-67
Mean	94.26	2.67		5.51	6.84	37.69
Exotic germplasm						
Range (low-high)	82-108.67	0.93-7.15	1.23-2.70	57.22-193.73	4.15-10.85	12.40-93.37
Mean	95.22	3.22		6.13	7.68	51.69

Table 3: SSR primer, alleles amplified, PIC value and heterozygosity

Sr no	SSR primer	Number of amplicons	PIC value	Expected heterozygosity (H_e)	Observed heterozygosity (H_o)	Reference
1	MCPI 4	3	0.34	0.37	0.41	Joobeur et al (2006)
2	MCPI 5	8	0.73	0.55	0.61	
3	MCPI 16	9	0.84	0.65	0.72	
4	MCPI 17	3	0.52	0.60	0.69	
5	MCPI 20	2	0.31	0.32	0.34	
6	MCPI 23	5	0.71	0.54	0.65	
7	MCPI 29	2	0.33	0.23	0.18	
8	MCPI 32	5	0.65	0.69	0.79	
9	MCPI 33	3	0.48	0.37	0.47	
10	MCPI 39	3	0.35	0.38	0.48	
11	WMA 1	4	0.68	0.63	0.69	Lee et al (2007)
12	WMA 2	3	0.42	0.46	0.65	
13	WMA 5	3	0.45	0.54	0.58	
14	WMA 6	2	0.37	0.29	0.23	
15	WMA 11	3	0.38	0.42	0.55	
16	WMA 15	7	0.72	0.76	0.88	
17	WMA 26	3	0.35	0.30	0.35	
18	BVWS 333	2	0.36	0.33	0.29	Zhang et al (2012)
19	BVWS 441	2	0.37	0.39	0.35	
20	BVWS 948	2	0.20	0.23	0.21	
21	BVWS 433	7	0.78	0.64	0.69	
22	BVWS 314	3	0.49	0.58	0.68	
23	BVWS 658	3	0.30	0.38	0.46	
24	BVWS 2048	4	0.54	0.49	0.58	
25	ASUW 19	6	0.69	0.63	0.70	Levi et al (2006)
26	CLG 7996	2	0.11	0.21	0.15	
27	CLG 8218	3	0.24	0.26	0.38	
28	CLG 8288	4	0.51	0.36	0.44	
29	Cgb 4765	3	0.42	0.60	0.59	
30	EST 00674	2	0.33	0.41	0.36	Guerra-Sanz (2002)
31	AF 032471.1	4	0.57	0.63	0.72	
32	EST 00691	3	0.52	0.38	0.41	
33	EST 00667	2	0.31	0.26	0.19	
	Total	120	15.37	14.88	16.55	
	Mean	3.63	0.46	0.45	0.50	

- Primer sequences are given in supplementary table

Table 1: Origin, source, fruit appearance and metric traits of eighty eight watermelon genotypes

<i>Citrullus colocynthis</i>			Fruit characteristics			Metric traits				
Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to maturity	Fruit weight (kg)	Yield/ Vine (kg)	TSS (°Brix)	Lycopene content (µg/g)
42	Tumba 1	India	Spherical	White	Green/dark green broad	121.00	0.08	0.17	2.73	5.00
43	Tumba 5	India	Spherical	White	Green/ dark green broad	119.00	0.06	0.14	1.87	3.10
44	EC-829816	Iran	Spherical	Pale white	Green/ dark green broad	116.50	0.10	0.23	1.40	3.67
45	EC-829818	Cyprus	Spherical	White	Green/ dark green medium	112.83	0.21	0.65	1.93	4.20
46	EC-829819	Unknown	Broad elliptic	Pinkish red	Light green/ narrow stripes	106.33	1.41	3.19	7.10	23.43
<i>Citrullus amarus</i>			Fruit characteristics			Metric traits				
41	EC-829810	Unknown	Elliptic	White	Light green/ absent	113.33	1.65	2.69	3.00	14.40
<i>Citrullus lanatus</i> var. <i>lanatus</i>			Fruit characteristics			Metric traits				
1	WM-5	India	Broad elliptic	Red	Solid grey/absent	85.33	1.76	4.64	6.83	40.40
2	WM-6	India	Broad elliptic	Pinkish red	Solid grey/absent	87.67	1.71	4.47	6.95	37.57
3	WM-10	India	Broad elliptic	Yellow	Grey / green narrow stripes	88.33	2.13	5.10	7.78	16.90
4	WM-11	India	Spherical	Yellow	Solid grey/absent	92.33	1.40	3.95	6.63	18.87
5	WM-14	India	Broad elliptic	Red	Solid grey /absent	90.83	2.33	5.05	6.83	50.67
6	WM-20	India	Broad elliptic	Yellow	Light green/dark green broad	82.00	2.13	5.05	8.35	19.93
7	WM-21	India	Broad elliptic	Yellow	Grey/ dark green narrow	87.00	1.70	4.21	7.78	22.47
8	WM-22	India	Elliptic	Yellow	Grey/ dark green medium	89.00	2.69	4.99	8.62	16.13
9	WM-26	India	Broad elliptic	Yellow	Green/ dark green medium	85.50	2.34	4.39	7.47	21.77
10	WM-29	India	Broad elliptic	Orange	Grey green/ absent	87.50	2.58	4.86	7.93	32.53
11	WM-30	India	Spherical	White	Grey/dark green narrow	87.67	1.58	3.57	8.00	14.33
12	WM-35	India	Spherical	Red	Grey/ diffused stripes	89.50	1.57	4.10	8.33	38.70
13	WM-36	India	Spherical	Red	Grey/dark green narrow	85.00	1.29	3.75	6.28	57.70
14	WM-38	India	Spherical	Orange	Grey/dark green narrow	85.17	1.92	4.65	7.90	33.77
15	WM-41	India	Broad elliptic	Red	Solid grey/absent	92.33	2.10	5.47	6.43	51.33
16	WM-43	India	Broad elliptic	Pinkish red	Green/ diffused	97.67	2.77	5.50	7.22	37.60

Table 1: Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ pattern	Days to maturity	Fruit weight (kg)	Yield/ vine (kg)	TSS (°Brix)	Lycopene content (µg/g)
17	WM-46	India	Broad elliptic	Red	Solid grey/absent	95.33	3.82	5.91	7.30	65.73
18	WM-50	India	Broad elliptic	Pinkish red	Grey/ green diffused	90.50	3.83	6.13	7.60	31.83
19	Jaipur	India	Broad elliptic	Pinkish red	Green/ dark green medium	97.67	2.68	5.76	6.07	38.23
20	Jaiselmer	India	Broad elliptic	Red	Green/ dark green medium	93.00	3.49	6.27	6.27	50.67
21	Bikaner	India	Broad elliptic	Pinkish red	Light green/dark green narrow	97.67	2.89	6.08	7.30	45.50
22	Barmer	India	Broad elliptic	Red	Green/ dark green medium	97.83	3.22	6.60	8.38	53.30
23	Thar Manik	India	Broad elliptic	Pinkish red	Light green/ dark green broad	89.67	2.53	5.18	6.93	46.73
24	Arka Manik	India	Broad elliptic	Red	Green/ dark green broad	85.50	3.24	6.76	10.08	67.03
25	Arka Jyoti	India	Broad elliptic	Red	Green/ dark green broad	89.50	3.76	7.52	9.97	61.87
26	IC-374795	India	Broad elliptic	Yellow	Light green/dark green broad	96.67	1.76	4.03	6.15	14.73
27	IC-611625	India	Broad elliptic	Pinkish red	Green/ dark green very narrow	97.83	3.03	6.78	4.03	36.00
28	IC-611623	India	Broad elliptic	White	Green/absent	107.33	3.72	6.99	5.33	13.73
29	IC-611626	India	Spherical	Red	Light green/diffused	102.83	3.58	7.62	5.93	42.30
30	IC-324819	India	Broad elliptic	Pinkish red	Light green/green very narrow	106.17	2.53	6.22	6.15	32.33
31	IC-374820	India	Broad elliptic	Pinkish red	Light green/diffused	107.50	1.73	4.34	3.77	34.87
32	IC-315321	India	Broad elliptic	Pinkish red	Green/ dark green broad	94.83	3.18	6.48	5.77	29.07
33	IC-374802	India	Broad elliptic	Pinkish red	Green/ dark green narrow	101.17	4.62	7.16	6.63	27.50
34	IC-374803	India	Broad elliptic	Pinkish red	Green/diffused	105.17	2.83	5.71	5.48	41.67
35	IC-599377	India	Broad elliptic	Pinkish red	Green/ dark green narrow	107.33	4.32	6.92	7.75	50.50
36	IC-315324	India	Broad elliptic	Red	Green/dark green medium	97.67	2.30	5.04	5.48	45.47
37	IC-13773	India	Broad elliptic	Pinkish red	Light green/ green narrow	96.50	1.93	4.22	5.52	41.20
38	IC-611624	India	Elliptic	Pinkish red	Green/diffused	99.00	3.73	7.34	6.43	24.70
39	IC-315328	India	Broad elliptic	Pinkish red	Light green/ green broad	99.33	2.50	5.31	4.22	35.70
40	IC-325808	India	Broad elliptic	Pinkish red	Green/absent	101.67	3.39	6.56	6.08	30.73
62	EC-829844	Turkey	Broad elliptic	White	Green/ dark green medium	97.33	1.82	4.01	5.85	16.57

Table 1: Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to fruit maturity	Fruit weight (kg)	Yield/ Vine (kg)	TSS (°Brix)	Lycopene content (µg/g)
63	EC-829846	Nigeria	Broad elliptic	Pinkish red	Green/dark green medium	106.50	3.52	7.21	7.12	38.53
64	EC-829848	Nigeria	Broad elliptic	Pinkish red	Green/diffused	101.00	3.27	6.12	6.50	44.63
65	EC-829849	Iran	Broad elliptic	Pinkish red	Grey/dark green narrow	107.67	0.93	2.50	4.37	49.33
66	EC-829850	Iran	Broad elliptic	Pinkish red	Green/dark green medium	104.67	4.22	6.49	5.62	32.37
67	EC-829851	Japan	Broad elliptic	Red	Light green/dark green medium	91.00	2.29	5.90	9.64	70.87
68	EC-829852	Japan	Elliptic	Red	Light green/ narrow dark green	89.67	2.54	6.42	9.03	72.80
69	Asahi Yamato	Japan	Broad elliptic	Red	Light green/diffused	91.50	3.17	6.54	9.80	77.20
70	EC-829853	Belize	Spherical	Pinkish red	Grey/dark green medium	95.33	5.93	7.32	7.39	32.43
71	EC-829854	Greece	Narrow elliptic	Pinkish red	Light green/dark green narrow	98.00	5.68	7.00	6.35	28.20
72	EC-829858	China	Broad elliptic	Red	Light green/ diffused	86.83	3.67	7.80	10.85	93.37
73	EC-829860	Zimbabwe	Broad elliptic	Pinkish red	Green/ dark green narrow	106.50	2.99	6.07	4.27	29.97
74	EC-829861	Zimbabwe	Broad elliptic	Pinkish red	Green/dark green narrow	104.00	1.88	4.29	5.20	22.77
75	EC-829862	Zimbabwe	Elliptic	Pinkish red	Light green/ absent	105.17	1.34	3.24	4.15	26.70
76	EC-829863	USSR	Broad elliptic	Pinkish red	Green/ dark green broad	106.33	5.64	7.04	7.43	32.53
77	EC-829864	USSR	Broad elliptic	Red	Light green/dark green broad	104.33	3.58	6.87	7.49	35.47
78	EC-829865	Pakistan	Spherical	Pinkish red	Light green/ dark green broad	108.50	1.82	4.52	5.75	20.67
79	EC-829866	Pakistan	Broad elliptic	Pinkish red	Green/dark green very broad	108.67	1.69	3.93	5.12	37.67
80	EC-829867	Spain	Elliptic	Red	Dark green/ absent	100.83	1.88	4.08	6.99	53.17
81	EC-829868	Spain	Spherical	Pinkish red	Green/ dark green medium	87.00	4.07	6.50	7.59	21.60
82	EC-829870	China	Narrow elliptic	Orange	Green/ dark green very broad	84.17	3.60	7.22	7.95	25.83
83	EC-829871	China	Broad elliptic	Red	Light green/dark green medium	93.33	3.75	7.70	7.27	91.17
84	EC0829872	South Korea	Broad elliptic	Red	Light green/ dark green medium	91.67	2.35	4.79	9.75	56.93
85	EC-829873	South Korea	Broad elliptic	Red	Light green/diffused	88.83	2.64	5.29	6.80	55.63
86	EC-829874	South Korea	Spherical	Red	Light green/ dark green diffused	88.00	2.29	5.37	9.58	79.43

Table 1. Contd.

Code	Accession number	Origin	Fruit shape	Flesh colour	Rind colour/ stripe pattern	Days to maturity	Fruit weight (kg)	Yield/ vine (kg)	TSS (°Brix)	Lycopene content (µg/g)
87	EC-829875	South Korea	Spherical	Red	Light green/absent	89.50	2.72	5.65	10.42	82.77
88	EC-829878	Canada	Spherical	Pale white	Light green/ dark green medium	82.00	3.39	7.53	9.91	12.40
<i>Citrullus lanatus</i>			Fruit characteristics			Metric traits				
	Accession number	Origin	Fruit shape	Flesh colour		Days to maturity	Fruit weight (kg)	Yield /vine (kg)	TSS (°Brix)	Lycopene Content (µg/g)
47	EC-829820	USA	Broad elliptic	Red	Grey/green diffused	87.17	3.05	6.40	10.25	84.30
48	EC-829822	USA	Narrow elliptic	Red	Light green/dark green medium	90.83	3.93	7.49	9.10	68.73
49	EC-829823	USA	Elliptic	Red	Green/dark green narrow	85.83	3.49	7.39	9.45	73.17
50	EC-829824	USA	Narrow elliptic	Red	Green/dark green medium	92.50	4.69	7.74	8.94	60.63
51	EC-829825	USA	Elliptic	Red	Light green/dark green broad	90.33	3.40	7.27	8.53	63.87
52	EC-829826	USA	Elliptic	Red	Light green/ dark green broad	87.67	3.66	6.93	7.97	59.57
53	EC-829827	USA	Narrow elliptic	Pinkish red	Light green/ dark green broad	97.17	2.85	6.04	5.70	47.10
54	EC-829828	USA	Elliptic	Red	Yellow rind/ narrow stripes	86.00	2.01	4.86	9.70	69.63
55	EC-829829	USA	Elliptic	Red	Light green/dark green medium	97.67	3.65	7.80	8.63	54.37
56	EC-829832	USA	Broad elliptic	Red	Green/dark green narrow	89.67	2.86	5.53	9.62	71.53
57	EC-829833	USA	Elliptic	Red	Green/ dark green broad	91.33	3.13	6.64	6.78	56.43
58	EC-829836	USA	Elliptic	Pinkish red	Green/absent	97.33	1.32	3.07	6.60	54.03
59	EC-829838	USA	Narrow elliptic	Red	Light green/dark green broad	84.50	4.24	8.16	8.97	64.17
60	EC-829840	USA	Broad elliptic	Red	Green/dark green medium	97.50	7.15	8.48	8.53	58.07
61	EC-829841	Turkmenistan	Broad elliptic	Pinkish red	Green/diffused	105.50	3.23	6.27	5.65	35.33
L.S.D						2.86	0.31	0.48	0.71	4.58

F test was significant @ P< 0.00001)

*Accessions starting with IC and EC number are obtained from GRIN-USDA, Griffin, USA

WM- Accessions were collected from Uttar Pradesh, India

Jaipur, Jaiselmer, Barmer, Bikaner and Thar Manik from Rajasthan, India

Arka Manik and Arka Jyoti from Indian Institute of Horticultural Research, Bangalore, India

Table 2: Range and mean for different fruit traits for *Citrullus colocynthis*, Indian landraces and exotic germplasm

Group	Days to fruit maturity	Fruit weight (kg)	Fruit yield/vine (kg)	Total soluble solids (°Brix)	Lycopene content (µg/g)
<i>Colocynthis</i>					
Range (low-high)	106 to 122	.06-1.41	0.14-3.19	1.40-7.10	3.10-23.43
Mean	133	0.36	0.86		
Landraces					
Range (low-high)	82-107.5	1.29-4.62	3.57-7.34	3.77-10.08	14.33-67
Mean	94.26	2.67	5.51	6.84	37.69
Exotic germplasm					
Range (low-high)	82-108.67	0.93-7.15	2.50-8.48	4.15-10.85	12.40-93.37
Mean	95.22	3.22	6.13	7.68	51.69

Table 3: SSR primer, annealing temperature, alleles amplified, PIC value and heterozygosity

Sr no	SSR primer	Annealing temperature (°C)	Number of alleles	PIC value	Expected heterozygosity (H_e)	Observed heterozygosity (H_o)	Reference
1	MCPI-4	50.5	3	0.34	0.37	0.41	Joobeur et al (2006)
2	MCPI-5	52.0	8	0.73	0.55	0.61	
3	MCPI-16	51.2	9	0.84	0.65	0.72	
4	MCPI-17	49.4	3	0.52	0.60	0.69	
5	MCPI-20	51.5	2	0.31	0.32	0.34	
6	MCPI-23	51.0	5	0.71	0.54	0.65	
7	MCPI-29	51.0	2	0.33	0.23	0.18	
8	MCPI-32	51.0	5	0.65	0.69	0.79	
9	MCPI-33	51.5	3	0.48	0.37	0.47	
10	MCPI-39	52.0	3	0.35	0.38	0.48	
11	WMA-1	50.0	4	0.68	0.63	0.69	Hee-Jeong et al (2007)
12	WMA-2	51.0	3	0.42	0.46	0.65	
13	WMA-5	52.0	3	0.45	0.54	0.58	
14	WMA-6	52.5	2	0.37	0.29	0.23	
15	WMA-11	48.0	3	0.38	0.42	0.55	
16	WMA-15	50.0	7	0.72	0.76	0.88	
17	WMA-26	48.5	3	0.35	0.30	0.35	
18	BVWS-333	49.2	2	0.36	0.33	0.29	Zhang et al (2012)
19	BVWS-441	47.2	2	0.37	0.39	0.35	
20	BVWS-948	50.7	2	0.20	0.23	0.21	
21	BVWS-433	51.0	7	0.78	0.64	0.69	
22	BVWS-314	53.4	3	0.49	0.58	0.68	
23	BVWS-658	50.2	3	0.30	0.38	0.46	
24	BVWS-2048	52.0	4	0.54	0.49	0.58	
25	ASU-19	47.8	6	0.69	0.63	0.70	Levi et al (2006)
26	CLG-7996	48.5	2	0.11	0.21	0.15	
27	CLG-8218	44.8	3	0.24	0.26	0.38	
28	CLG-8288	45.0	4	0.51	0.36	0.44	
29	Cgb-4765	52.0	3	0.42	0.60	0.59	
30	EST-00674	55.0	2	0.33	0.41	0.36	Guerra-Sanz (2002)
31	AF032471.1	48.4	4	0.57	0.63	0.72	
32	EST-00691	48.0	3	0.52	0.38	0.41	
33	EST-00667	54.4	2	0.31	0.26	0.19	
	Total		120	15.37	14.88	16.55	
	Mean		3.63	0.46	0.45	0.50

Figure 1: Neighbour Joining (NJ) tree

Figure 2: Factorial correspondent analysis (FCA) plot

Factorial analysis: (Axes 1 / 2)