# Molecular analysis of genetic diversity in melon landraces (Cucumis melo L .) from Myanmar and their relationship with melon germplasm from East and South Asia 

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#### Abstract

Genetic diversity of Myanmar melon was evaluated by analysis of 27 RAPD markers and morphological characters using 41 accessions of melon landraces of which 36 accessions were small-seed type. The gene diversity was 0.239 , higher than for group Conomon from East Asia and equivalent to Indian melon populations. Melon accessions were classified into six major clusters. The largest cluster IV comprised mainly group Conomon which was closely related to cluster V consisting of mainly group Agrestis. Most of the accessions of group Cantalupensis were grouped into clusters II or VII and were distantly related to groups Conomon and Agrestis. The genetic relationship to melon accessions from neighboring countries was analyzed. The 24 accessions of


[^0]clusters IV and V were mostly clustered together with small-seed type melon of India, but the 14 accessions of clusters VI and VII were mostly clustered together with large-seed type melon of India. These results indicated that the genetic diversity of Indian melon is conserved in Myanmar. Genetic introgression among melon groups through spontaneous hybridization was also indicated and was considered important to maintain or increase the genetic diversity in Myanmar.

Keywords Cucumis melo - Genetic diversity . Genetic resources • Myanmar • RAPD • Seed size

## Introduction

Melon (Cucumis melo L.) is one of the important horticultural crops of the world. It is morphologically diverse: the size, shape, color of fruits and rind firmness vary greatly among melon cultivars and market types (Bates and Robinson 1995). The fruits are usually spherical or oval oblong and some are oblate or elongate. The fruit surfaces are smooth, some are deeply ridged and others are covered with corky (reticulate) netting. Vein tracks, also called sutures, are areas of longitudinal stripes on the fruit surface without netting. Naudin (1859) classified the different forms of melon into botanical varieties. Several reclassifications later Munger and Robinson (1991) classified melon into seven groups: Cantalupensis,

Indorus, Flexuosus, Conomon, Chito and Dudaim, Momordica and Agrestis. From their seed length, these melon groups can be classified into small-seed type ( $<9.0 \mathrm{~mm}$ ), including groups Conomon and Agrestis, and large-seed type ( $\geq 9.0 \mathrm{~mm}$ ), including groups Catalupensis and Indorus (Fujishita 1983; Akashi et al. 2002). The former two groups are assigned to subsp. agrestis with short hairs on the ovary, while the latter two groups are assigned to subsp. melo with long hairs on the ovary (Pitrat et al. 2000; Jeffrey 2001). In South and Southeast Asia, especially in Myanmar, both seed types are commonly cultivated and all groups, except Inodorus, are cultivated.

Tanaka et al. (2007) confirmed that groups Cantalupensis and Inodorus have sweet flesh and are cultivated mainly in Europe and USA, groups Momordica and Conomon have low sugar amounts and a smooth skin and are cultivated mainly in South and East Asia. These two geographical groups also differ in seed length: longer than 9.0 mm in Cantalupensis and Indorus and shorter than 8.5 mm in Conomon, but not in group Momordica (Fujishita 1983). Group Conomon, as defined by Munger and Robinson (1991), is divided into C. melo var. conomon Thunb. (group Conomon var. conomon) and C. melo var. makuwa Makino (group Conomon var. makuwa); these two varieties have been cultivated as different crops in Japan (Kitamura 1950). Group Conomon var. makuwa is also cultivated in Korea and China; its fruits have a smooth skin, are sweet and fragrant when fully ripened, and are eaten raw. Group Conomon var. conomon fruits have a smooth skin, are sometimes eaten raw, but are usually cooked or pickled; although they do not taste sweet, fully ripened fruits are slightly sweet and fragrant. Group Conomon is an important genetic resources for disease resistance, as reviewed by Akashi et al. (2002).

Genetic diversity is an important aspect of crop genetic resources, and can be determined by using morphological and molecular markers. Molecular markers based on DNA sequence polymorphism are independent of environmental conditions and show higher levels of polymorphism. Therefore, in the last decade molecular markers have been frequently used to analyze genetic diversity. Evaluation studies of genetic diversity in melon have used several types of molecular markers: restriction fragment length polymorphisms (RFLPs; Neuhausen 1992), isozymes (Akashi et al. 2002; McCreight et al. 2004), amplified
fragment length polymorphism (AFLPs; Garcia-Mas et al. 2000), random amplified polymorphic DNA (RAPD; Mliki et al. 2001; Lopez-Sese et al. 2003; Staub et al. 2004; Nakata et al. 2005; Dhillon et al. 2007; Tanaka et al. 2007) and simple sequence repeats (SSRs; Monforte et al. 2003; Dhillon et al. 2007). All these markers have been equally informative in establishing genetic relationships between melon genotypes.

Akashi et al. (2002), McCreight et al. (2004) and Tanaka et al. (2007) evaluated genetic variation in East and South Asian melon by analysis of isozyme and RAPD polymorphism; they showed that Indian melon was rich in genetic diversity compared with East Asian melon. Akashi et al. (2002) also showed that Indian cultivated melon had varied seed length ( $4.0-13.0 \mathrm{~mm}$ ) and indicated its importance as novel genetic resources that can be used in various breeding programs. However, less attention have been paid to East and South Asian melon, and thus extensive studies on the genetic structure of East and South Asian melon is required.

Myanmar is the second largest country in Southeast Asia; it shares a border with Bangladesh and India in the west and China, Laos, and Thailand in the east, and thus is an important area for the movement of people, culture, goods and plants from India to East Asia by skirting the southern slope of Himalayas. These areas are also considered important for domestication of Cucurbitaceae crops, because various wild species of Cucurbitaceae crops, such as cucumber, bitter gourd, luffa, grow wild in these areas. Especially, wild taxa of Cucumis, such as $C$. sativus var. hardwickii (Royle) Gabaev and C. hystrix Charkr., grow in only these areas. Farmers grow different melon cultivars in dry and rainy seasons in northeast India (Kato et al. 2006). The weedy melon classified as group Agrestis grows in Myanmar and is not cultivated by farmers, but the mature fruit is used as a vegetable when mainly fruit flesh is cooked and the seeds are discarded. These facts indicate that Myanmar melon has diverse morphological and physiological traits. Because of its diverse uses, melon is becoming an important horticultural crop and one of the future potential horticultural crops in Myanmar (MAS 2006). Tanaka et al. (2007) analyzed genetic variation in Myanmar melon using five accessions of local melon. However, a comprehensive analysis of genetic variation in local melon
landraces has not been done. To increase the usefulness of different types of melon germplasm for melon breeders and growers, characterization of morphological, biochemical and molecular aspects of Myanmar melon is required.

Thus, in this study we used RAPD makers (1) to assess melon genetic diversity together with fruit morphological data, and (2) to determine the genetic relationship with melon landraces from neighboring countries by incorporating the results of Tanaka et al. (2007). These analyses provided insight into the horticultural worth of Myanmar melons, which is essential for the organization and conservation of melon genetic resources and their future use.

## Materials and methods

Plant materials
To evaluate the genetic diversity of Myanmar melon, 31 accessions of melon landraces collected from seven States and Divisions were analyzed (Fig. 1). Table 1 summarizes details of the plant materials. Seeds of two melon accessions were provided by the National Institute of Vegetable and Tea Science (NIVTS), Japan. RAPD analysis was done using a single plant from each accession. Ten accessions of Myanmar melon from the United States Department of Agriculture (USDA) were analyzed by Tanaka et al. (2007): Thus a total of 41 accessions and their RAPD data were used in this study. One accession of Cucumis sativus var. hardwickii was also included as an outgroup for comparison.

## DNA extraction

Seeds were sown on filter paper and were grown at $26^{\circ} \mathrm{C}$ in a 16 h light to 8 h dark cycle at light intensity $46.5 \mu \mathrm{MS}^{-1} \mathrm{~m}^{-2}$. Cotyledons of 10 -day-old seedlings were ground individually in liquid nitrogen, and total DNA was extracted by using the procedure of Murray and Thompson (1980) with minor modifications.

RAPD analysis
Eighteen random primers selected for their ability to detect polymorphism and for their stability in PCR


Fig. 1 Map of Myanmar. Boxes indicate seven states where melon accessions were collected
amplification were used for RAPD analysis as described by Tanaka et al. (2007; Table 2). PCR amplification was done in a $10 \mu \mathrm{l}$ mixture containing 50 ng genomic DNA, $1 \mu \mathrm{l}$ PCR buffer (Sigma ${ }^{\circledR}$, USA: 10 mM Tris- $\mathrm{HCl}(\mathrm{pH} 8.3), 50 \mathrm{mM} \mathrm{KCl})$, $2.5 \mathrm{mM} \mathrm{MgCl} 2,0.25 \mathrm{U}$ Taq polymerase (Pharmacia for primer A07 and Sigma for others), 0.1 mM dNTP and $0.5 \mu \mathrm{M}$ primer by using an i-Cycler (Bio-Rad, USA), and PC-707 (ASTEC, Japan). An initial denaturing step at $95^{\circ} \mathrm{C}$ for $3 \mathrm{~min}, 40 \mathrm{PCR}$ cycles at $94^{\circ} \mathrm{C}$ for $1 \mathrm{~min}, 40^{\circ} \mathrm{C}$ for 2 min , and $72^{\circ} \mathrm{C}$ for 2 min were done, and then a final extension at $72^{\circ} \mathrm{C}$ for 5 min . After amplification, samples underwent electrophoresis on $1.5 \%$ agarose gel (Takara, Japan) at constant voltage 100 V (Mupid-2, Cosmo Bio, Japan). Then the PCR products were visualized with ethidium bromide staining and their polymorphisms were evaluated.

Table 1 Melon accessions analyzed in this study

| Acccession no. ${ }^{\text {a }}$ | Country of origin | Collected area | Group | Seed source ${ }^{\text {b }}$ | Seed | Cluster ${ }^{\text {c }}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| CSH1 | India | - | C. sativus var. hardwickii | Okayama Univ. | - | I |
| US96-1 | Myanmar | Mandalay | Cantalupensis | USDA (PI 200813) | Large | II |
| US96-2 | Myanmar | Mandalay | Cantalupensis | USDA (PI 200813) | Large | II |
| My238 | Myanmar | Yangon | Conomon | Okayama Univ. | Small | III |
| Cho4 | Myanmar | Mandalay/Taungthar | Momordica | VFRDC | Small | IV a |
| Cho6 | Myanmar | Yangon | Momordica | VFRDC | Small | IV b |
| EA30 | Myanmar | Mandalay | Momordica | NIVTS (650057) | Small | IV b |
| OU284-1 | Myanmar | Mandalay | Conomon | USDA (PI 200814) | Small | IV b |
| OU181-1 | Myanmar | Mandalay | Conomon | USDA (PI 200819) | Small | IV b |
| OU181-2 | Myanmar | Mandalay/Myingyan | Conomon | USDA (PI 200819) | Small | IV b |
| M25 | Myanmar | Mandalay/Myingyan | Cantalupensis | VFRDC | Small | IV b |
| Cho2 | Myanmar | Mandalay/Taungthar | Conomon | VFRDC | Small | IV b |
| My23 | Myanmar | Mandalay/Tat kone | Conomon | Okayama Univ. | Small | IV b |
| Cho7 | Myanmar | Yangon | Momordica | VFRDC | Small | IV b |
| My209 | Myanmar | Rakhaine/Thandwe | Conomon | Okayama Univ. | Small | IV b |
| My154 | Myanmar | Magway/Pakokhu | Agrestis | Okayama Univ. | Small | IV c |
| OU284-2 | Myanmar | Mandalay | Conomon | USDA (PI 200814) | Small | IV c |
| OU290-1 | Myanmar | Kachin | Conomon | USDA (PI 200817) | Small | IV c |
| M16 | Myanmar | Mandalay/Taungthar | Conomon | VFRDC | Small | IV c |
| OU290-2 | Myanmar | Kachin | Conomon | USDA (PI 200817) | Small | IV c |
| My140 | Myanmar | Magway/Pakokhu | Agrestis | Okayama Univ. | Small | V |
| My206 | Myanmar | Rakhaine/Thandwe | Conomon | Okayama Univ. | Small | V |
| My207 | Myanmar | Rakhaine/Thandwe | Conomon | Okayama Univ. | Small | V |
| My133 | Myanmar | Mandalay/Popa | Agrestis | Okayama Univ. | Small | V |
| M27 | Myanmar | Arrawaddy/Athoke | Agrestis | VFRDC | Small | V |
| M28 | Myanmar | Mon/Mlemine | Agrestis | VFRDC | Small | V |
| My169 | Myanmar | Magway | Agrestis | Okayama Univ. | Small | V |
| M29 | Myanmar | Mon/Molemine | Conomon | VFRDC | Small | V |
| M26 | Myanmar | Arrawaddy/Athoke | Conomon | VFRDC | Small | VI |
| M21 | Myanmar | Mandalay | Conomon | VFRDC | Small | VI |
| M6 | Myanmar | Yangon | Conomon | VFRDC | Large | VI |
| My171 | Myanmar | Magway | Agrestis | Okayama Univ. | Small | VI |
| M7 | Myanmar | Mandalay/Myingyan | Conomon | VFRDC | Small | VI |
| Chol | Myanmar | Mandalay/Taungthar | Momordica | VFRDC | Small | VI |
| M15 | Myanmar | Mandalay/Taungthar | Conomon | VFRDC | Small | VII |
| OU176-1 | Myanmar | Mandalay | Conomon | USDA (PI 200816) | Small | VII |
| OU176-2 | Myanmar | Mandalay | Conomon | USDA (PI 200816) | Small | VII |
| Cho3 | Myanmar | Mandalay/Taungthar | Conomon | VFRDC | Small | VII |
| M1 | Myanmar | Mandalay/Myingyan | Cantalupensis | VFRDC | Small | VII |
| Cho5 | Myanmar | Rakhaine/Thandwe | Momordica | VFRDC | Large | VII |
| EA65 | Myanmar | Unknown | Cantalupensis | NIVTS (940261) | Large | VII |
| My115 | Myanmar | Mandalay/Myingyan | Conomon | Okayama Univ. | Small | VII |

[^1]Table 2 Eighteen random primers used in this study and the size of polymorphic bands
${ }^{\text {a }}$ The size of marker bands is indicated in base pairs. The number of accessions amplified each marker band is shown in the parenthesis

| Primer no. | Sequence $\left(5^{\prime} \rightarrow 3^{\prime}\right)$ | Polymorphic bands |  |
| :--- | :--- | :--- | :--- |
|  |  | Number | Size $^{\mathrm{a}}$ |
| A07 | GATGGATTTGGG | 2 | $1,353(39), 872(27)$ |
| A20 | TTGCCGGGACCA | 2 | $1,100(39), 800(4)$ |
| A22 | TCCAAGCTACCA | 1 | $1,520(1)$ |
| A23 | AAGTGGTGGTAT | 1 | $1,200(7)$ |
| A26 | GGTGAGGATTCA | 1 | $1,400(3)$ |
| A31 | GGTGGTGGTATC | 1 | $800(28)$ |
| A39 | CCTGAGGTAACT | 1 | $2,027(7)$ |
| A41 | TGGTACGGTATA | 3 | $1,353(3), 1,020(9), 930(33)$ |
| A57 | ATCATTGGCGAA | 1 | $800(38)$ |
| B15 | CCTTGGCATCGG | 1 | $600(22)$ |
| B32 | ATCATCGTACGT | 2 | $900(22), 700(40)$ |
| B68 | CACACTCGTCAT | 1 | $1,078(36)$ |
| B71 | GGACCTCCATCG | 1 | $1,220(16)$ |
| B84 | CTTATGGATCCG | 3 | $700(3), 600(34), 550(4)$ |
| B86 | ATCGAGCGAACG | 2 | $1,500(0), 1,350(35)$ |
| B96 | GTGAAGACTATG | 2 | $850(1), 750(39)$ |
| B99 | TTCTGCTCGAAA | 1 | $1,400(10)$ |
| C00 | GAGTTGTATGCG | 1 | $1,350(12)$ |
| Total |  | 27 |  |

Evaluation of morphological traits
Qualitative and quantitative traits of fruit and seeds were asessed by growing them in a greenhouse. Table 3 summarizes nine traits assessed in this study: (1) length and (2) width of seeds were measured for a representative five seeds; (3) fruit weight, (4) fruit diameter and (5) fruit height were measured for each fruit; (6) exocarp skin was classified as having stripes and the color as white, green, yellow and orange with or without spots; (7) mesocarp flesh color and (8) placenta color were classified as green, white, orange and yellow; (9) soluble solid content was measured by Brix grade.

Akashi et al. (2002) showed that seed length of melon generally differed between groups and seed length was a good character for a rough classification into each group of $C$. melo. Because most Myanmar melons have not been systematically classified into varieties, they were classified into small-seed type ( $<9.0 \mathrm{~mm}$ ) and largeseed type ( $\geq 9.0 \mathrm{~mm}$ ) based on seed length (Table 3).

## Data analysis

Each marker fragment was scored as 1 for present and 0 for absent. From RAPD data of 42 accessions
the genetic similarity (GS) among accessions was calculated as described by Apostol et al. (1993), and the gene diversity (D) within each group and genetic distance (GD) were calculated as described by Weir (1996) and Nei (1972), respectively. A dendrogram was constructed by using the Phylip program by using the unweighted pair group method with arithmetic mean (UPGMA) method. Principal coordinate analysis (PCO; Gower 1966) based on the genetic similarity matrix was done to show multiple dimensions of each group and the accessions in a scatterplot.

## Results

Classification based on fruit characters and seed length

Most accessions produced non-netted fruits, while five accessions produced netted fruit. Fruit weight varied significantly among melon accessions from 10 g to $1,400 \mathrm{~g}$ per fruit (Table 3). Seven accessions classified as group Agrestis were collected as weedy melon characterized by small-fruits of weight 40 g or

Table 3 Seed size and fruit characters of 41 accessions of Myanmar melon

| Accession no. | Seed Size |  |  | Fruit |  |  | Color of ${ }^{\text {a }}$ |  |  | Brix <br> ${ }^{\circ}$ ) | Cluster ${ }^{\text {b }}$ |  | Group |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Class | Length (mm) | Width (mm) | Weight (g) | Height (cm) | Diameter <br> (cm) | Skin | Flesh | Placenta |  | 41 accs. | $153$ <br> accs. |  |
| US96-1 | Large | 9.0 | 4.0 | 600 | 13.7 | 8.0 | WG | O | W | 9.0 | II | II' | Cantalupensis |
| US96-2 | Large | 9.0 | 4.0 | 600 | 17.5 | 8.0 | GO | O | O | 3.8 | II | III' | Cantalupensis |
| My238 | Small | 6.0 | 3.0 | 200 | 8.5 | 5.0 | YO | W | W | 11.0 | III | $\mathrm{V}^{\prime}$ | Conomon |
| Cho4 | Small | 7.0 | 3.0 | 125 | 6.5 | 6.0 | W (spotted) | WG | WG | 4.0 | IV | VII' | Momordica |
| Cho6 | Small | 7.0 | 3.0 | 500 | 17.5 | 7.0 | GY | WG | W | 2.8 | IV | VIII' | Momordica |
| EA30 | Small | 8.0 | 5.0 | 800 | 16.5 | 10.0 | Y | G | O | 6.5 | IV | VIII ${ }^{\prime}$ | Momordica |
| OU284-1 | Small | 5.0 | 2.0 | 100 | 7.5 | 6.0 | YG (stripe) | G | W | 6.2 | IV | VIII ${ }^{\prime}$ | Conomon |
| OU181-1 | Small | 5.0 | 2.0 | 600 | 13.7 | 8.0 | WG | O | W | 9.0 | IV | VIII' | Conomon |
| OU181-2 | Small | 5.0 | 2.0 | 100 | 8.0 | 4.5 | O | GW | O | 6.5 | IV | VIII' | Conomon |
| M25 | Small | 8.3 | 3.8 | 500 | 16.5 | 8.0 | $\begin{aligned} & \text { Y (brown } \\ & \text { spots) } \end{aligned}$ | - | - | - | IV | VIII' | Cantalupensis |
| Cho2 | Small | 6.0 | 3.0 | 200 | 10.0 | 6.5 | G | GY | Y | 4.0 | IV | VIII ${ }^{\prime}$ | Conomon |
| My23 | Small | 5.5 | 2.5 | 200 | 11.0 | 6.0 | GY | W | Y | 8.8 | IV | VIII ${ }^{\prime}$ | Conomon |
| Cho7 | Small | 8.0 | 3.5 | 700 | 16.7 | 10.0 | G | W | W | 5.2 | IV | VIII' | Momordica |
| My209 | Small | 7.5 | 2.5 | 1,300 | 21.5 | 9.5 | Y (stripe) | O | O | 6.5 | IV | VIII ${ }^{\prime}$ | Conomon |
| My154 | Small | 5.5 | 2.5 | 10 | 3.0 | 2.5 | GW | G | G | 5.5 | IV | VIII' | Agrestis |
| OU284-2 | Small | 5.0 | 2.0 | 100 | 7.5 | 6.0 | YG (stripe) | G | W | 6.2 | IV | VIII' | Conomon |
| OU290-1 | Small | 4.0 | 3.0 | 400 | 16.0 | 7.5 | GY | WO | W | 4.2 | IV | VIII' | Conomon |
| M16 | Small | 6.0 | 3.1 | 450 | 13.0 | 7.0 | GW (stripe) | W | YW | 5.8 | IV | VIII ${ }^{\prime}$ | Conomon |
| OU290-2 | Small | 4.0 | 3.0 | 400 | 16.0 | 7.5 | GY | WO | W | 4.2 | IV | VIII ${ }^{\prime}$ | Conomon |
| My140 | Small | 5.0 | 2.0 | 30 | 3.0 | 2.5 | G (stripe) | W | W | - | V | VIII' | Agrestis |
| My206 | Small | 8.0 | 3.5 | 60 | 8.5 | 3.8 | G | W | W | 7.0 | V | VIII' | Conomon |
| My207 | Small | 8.0 | 3.5 | 400 | 12.5 | 6.5 | G | WO | O | 4.8 | V | VIII' | Conomon |
| My133 | Small | 4.0 | 3.0 | 40 | 3.5 | 3.5 | YG | W | W | - | V | VIII' | Agrestis |
| M27 | Small | 6.0 | 3.1 | 33 | 2.8 | 2.5 | G (stripe) | W | W | - | V | VIII' | Agrestis |
| M28 | Small | 6.7 | 3.2 | 29 | 2.6 | 2.4 | G (stripe) | W | W | - | V | III' | Agrestis |
| My169 | Small | 5.0 | 2.5 | 25 | 5.0 | 3.0 | G (stripe) | W | W | 7.0 | V | VIII' | Agrestis |
| M29 | Small | 7.1 | 2.9 | 600 | 12.0 | 9.0 | YG | W | W | - | V | III' | Conomon |
| M26 | Small | 5.3 | 2.7 | 29 | 2.5 | 2.4 | G (stripe) | W | W | - | VI | VIII ${ }^{\prime}$ | Conomon |
| M21 | Small | 6.9 | 3.0 | 54 | 5.7 | 4.3 | W (stripe) | WY | WY | 7.6 | VI | VIII' | Conomon |
| M6 | Large | 9.3 | 4.4 | 220 | 9.3 | 5.6 | Y | Y | OW | 10.6 | VI | IV ${ }^{\prime}$ | Conomon |
| My171 | Small | 5.0 | 2.0 | 20 | 4.0 | 3.0 | GY | W | WY | 4.2 | VI | IV ${ }^{\prime}$ | Agrestis |
| M7 | Small | 8.0 | 3.1 | 180 | 7.3 | 4.2 | GY | YW | YW | 5.8 | VI | IV ${ }^{\prime}$ | Conomon |
| Chol | Small | 8.0 | 3.0 | 500 | 15.0 | 8.0 | GY | W | W | 6.0 | VI | IV ${ }^{\prime}$ | Momordica |
| M15 | Small | 6.1 | 3.2 | 600 | 10.0 | 7.0 | W | YW | WY | 7.7 | VII | IX ${ }^{\prime}$ | Conomon |
| OU176-1 | Small | 6.0 | 3.0 | 1,400 | 10.5 | 28.0 | GO | O | O | 6.0 | VII | III' | Conomon |
| OU176-2 | Small | 6.0 | 3.0 | 1,400 | 10.5 | 28.0 | GO | O | O | 6.0 | VII | III' | Conomon |
| Cho3 | Small | 6.0 | 3.0 | 200 | 8.5 | 4.0 | G | WG | WG | 10.0 | VII | IV ${ }^{\prime}$ | Conomon |
| M1 | Small | 6.7 | 2.6 | 230 | 8.7 | 4.6 | Y | OY | OY | 10.2 | VII | IV ${ }^{\prime}$ | Cantalupensis |
| Cho5 | Large | 10.0 | 4.0 | 400 | 17.0 | 7.0 | Y | W | W | 5.2 | VII | IV ${ }^{\prime}$ | Momordica |
| EA65 | Large | 10.0 | 4.0 | 600 | 9.5 | 10.5 | WG | WG | O | 6.8 | VII | III' | Cantalupensis |
| My115 | Small | 6.0 | 3.0 | 400 | 15.0 | 8.0 | GO | W | WO | 6.0 | VII | IV ${ }^{\prime}$ | Conomon |

-, not tested
${ }^{\text {a }}$ Colors are indicated by: $G$ green; $O$ orange; $Y$ yellow; $W$ white
${ }^{\mathrm{b}}$ The results of this study (41 accs.) and the combined analysis (153 accs.) including data of Tanaka et al. (2007)

Fig. 2 Photos of typical accessions of four groups of Cucumis melo analyzed in this study

less (Fig. 2). Six accessions had a cracked exocarp skin at the fully matured stage and their flesh texture was rather powdery. Therefore, these accessions were classified as group Momordica. Five accessions that produced weakly netted fruits were classified as group Cantalupensis, because the shelf life of their fruits is not long (data not shown). The other accessions had smooth and thin exocarp skin and small yellow seeds, and thus were classified as group Conomon. The brix values were $2.8^{\circ}-11.0^{\circ}$ among 41 accessions, while they were $4.2^{\circ}-7.0^{\circ}$ and $2.8^{\circ}-6.5^{\circ}$ in groups, Agrestis and Momordica, respectively, and were $3.8^{\circ}-10.2^{\circ}$ and from $4.0^{\circ}$ to $11.0^{\circ}$ in groups Cantalupensis and Conomon, respectively.

Seed length of Myanmar melon was $4.0-10.0 \mathrm{~mm}$. According to criteria of Akashi et al. (2002), five accessions in group Cantalupensis (US96-1, US96-2, and EA65), Momordica (Cho5), and Conomon (M6) were classified as large-seed type, and the remainder were classified as small-seed type. In this study, in the seven accessions of group Agrestis, the seed length was $4.0-6.7 \mathrm{~mm}$ and they were classified as
small-seed type. However, in the six accessions of group Momordica and the five accessions of group Cantalupensis, the seed length was $7.0-10.0 \mathrm{~mm}$ and $6.7-10.0 \mathrm{~mm}$, respectively. In the 22 accessions of group Conomon, the seed length was $4.0-9.3 \mathrm{~mm}$, and all except M6 were classified as small-seed type.

RAPD profile, genetic diversity and genetic distance

As shown in Table 2, eighteen primers produced 27 polymorphic bands which ranged in size from 550 to 2027 bp . The average number of marker bands was 1.5 per primer. The most polymorphic band was generated by primer B32, and a marker band of 900 bp was amplified in 22 accessions among 41 melon accessions. On the contrary, the least polymorphic band was generated by primer B86, and all accessions lacked a marker band of $1,500 \mathrm{bp}$. The frequency of two marker bands, A39-2027 and C001350, was significantly different among large-seed type and small-seed type ( $x^{2}=15.93,7.08, d f=1$ ),
respectively, and was also different among melon groups. The frequency of a marker band B15-600 significantly differed in the four melon groups Conomon, Agrestis, Momordica and Cantalupensis $\left(x^{2}=13.63, d f=3\right)$, and all accessions of group Agrestis failed to amplify this band. The inter-group difference was also significant for four additional markers, A07-1353 ( $x^{2}=8.67$ ), A07-872 ( $\left.x^{2}=7.89\right)$, A39-2027 ( $x^{2}=8.34$ ), and C00-1350 $\left(x^{2}=11.23\right)$.

Gene diversity (D) was 0.239 in the 41 accessions of Myanmar melon: 0.231 in large-seed type and 0.218 in small-seed type (Table 4). These values were higher than those of group Conomon from China, Korea and Japan and equivalent to those of Indian melon populations. Gene diversity (D) was also different in the four groups: 0.157 in group Agrestis, 0.158 in group Momordica, 0.222 in group Conomon and 0.255 in group Cantalupensis.

The genetic distance (GD) between the 41 accessions of melon was calculated from the presence or absence of 27 RAPD markers. The average GD was 0.25 and ranged from 0.04 to 0.52 (Table 5). The
largest GD was recorded between M15 and US96-1. The smallest GD was between ten pairs of accessions.

Genetic relationship among melon landraces in Myanmar

To determine the genetic relationship among melon landraces, a dendrogram was constructed based on GD values by using the UPGMA method. The 41 accessions of melon were grouped into six major clusters in addition to Cluster I formed by the outgroup accession, C. sativus var. hardwickii (Fig. 3). Cluster II contained two accessions, both from USDA accession PI 200813, which was collected in Mandalay in April, 1952. They were classified as group Cantalupensis of the large-seed type and with orange fruit flesh. Accession My238 alone formed Cluster III; it was collected in Yangon in 2000, and its fruit looks quite similar to the famous Japanese cultivar 'Kinpyo', having yellowish orange skin and white flesh (Fig. 2). Cluster IV formed the largest group comprising 16 accessions among which

Table 4 Gene diversity and number of melon plants classified into 11 clusters in each population based on 153 accessions

| Country | Variety/seed size ${ }^{\text {a }}$ | Area | No. of accessions | Cluster |  |  |  |  |  |  |  |  |  |  | Gene diversity |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  | I' | II' | III' | IV' | $\mathrm{V}^{\prime}$ |  | VII' | VIII' | IX ${ }^{\prime}$ | $\mathrm{X}^{\prime}$ | $\mathrm{XI}^{\prime}$ |  |
| India | Large-seed type | West | 6 | 2 | - | - | 4 | - | - | - | - | - | - | - | 0.253 |
|  |  | North | 8 | - | - | 3 | 3 | - | - | - | - | - | - | 2 | 0.278 |
|  |  | Center | 6 | - | 2 | 1 | 1 | - | - | - | 2 | - | - | - | 0.294 |
|  |  | South | 10 | - |  | 5 | 4 | - | - | 1 |  | - | - | - | 0.273 |
|  |  | East | 12 | - | 5 | 1 | 1 | - | 1 | 1 | 2 | - | - | 1 | 0.305 |
|  | Small-seed type | West | 6 | - |  | 1 | 1 | - | - | - | 3 | - | - | 1 | 0.239 |
|  |  | North | 4 | - |  | 2 | - | - | - | - | - | - | 2 | - | 0.218 |
|  |  | Center | 10 | - | 1 | - | - | - | - | 3 | 5 | 1 | - | - | 0.224 |
|  |  | South | 4 | - | - | - | - | - | - | 1 | 3 | - | - | - | 0.148 |
|  |  | East | 8 | - | 1 | - | - | 1 | 1 | - | 3 | - | - | 2 | 0.278 |
| Myanmar | Large-seed type |  | 5 | - | 1 | 1 | 2 | - | - | - | 1 | - | - | - | 0.231 |
|  | Small-seed type |  | 36 | - | - | 5 | 6 | 1 | - | 1 | 22 | 1 | - | - | 0.218 |
| China | var. makuwa |  | 14 | - | - | - | - | 13 | - | 1 | - | - | - | - | 0.104 |
|  | var. conomon |  | 7 | - | - | - | - | 7 | - | - | - | - | - | - | 0.076 |
| Korea | var. makuwa |  | 6 | - | - | - | - | 6 | - | - | - | - | - | - | 0.091 |
| Japan | var. makuwa |  | 6 | - | - | - | - | 6 | - | - | - | - | - | - | 0.119 |
|  | var. conomon |  | 5 | - | - | - | - | 5 | - | - | - | - | - | - | 0.071 |
| Total |  |  | 153 | 2 | 10 | 19 | 22 | 39 | 2 | 8 | 41 | 2 | 2 | 6 |  |

[^2]Table 5 Genetic distance between 41 melon landraces from Myanmar based on the frequency of 27 RAPD markers



Fig. 3 Genetic relationship between 41 accessions of melon landraces shown by UPGMA cluster analysis based on GD. CSH1; C. sativus var. hardwickii used as an outgroup

10 accessions were group Conomon; all 16 accessions were small-seed type. They were further separated into three subclusters: (1) one accession from Mandalay (Cho4), (2) 10 accessions mainly from Mandalay area (7 from Mandalay, 2 from Yangon, 1 from Rakhaine), and (3) five accessions from Mandalay and other areas (2 from Kachin, 2 from Mandalay, 1 from Magway). Cluster V contained eight accessions from various areas other than Mandalay, among which five accessions were group Agrestis (Table 1), and their fruit weight was less than 60 g in six of the eight accessions. Cluster VI comprised six accessions, including one accession each of groups Momordica and Agrestis. Cluster VII comprised eight accessions mainly from Mandalay, including two accessions of group Cantalupensis and one accession of group Momordica.

Principal co-ordinate analysis (PCO) was done to visualize the relationship among the 41 accessions


Fig. 4 Distribution of the first two principal co-ordinates of 41 accessions of melon landraces from Myanmar. CSH1; C. sativus var. hardwickii used as an outgroup
and the outgroup (Fig. 4). The first two principal components explained $29.6 \%$ of the total variance, 19.3 and $10.2 \%$, respectively. Groups Agrestis and Conomon were separated from other groups of melon, mainly by the second PCO axis. One accession, C. sativus var. hardwickii, was in the right upper quadrant, distantly related to the group Agrestis accessions, which were grouped closely in the same quadrant. Most melon accessions of cluster IV were grouped closely in the right lower quadrant, and those of cluster VII were grouped in the left lower quadrant. Cluster VI accessions appeared in the upper two quadrants. Therefore, we concluded that grouping by using the UPGMA method could be well reproduced on a PCO plot.

## Discussion

Cultivation of the $\mathrm{F}_{1}$ hybrid of sweet and netted melon is now popular in the Mandalay area. These hybrid cultivars were introduced mainly from Taiwan and replaced the local landraces of melon. Therefore, we collected genetic resources of Myanmar melon and evaluated their genetic diversity. Local landraces of Myanmar melon comprised groups Conomon, Agrestis, Momordica, and Cantalupensis, among which group Conomon was the most popular (Fig. 2; Table 3).

The length of melon seed generally differs between groups: seeds of group Cantalupensis are
longer than 9 mm (large-seed type) and seeds of groups Conomon and Agrestis are $5-8.5 \mathrm{~mm}$ (smallseed type; Fujishita 1983; Akashi et al. 2002). Group Momordica is intermediate having both large- and small-seed types. In this study, among melon accessions from Myanmar, all group Conomon accessions were small-seed type, except the large-seed M6 (Table 3). Their seed size was similar to that of group Agrestis, and fruit weight was over 100 g (average about 370 g ) in most accessions. Group Conomon is widely grown in Myanmar and is used for salads or is cooked with meat for main dishes. Group Momordica is commonly found in South and Southeast Asia. Its seed length was $7-10 \mathrm{~mm}$, comprising both small- and large-seed types, as in India (Kato et al. 2006). In northeast India, local farmers grow "Phut" type (group Momordica) in the dry season and distinguish it from small-seed type grown in the rainy season (Kato et al. 2006), but in Myanmar they are grown and consumed as the same type of melon. Melon accessions of group Agrestis are characterized by producing many small fruits and are called weedy melon (Fujishita 1983) and "Kasit" in Myanmar. Detailed data of production and consumption of "Kasit" do not exist, but "Kasit" is produced and consumed almost all over the country year round. Local farmers generally remove "Kasit" from their fields in the early stage of crop growth, but leave it growing in the later stage. After harvesting the crop, they pick "Kasit" fruits in open fields to use as a vegetable.

In this study, melon accessions from Myanmar were classified into six major clusters by RAPD analysis (Fig. 3). The largest cluster IV comprised mainly group Conomon and was separated into three subclusters (Table 1). Cluster V was closely related to cluster IV, and comprised mainly group Agrestis. Most accessions of group Cantalupensis were grouped into cluster II or VII, and were distantly related to accessions of clusters IV and V. From these results, we tried to explain the genetic relationship among geographical groups within Myanmar. However, only two to four accessions were collected from most states, except from Mandalay. Therefore, more samples should be collected from the other states. Akashi et al. (2002) showed interesting results about the origin of weedy melon based on isozyme polymorphism. They found that melon accessions of group Agrestis
from India and Japan are distantly related, but they are closely related to cultivated melon with small seeds in these countries. They strongly suggested genetic introgression between weedy melon (group Agrestis) and small-seed type cultivated melon in each area, because melon is generally open-pollinated by bees. The results of our study agree well with their conclusion.

Because RAPD is more polymorphic than isozymes, groups Conomon and Agrestis that showed the same isozyme type were separated by RAPD analysis. However, some accessions of these two groups were intermixed in clusters IV and V (Table 3). Four accessions of group Momordica, and one accession of group Cantalupensis (M25) were grouped in cluster IV together with groups Conomon and Agrestis. These results show that the classification into melon groups mainly based on morphological characters dose not necessarily correspond to the genetic relationship shown by molecular analysis and may suggest genetic introgression among melon groups because of their outcrossing nature. An example of spontaneous hybridization within groups was observed for group Conomon accession M29. We cultivated ten plants of M29 using seeds collected from one fruit. The fruit size differed among these plants, at $60-1,400 \mathrm{~g}$. Among various factors that determine the genetic composition of plant populations, the mating system is the most influential. High polymorphism levels of the magnitude observed in this study for C. melo have been reported in outcrossing species, but predominantly selfing or clonal species or both generally show much higher proportions (45-80\%) of monomorphic loci (Zhivotovsky 1999). Similarly, relatively low differentiation among groups and the genetic diversity within groups are characteristic for predominantly outcrossing species, but this relationship for inbreeding species tends to be high (Nybom 2004).

Tanaka et al. (in preparation) estimated the proportion of small-seed type melon in various areas of the world from 100 seed weight in the GRIN database (USDA-ARS, http://www.ars-grin.gov/ $\mathrm{npgs} /$. The small-seed type was $6.2 \%$ in Europe, USA and Russia and was $3.1 \%$ in West and Central Asia. Its frequency increased from West Asia towards the east from $51.4 \%$ in South Asia to $62.2 \%$ in East Asia. In another study (Akashi et al. 2002), the
frequency of small-seed type increases markedly in areas east of the Middle East at $67.6 \%$ in South Asia, $73.3 \%$ in Southeast Asia and $100 \%$ in East Asia. Therefore, the high frequency of the small-seed type in the accessions of this study ( $87.8 \%$ ) reflects the tendency in this area.

Random amplified polymorphic DNA data of this study were compared with those of Tanaka et al. (2007) to know the genetic relationship with melon populations of neighboring countries. Gene diversity (D) of Myanmar melon was 0.231 in large-seed type and 0.218 in small-seed type (Table 4). These D values were as high as those in Indian melon populations (average $=0.251$ ), and were much higher than in East Asian melon populations (average $=0.092$ ). Therefore, genetic diversity may not have decreased with transmission of melon from northeast India to Myanmar, but severe selection must have occurred with transmission from Myanmar to the east.

The genetic relationship among 153 melon accessions [31 in this study and 122 from Tanaka et al. (2007)] was analyzed by cluster analysis. They were grouped into 11 clusters; Table 4 shows the number of accessions assigned to each cluster. Although the dendrogram of these 153 accessions is not shown, six clusters ( $\mathrm{V}^{\prime}-\mathrm{X}^{\prime}$ ) formed one large group and three clusters ( $\mathrm{II}^{\prime}-\mathrm{IV}^{\prime}$ ) formed another large group indicating clear genetic differentiation between small-seed type (Clusters $\mathrm{V}^{\prime}-\mathrm{X}^{\prime}$ ) and large-seed type (Clusters $\mathrm{II}^{\prime}-\mathrm{IV}^{\prime}$ ), as also shown by Akashi et al. (2002), Yashiro et al. (2005), and Tanaka et al. (2007). Among melon accessions from Myanmar, 24 accessions of clusters IV and V (from 41 accessions) were mostly grouped in cluster VIII' (from 153 accessions) together with small-seed type accessions of India, with the exception of three accessions (Cho4, M28, and M29; Table 3). Fourteen accessions of clusters VI and VII were mostly grouped in clusters III' and IV' together with large-seed type accessions of India, with the exception of three accessions (M15, M21, and M26; Table 3). This result indicated that genetic diversity in Indian melon was conserved also in Myanmar.

Regarding the differentiation and establishment of group Conomon var. makuwa and var. conomon, Tanaka et al. (2007) suggested that accessions PI 210542 and PI 124112, both from east India, could be the primitive type. Myanmar accession My238 of
cluster III was grouped in cluster $\mathrm{V}^{\prime}$ together with accessions of group Conomon from East Asia and PI 210542 (Table 3). Therefore, accession My238 might be also the primitive type of group Conomon in East Asia. However, its fruit character is quite similar to the famous Japanese cultivar 'Kinpyo' having a yellowish orange skin and white flesh (Fig. 2). My238 accession formed cluster III as a single accession, indicating My238 distantly related with other Myanmar accessions (Fig. 3). Therefore, further study is necessary to confirm if My238 is the primitive type of group Conomon or it is introduced from East Asia.

However, a striking difference was detected when the clustering pattern was compared between largeand small-seed types. In India, especially in east India, large-seed type and small-seed type were mostly separated into clusters $\mathrm{II}^{\prime}-\mathrm{IV}^{\prime}$ and clusters $\mathrm{V}^{\prime}-\mathrm{X}^{\prime}$, respectively (Table 4). Nearly one-third of the small-seed type accessions from Myanmar were grouped in the "large-seed type clusters $\mathrm{II}^{\prime}-\mathrm{IV}^{\prime}$ " indicating the presence of inter-group hybrids produced in the farmer's field but they were not eliminated from the farmer's field and used as usual. The presence of inter-group hybrids should be important to maintain or increase genetic diversity in Myanmar.

This study provided a general idea of the genetic diversity of Myanmar melon. The genetically diverse melon resources constitute raw materials that can potentially contribute to improve various traits of interest. Information based on systematic observations and evaluations of these resources is still very limited in Myanmar. However, even in this study, the melon accessions studied did not cover various agroecological zones of Myanmar. Therefore, germplasm collection of Myanmar melon should be increased to cover whole areas of Myanmar and should be analyzed from various aspects to understand the genetic diversity of Myanmar melon.

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## References

Akashi Y, Fukuda N, Wako T, Masuda M, Kato K (2002) Genetic variation and phylogenetic relationships in East and South Asian melons Cucumis melo L., based on the analysis of five isozymes. Euphytica 125:385-396. doi: 10.1023/A:1016086206423

Apostol BL, Black WCIV, Miller BR, Reiter P, Beaty BJ (1993) Estimation of the number of full sibling families at an oviposition site using RAPD-PCR markers: applications to the mosquito Aedes aegypti. Theor Appl Genet 86:991-1000. doi:10.1007/BF00211052
Bates DM, Robinson RW (1995) Cucumber, melons and watermelons, Cucumis and Citrullus (Cucurbitaceae). In: Smartt J, Simmonds NW (eds) Evolution of crop plants, vol 10158. Wiley, New York, pp 89-111
Dhillon NPS, Ranjana R, Singh K, Eduardo I, Monforte AJ, Pitrat M, Dhillon NK, Singh PP (2007) Diversity among landraces of Indian snapmelon (Cucumis melo var. momordica). Genet Resour Crop Evol 54:1267-1283. doi: 10.1007/s10722-006-9108-2

Fujishita N (1983) Genetic diversity and phylogenetic differentiation in melon. Curr Top Plant Breed 24:3-21 in Japanese
Garcia-Mas J, Oliver M, Gomez-Paniagua H, de Vicente MC (2000) Comparing AFLP, RAPD and RFLP markers for measuring genetic diversity in melon. Theor Appl Genet 101:860-864. doi:10.1007/s001220051553
Gower JC (1966) Some distance properties of latent root and vector methods used in multivariate analysis. Biometrika 53:325-338
Jeffrey C (2001) Cucurbitaceae. In: Hanelt P, Institute of Plant Genetics and Crop Plant Research (eds) Mansfeld's encyclopedia on agricultural and horticultural crops. Springer, Berlin, pp 1510-1557
Kato K, Yoshino H, Matsuura S, Akashi Y, Tanaka K (2006) Cucurbitaceae crop. In: Takeda K (ed) Genetic assay and study of crop germplasm in and around China, 3rd edn. Okayama University, Okayama, pp 69-85
Kitamura S (1950) Notes on Cucumis of far East. Acta Phytotax Geobot 14:41-44
Lopez-Sese AI, Staub JE, Gomez-Gullamon ML (2003) Genetic anysis of Spanish melon (Cucumis melo L.) germplasm using a standardized molecular marker array and reference accessions. Theor Appl Genet 108:41-52. doi:10.1007/s00122-003-1404-z
McCreight JD, Staub JE, Lopez-Sese A, Chung S (2004) Isozyme variation in Indian and Chinese melon (Cucumis melo L.) germplasm collections. J Am Soc Hortic Sci 129:811-818
Ministry of Agricultural Services (MAS) (2006) Annual report of horticulture. MAS, Myanmar

Mliki A, Staub JE, Sun Z, Ghorbel A (2001) Genetic diversity in melon (Cucumis melo L.): an evaluation of African germplasm. Genet Resour Crop Evol 48:587-597. doi: 10.1023/A:1013840517032

Monforte AJ, Garcia-Mas J, Arus P (2003) Genetic variability in melon based on microsatellite variation. Plant Breed 122:153-157. doi:10.1046/j.1439-0523.2003.00848.x
Munger HM, Robinson RW (1991) Nomenclature of Cucumis melo L. Cucurbit Genet Coop Rep 14:43-44
Murray GC, Thompson WF (1980) Rapid isolation of high molecular weight DNA. Nucleic Acids Res 8:4321-4325. doi:10.1093/nar/8.19.4321
Nakata E, Staub JE, Lopez-Sese AI, Katzir N (2005) Genetic diversity of Japanese melon cultivars (Cucumis melo L.) as assessed by random amplified polymorphic DNA and simple sequence repeat markers. Genet Resour Crop Evol 52:405-419. doi:10.1007/s10722-005-2258-9
Naudin C (1859) Especes et des varietes du genra cucumis. Sci Nat 11:5-87
Nei M (1972) Genetic distance between populations. Am Nat 106:283-292. doi:10.1086/282771
Neuhausen SL (1992) Evaluation of restriction fragment length polymorphism in Cucumis melo. Theor Appl Genet 83:379-384. doi:10.1007/BF00224286
Nybom H (2004) Comparison of different nuclear DNA makers for estimating intraspecific genetic diversity in plants. Mol Ecol 13:1143-1155. doi:10.1111/j.1365-294X.2004. 02141.x

Pitrat M, Hanelt P, Hammer K (2000) Some comments on intraspecific classification of cultivars of melon. Proc Cucurbitaceae 2000:29-36
Staub JE, Lopez-Sese AI, Fanourakis N (2004) Diversity among melon landraces (Cucumis melo L.) from Greece and their genetic relationships with other melon germplasm of diverse origins. Euphytica 136:151-166. doi: 10.1023/B:EUPH. 0000030667.63614. bd

Tanaka K, Nishitani A, Akashi Y, Nishida H, Yoshino H, Kato K (2007) Molecular characterization of South and East Asian melon, Cucumis melo L., and the origin of group Conomon var. makuwa and var. conomon revealed by RAPD analysis. Euphytica 153:233-247. doi:10.1007/ s10681-006-9259-4
Weir BS (1996) Genetic data analysis II. Sinauer Associate Inc. Publisher, Massachusetts
Yashiro K, Iwata H, Akashi Y, Tomita K, Kuzuya M, Tsumura Y, Kato K (2005) Genetic relationship among East and South Asian melon (Cucumis melo L.) revealed by AFLP analysis. Breed Sci 55:197-206. doi:10.1270/jsbbs.55.197
Zhivotovsky LA (1999) Estimating population structure in diploids with multilocus dominant DNA makers. Mol Ecol 8:907-913. doi:10.1046/j.1365-294x.1999.00620.x


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[^1]:    ${ }^{\text {a }}$ Melon accessions are listed in the order of the dendrogram of Fig. 3
    ${ }^{\mathrm{b}}$ NIVTS National Institute of Vegetable and Tea Science, Japan; USDA United States Department of Agriculture, USA; VFRDC Vegetable and Fruit Research and Development Center, Myanmar. Ten accessions from USDA have been analyzed by Tanaka et al. (2007)
    ${ }^{c}$ The results of this study

[^2]:    ${ }^{\text {a }}$ var. makuwa and var. conomon indicate Group Conomon var. makuwa and var. conomon, respectively

