AFRICAN LEAFY VEGETABLES AGROMORPHOLOGICAL CHARACTERIZATION AND PARTICIPATORY PLANT BREEDING ON FOUR SPECIES

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Modern agriculture, characterized by the use of new homogenous varieties with high yield potential (instead of traditional ones) results in the erosion of genetic diversity. The collection and characterization of local germplasm become necessary. Thus the 64 accessions collected during our three surveys and the 18 accessions of ISRA/CDH genebank have been characterized at the Horticultural Developpement Center (CDH) of Senegal. They were 48 accessions of *Hibiscus sabdariffa*, 19 accessions of *Vigna unguiculata*, 9 accessions of *Amaranthus spp*, and 6 accessions of *Moringa oleifera*. Descriptors of IPGRI & AVRDC-GRSU were used. This characterization shows the existence of an intra-specific diversity of the four species studied. Seeds (or genes) can also spread up to 800 km based on the variables used. In average, it is noted that 35% of the accessions were found twice. The analysis of the specific diversity of the accessions in the visited localities using genetic diversity Richness Index has revealed that 57 % of the accessions are subjected to genetic erosion. The agro-morphological characterization should be completed by other studies using biochemical and molecular tools in order to verify the above results. Using more accessions for each species and others characterization methods could help set up a core collection which is the best way for germplasm conservation. In order to increase the adoption rate of the new cultivars of the traditional leaf-vegetables, a participatory selection, on the new cultivars based on users' criteria preferences, was carried out at CDH research station, with heavy involvement of the users (producers, consumers and traders). The scoring matrix has been used. The cultivars were ranked by the users, adoption rate should be maximal if these cultivars keep the same performance in the farmer's fields. These new cultivars are now diseminated using on farm conservation in pilot sites (3) in the country and at research station.

Key words: *leafy vegetables, characterization, diversity, participatory, cultivar*.

INTRODUCTION

Phytogenetic resources preservations are essential for food security and sustainable agriculture. However, plant genetic diversity is dangerously affected. Wild plants are disappearing with the development of the habitation. In the field, farmers abandon the traditional varieties for the new improved ones (CGRAI, 1994). It is worrisome to remark that the traditional knowledge on the grown and wild species is quickly disappearing. Most of this knowledge is kept by the women producers but, is not widely known by the general public. With the changes in the societies, youngsters do not usually acquire the experiences of the elders. The modernization of agriculture, whereby new homogenous varieties with high yield potential are used, leads to narrowing of the genetic diversity (Guarino et al. 1995). Therefore, the collection of diverse local ecotypes becomes a condition for ensuring food security. In general, the genetic material obtained by collecting is minute (CHADHA *& al.*, 2000). The conservation needs (*in situ* and *ex situ*) and later uses (plant improvement, food, pharmacognosy etc.) require seeds increase and better phenotypic characteristics of the material. That's why the regeneration or multiplication and the characterization become necessary steps for further use of the plants. This seed multiplication for getting enough material will allow a sound duplication of the seeds for

preservation. Part of the genetic material is on midterm storage, another part is the working collection (short term storage) and a third part is on long term storage (national and international gene bank) (CHADHA & *al*., 2000).

During our three surveys, we collected species of cowpea *Vigna unguiculata* (L.) WALP), Hibiscus (*Hibiscus sabdariffa* L.), amaranths (*Amaranthus* L. spp) and nebeday (*Moringa oleifera* LAM. Thus, 9 varieties of amaranths, 6 of nebeday, 48 of hibiscus and 19 of cowpeas have been planted at the ISRA-CDH research station.

To increase the level of adoption of the new cultivars of leafy vegetable selected a participatory breeding session was carried out at ISRA-CDH research station. This selection was done with a strong participation of farmers, traders and consumers of these traditional leafy vegetables with also the main stakeholders of research such Non Governmental Organizations (NGOs).

1. Methodology

Experiments were conducted at the Senegalese Institute of Agricultural Research/ Horticultural Development Center (ISRA-CDH) Research Station. A completely randomized design with single blocs without replications has been used (IPGRI, 2001). Sowing was done manually on Mai 23 2003 after a fallow. All the accessions: hibiscus, amaranths, nebeday and cowpeas have been cultivated according to the recommendations in the technical guidelines (BENIEST, (1987); ISRA-CNRA, (1987); VAN DE PLAS, (1980) et ROUSSEL & *al*., (1995)). Characterization keys for nebeday, cowpea and amaranths of the Asian Vegetable Research and Development Center (AVRDC, 2002 and CHADHA, 2000) have been used. A simplified key version of BRICAGE (1978) has been used for hibiscus. The most studied varieties have been used as reference or check and are Vcdh, Vimto, and Koor (bissap), ISRABambey1, ISRABambey2 and ISRABambey3 (cowpea), AVRDC1 et AVRDC2 (amaranth) and for nebeday the accession MAVRDC.
Different parameters of floral and vegetative development, leaf and seed production were studied. These parameters were measured on the thinned plants as well on those remaining on the plot. The measurements were made on a sample of 5 accessions for hibiscus and amaranth, 10 for cowpea and 3 for nebeday. 11 variables have been measured on the plants of the 48 accessions of hibiscus, 16 variables on 19 accessions of cowpea, 29 variables on 9 accessions of amaranths and 32 variables on 6 accessions of nebeday.
Dry matter measurement for all accessions of the 4 species was done on a 20 gram sample of wet leaves dried on a piece of paper in the laboratory in the shade for 14 days. Measurements on the length and the width were done using a sample of 5 leaves chosen randomly. Measurements of the length and diameter of the pods of cowpea and nebeday were done on 10 plants. A foot – ruler was used to measure the diameter and a graduated stick of 50 cm long was used to measure the length.
Total biomass was measured using a portable scale and sample less than 600 g were weighed using an electronic scale (SARTORIUS of 600 g maximum weight). Among the 11 variables measured on hibiscus, 9 were used in the ANOVA, the remaining 2 (fresh leaves (20 g) and drying time under the shade (14 days)) were the same for all accessions. The 9 variables are: the height and the ground cover, total biomass per plant, total fresh weight, the mean length and width of the leaf, the ratio length on width ((L/l<1 (1), from 1 to 1.9 (2), from 2 to 2.9 (3) ≥ (4)), dry weight of the leaves and dry matter.
The 16 variables measured on cowpea are total biomass and the mean diameter per plant, the mean height and ground cover, growth habit ((erect (1) and spreading (2)), fresh leaves per plant, main stem color, mean number of nodules per plant, total nodules of 5 plants, sample weight of the fresh leaves, weight of the leaves dried under the shade, drying time, dry matter content, emergence rate in the field. 120 days after sowing, the measured variables were plants height,

mean length and width of the limb, the length of the lateral inflorescence, leaf color, leaf shape, thickness of the axillary leaves, branch development indixes, hairiness and color of the stems, leaf margin shape, types of venation, petiole pigmentation, sex, inflorescence density and leaf color.
32 variables have been measured on nebeday 266 days after sowing: they are: plant growth habit, the length and width of the leaves, color taste before cooking of the leaves, length and width of the leaflets, petiole color, flower color and smell, color and mean length and width of the pods, mean number of seeds per pods, powder conversion coefficient of the leaves. Variables measured at 41 days after sowing are: stem diameter, height and number of leaves per plant, dry matter content and drying time of the leaves. 78 days after sowing, parameters which were measured concern: main stem diameter, plant height, flowering rate, number of lateral branches, 5 leaves weight and the weight of the limb, total weight and number of the leaves per plant, total consumable leaf weight and harvest index, dry matter content and finally drying time.
For all species, all data were stored in excel, percent dry matter content data were transformed using Arc sinus √%. Data were analyzed using multivariate statistics R1.6.1 (GENTLEMAN & IHAKA, 2002). A quantidendrogramm was made from the hibiscus data and daisydendrogams were made using data from cowpea, amaranths and nebeday. Inter-specific genetic diversity of the four species has been studied in all the sites. Richness genetic diversity index has been used. It defines the number of accessions found in the site and combines the richness of the species in the site and the probability of finding the accession or the species. To increase the level of adoption by farmers of the cultivars selected, a participatory selection sesssion was organized at ISRA CDH reserach station. The method of matric notation or Scoring matrix (CERAAS, 2002) was used for the sesssion. All the cultivars for each especies are ranked by users.

2. RESULTS

2.1. AGROMORPHPLOGOCAL CHARACTERIZATION

Hibiscus (*Hibiscus sabdariffa* L.*)*

There are four groups for Hibiscus: Group 1 is composed of 21 accessions, group 2 has 9, group 3 has 3 and group 4 has 18. The number of duplication is respectively 7, 4, 1 and 7 for group 1, 2, 3, and 4 which represents 37 % overall (19/51). There are 3 duplications out of the 9 accessions collected in one site; the 16 remaining duplications are from accessions collected from distant sites reaching sometimes 800 km. The high duplication rate shows the importance of the exchange of seeds between the producers within a village or at the national level. There is an intra-specific variability between some accessions as shown by 63 % dissimilarity between them (Figure 1).
Four out of 16 variables have been chosen for comparison of the different hibiscus groups. They are plant height, fresh leaf weight, leaf width and dry matter content. Choices of the four variables were explained by the fact that height is generally correlated with productivity. Leaf fresh weight and dry matter content are productivity criteria and leaf width is a criterion of producer's preference. The big leaves are the ultimate choice of the producers.
For plant height, the 2 groups contain accessions with smaller plants than the three others. These latter ones seem to have plants of nearly equal height and the same trend exist for the width of the leaves. Dry matter content does not seem different between the two groups. Fresh leaf weight was bigger for the group 3 followed by group 2 and 4, while group 1 has the lowest production
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Richness diversity index shows that Diaobe and Matam markets present more genetic diversity, meaning the chance of collecting samples during the next survey are greater and these markets are followed by Tamba and Missira (Figure 2). The high diversity in these markets could be explained by the fact that they represent a place of seed exchange and located in a relatively high rainfall zone where people produce and consume leaves as vegetables (Diouf & *al*., 1999). In these areas, most of the producers contacted, buy their seeds in the markets. A less important fact is that Diaobe marketplace has a national and international and even a sub regional magnitude. Of the agro-morphological characteristics which are for the most time the same as the preference criteria of the producers (large green leaves, productivity etc.), plants N°7 (or L7) and plant N°28 (or L28) of the group 1 and plant N° 24 (or L24) of group 4 have been selected and will be multiplied in the future.

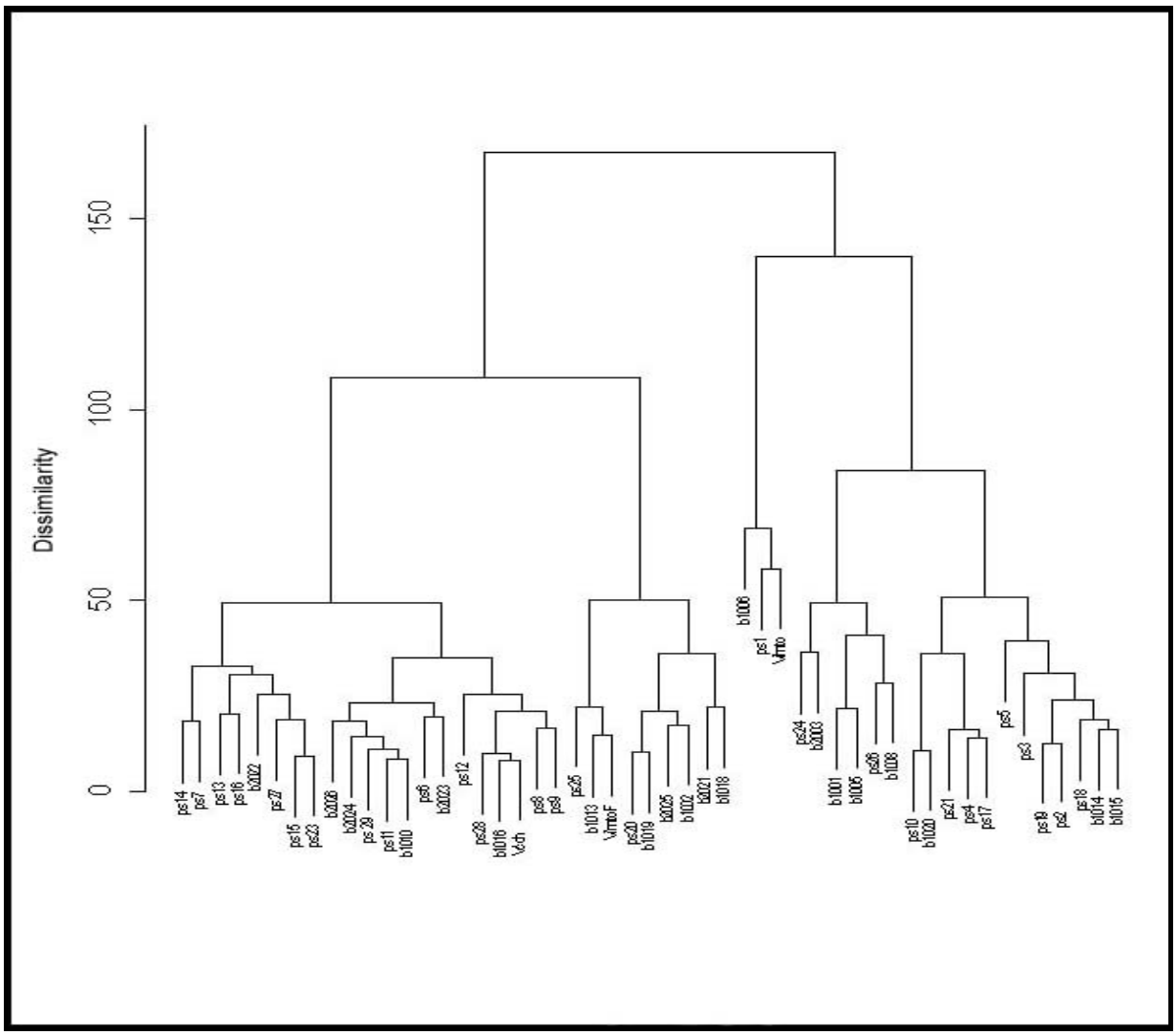

Figure1: Daisydendrogram of roselle accessions

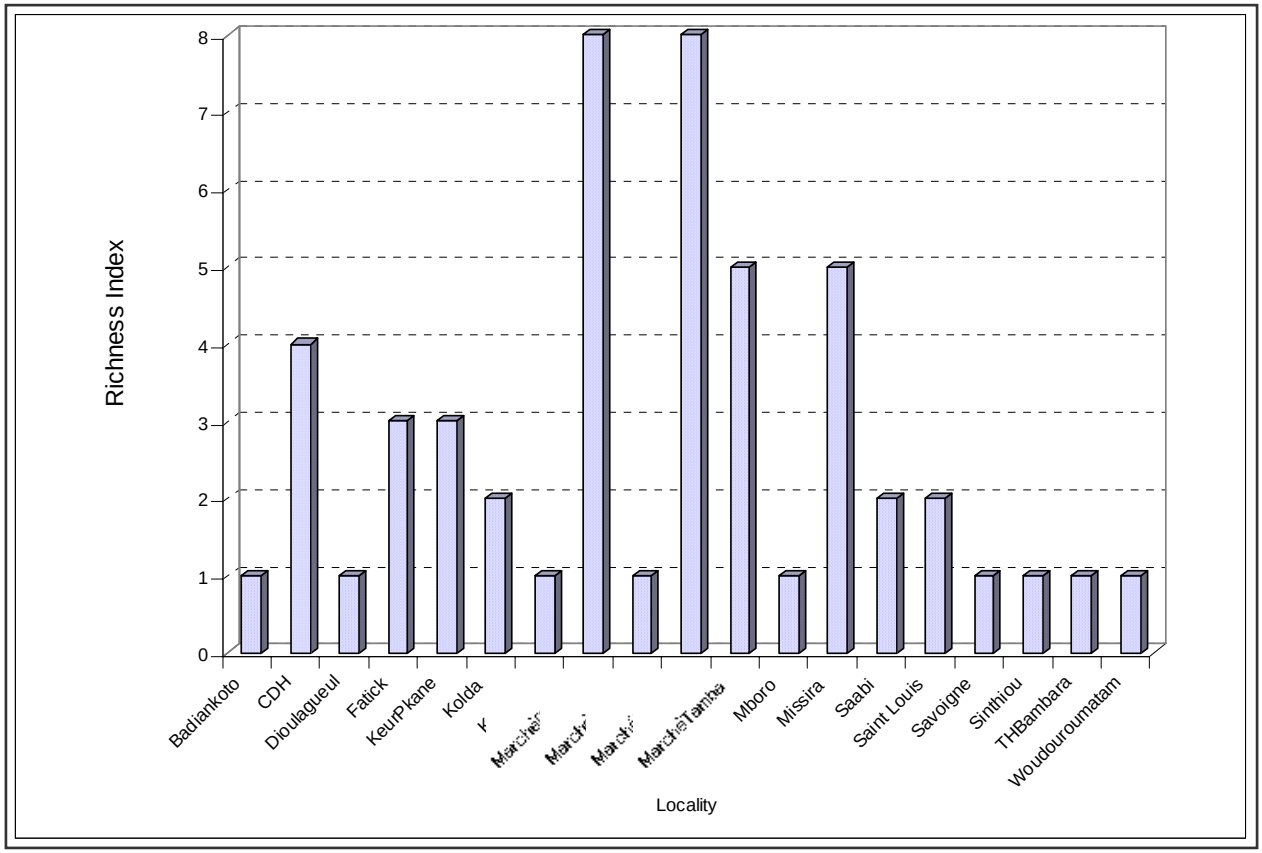

Figure 2: Degree of intra-specific diversity of roselle in the different localities.

Cowpea (*Vigna unguiculata* (L.) WALP):

There are 8 groups or classes and the number of duplications represents 37 % ((7/19)*100=37%) of the accessions (Figure 3). The group 8, followed by groups 3 and 4 has a relatively higher mean height than the others. For fresh leaf production, group 2 is better than the others followed by groups 3, 4 and 5 with group 6 having the lowest production. There was no apparent difference in dry matter content and in leaf width. For the number of fixing nitrogen nodules per plant, groups 3 and 5 followed by groups 2 and 6 seem to have a superior nodulating capacity, while groups 1, 4 and 7 have the lowest nodulating level. With regards to the preferences of the women farmers (erect growth habit, green leaves, lateness etc.) the 7 selected plants are: N 111 (or Kolda 11), N183 (Yelingara GF), Mougne, Bambey 21, N191 (ou fatick 9), N1011 (or Kolda 1) and N17 (or YelingaraPF).

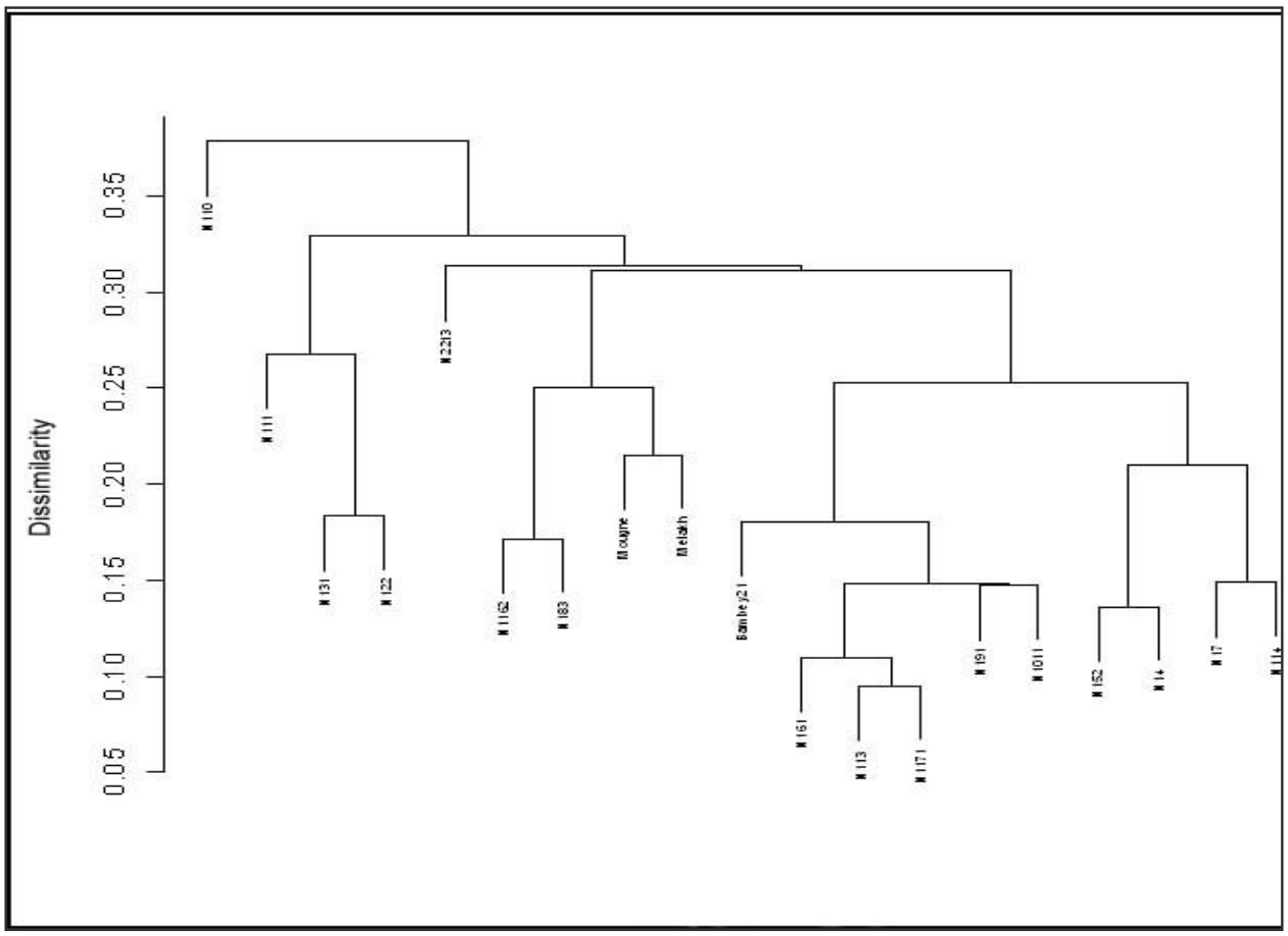

Figure 3 : Daisydendrogram of cowpea accessions

Amaranths (*Amaranthus* L. spp):

There are 6 groups or classes and the number of duplications represent 33 % ((3/9)*100=33%) of the accessions (Figure 4). The graph shows an interesting aspect of the seed management by the farmers. Knowing that Afaro Tamba1, Afaro Tamba 2 and Afaro Tamba 3 are from different samples taken in the same area where three women farmers have neighbouring plots; one can say that they grow a mixture of traditional varieties and sometimes a mixture of species. Moreover, there is an active seed exchange between the women producers in that some duplications (Afaro2/AhamadiNoba and Adioulagueul/Amissira) are composed of accessions taken from distant locations that can reach 100 km. However, it is true that more sophisticated molecular studies could confirm the existence of these duplications.

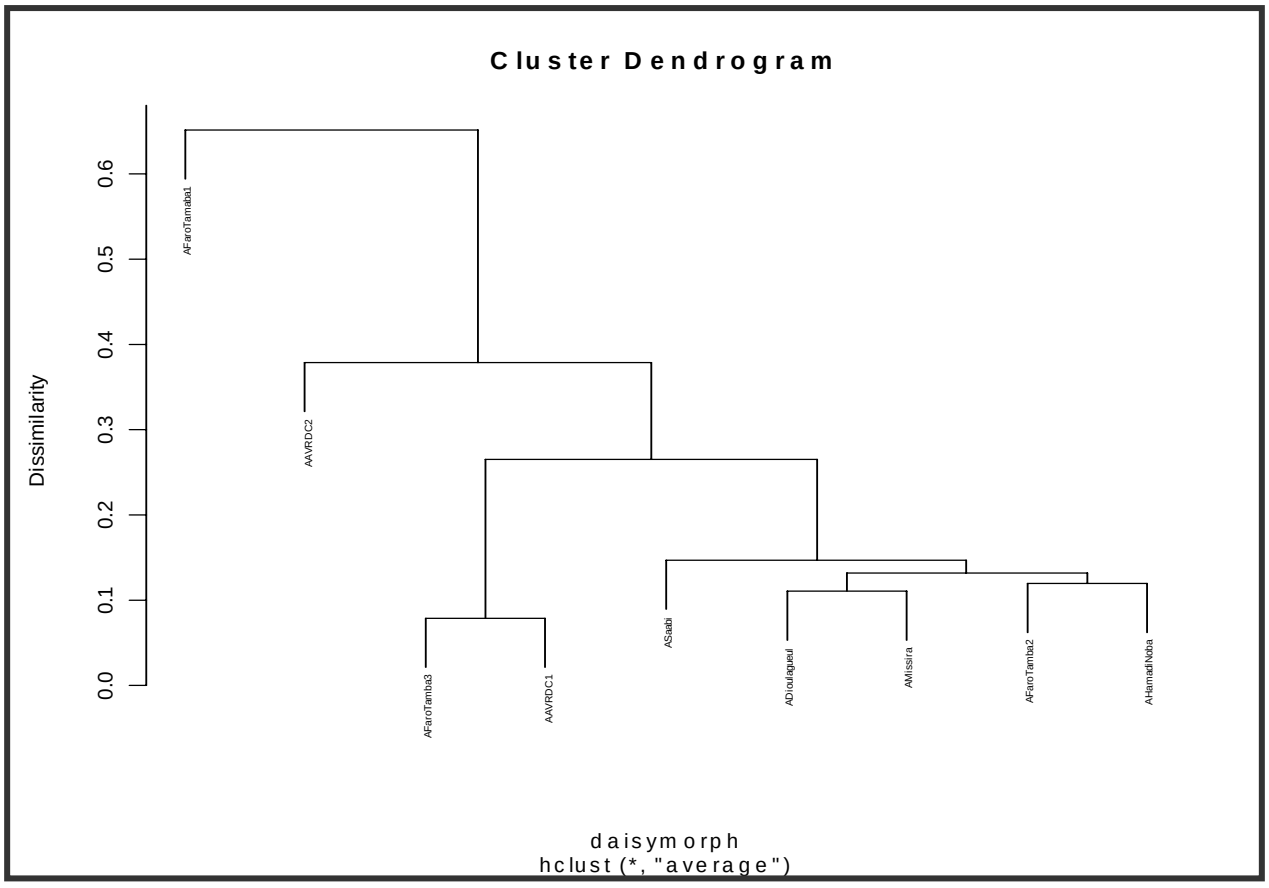

Figure 4: Daisydendrogram of Amaranth accessions.

Nebeday (*Moringa oleifera* Lam);

There are four groups or classes and the number of duplications represent 33% ((2/6)*100=33%) of the accessions (Figure 5). MAVRDC is completely different from the other accessions. The duplications (MCDH/MKothiary and MTamba/MDiourbel) contain accessions taken from distance reaching 600 km. These observations lead to two hypotheses: Either the seeds have been exchanged or the measured variables could not discriminate between accessions. Only molecular studies could clarify this issue. The accessions MAVRDC, MCDH and MKothiary have been selected based on the women farmers criteria and therefore will be studied later.

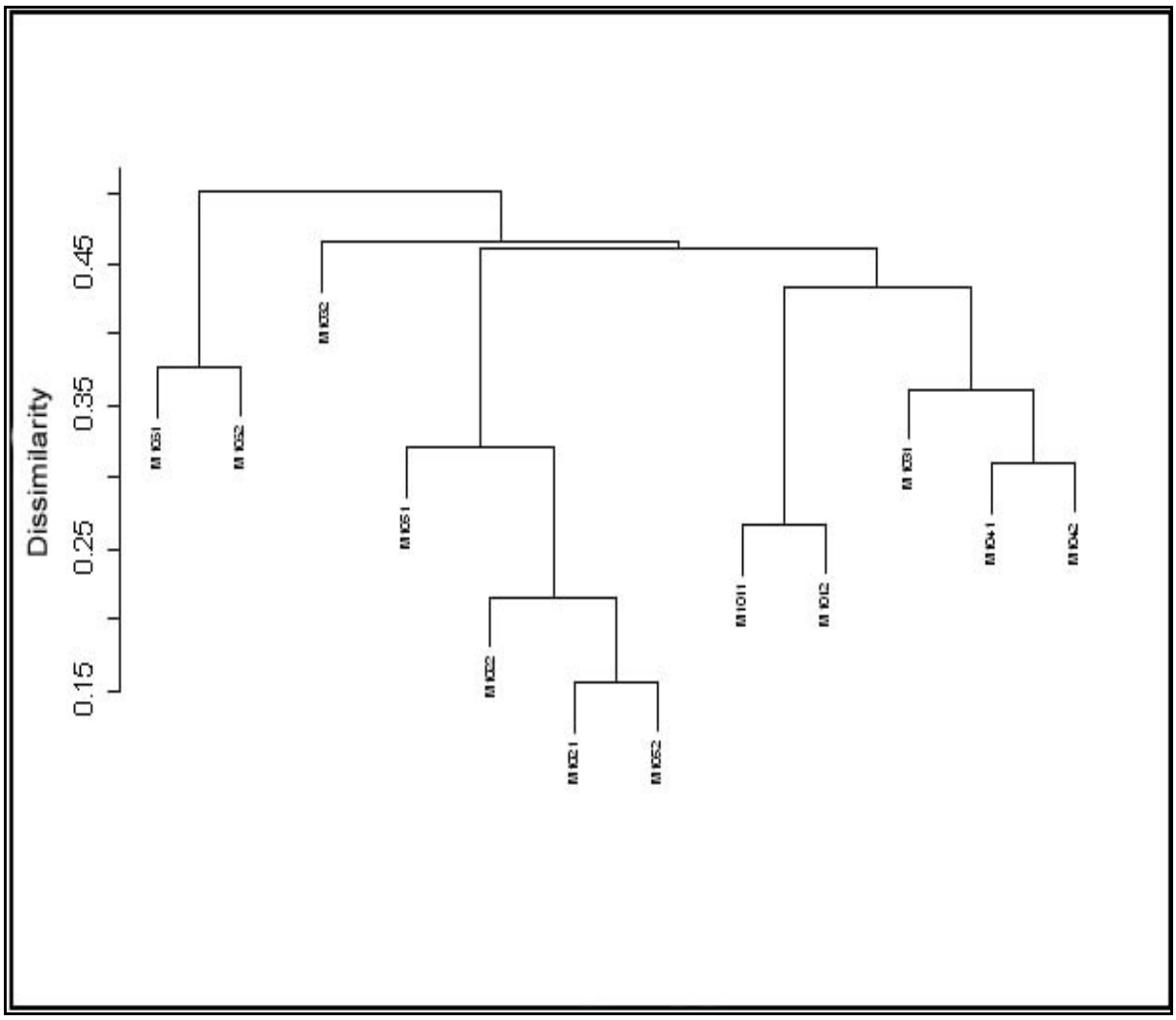

Figure 5: Daisydendrogram of moringa accessions

Inter-specific diversity of the localities.

Richness index results allowed the study the inter-specific diversity level of the visited localities. By analyzing the graph, 55% of the localities are highly subjected to genetic erosion, 27 % are under medium erosion and 18 % are under no genetic erosion risk (Figure 6).

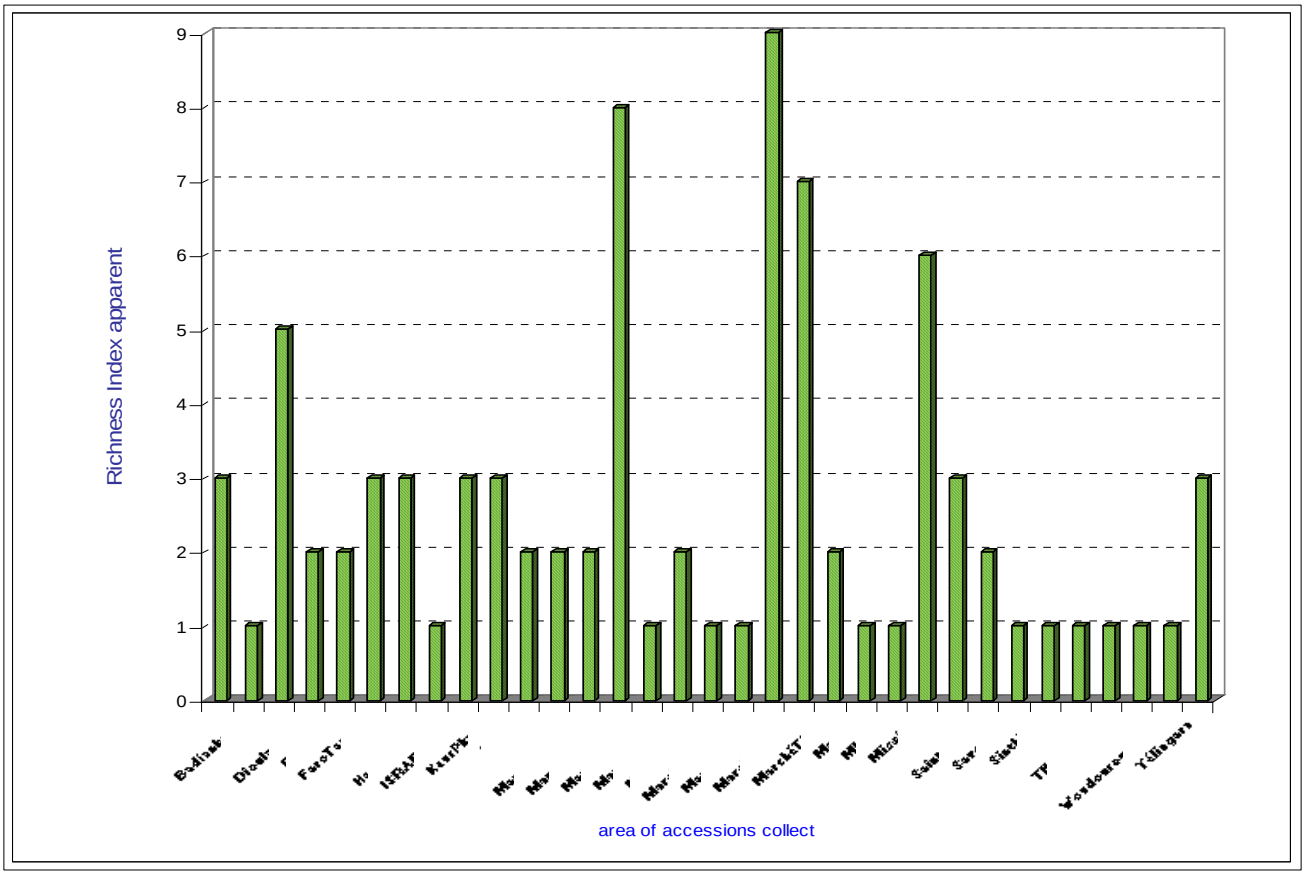

Figure 6: Site-specific diversity of the localities.

2.2. PARTICIPATORY PLANT BREEDING OF ALVs CULTIVARS

Bissap or roselle (*Hibiscus sabdariffa* L.)

While being based on a total of seven criteria of selection (acidity, leaf width, leaf shape, leaf color, easiest to sale, capacity of regeneration after harvest & yield), the cultivar L24 is considered to be more interesting for five criteria according to leaf width as leaf shape, easiest to sale, and the capacity of regeneration after harvest. On the other hand, the cultivar Koor is appreciated as last for the first 4 criteria above in addition to the color of its laeves which is not at all attractive. L24 remains among the cultivars tested most attractive for the two others criteria (color and acidity of the leaves). The results of the analysis of acidity showed the predominance of 4 cultivars which are VCDH, L22, L24 and ACCM. It is the same for the color for the leaves where ACCM, L7, L24 and L22 are preferred. Thus, the classification of the cultivars according to the method of matric notation devotes by far L24 like the cultivar preferred it is followed respectively: L7, ACCM, L22, VCDH, L28, Vimto, VF and Koor. Cultivar L24 show a better performance than the others cultivars (Figure 1). It was reported the preference of farmers growing roselle on the cultivars with green and broad leaves (DIOUF *& al*., 2004). For all the cultivars subject to this study the five first are green type and the last four the red type, as showed on Figure 7. But it should be noted that the method of matric notation used during this participatory breeding session confirm the preference of the users. These results come to confirm those of DIOUF *& al*. (2004) on the preference of users for the green type of roselle.

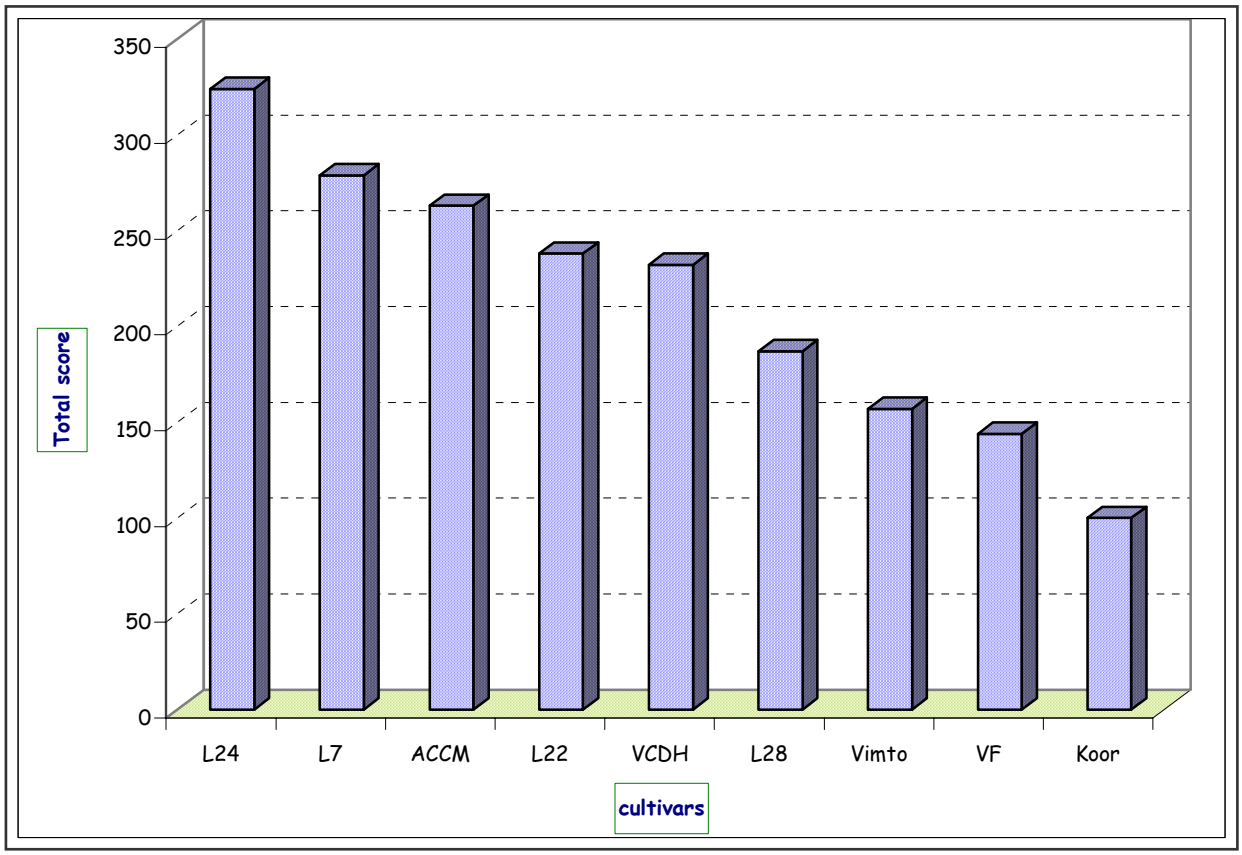

Figure 7: Ranking *Hibiscus* (roselle or bissap) cultivars according to total score.

Cowpea (*Vigna unguiculata* (L.) WALP)

The cultivars Kolda1, YélingaraGF and Fatick9 are respectively latest. According to the tenderness or times of cooking of the leaves we have respectively AVRDC, Kolda1, YélingaraGF and Fatick9. The cultivars Fatick9 and Kolda1 have leaves color more attractive than the others, they are followed by YelingaraGF and the AVRDC. Referring to plant canopy, the cultivar Kolda1 become the best one. For all the 5 criterion of selection, the cultivar Kolda1 is very well appreciated for its lateness, the length of its leaves and for the canopy contrary to Mougne which is less appreciated. In summary, the four better cultivars according to users' classification are respectively in decreasing order Kolda1, YélingaraGF, AVRDC and Fatick9 (Figure 8). The three better cultivars (Kolda1, YélingaraGF and Fatick9) are latest, have broad leaves and erect type. These same criteria of preferences were reported by DIOUF & *al*.(2004).

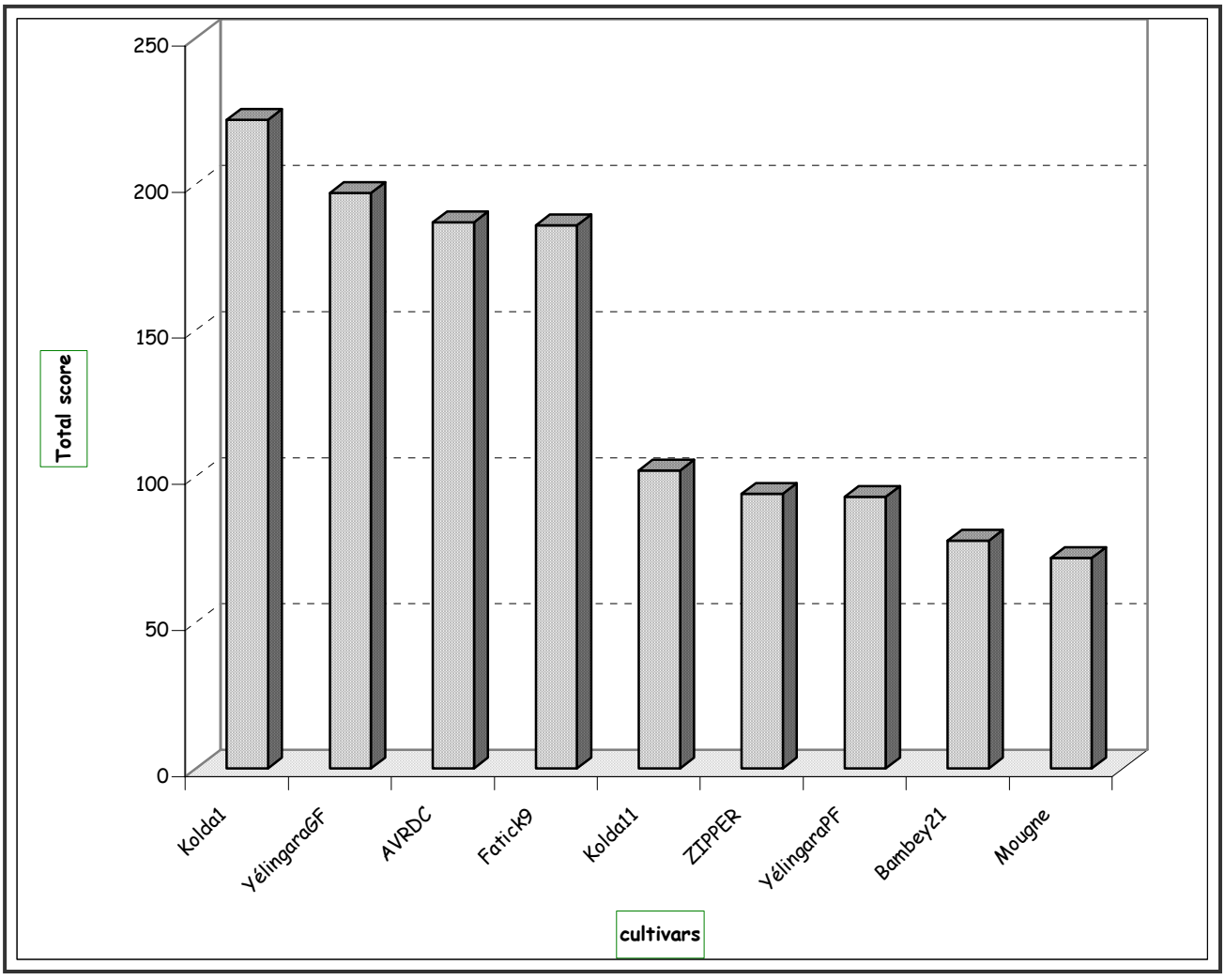

Figure 8: Ranking *Cowpea* cultivars according to total score

Amaranth (*Amaranthus* L. spp)

Taking into consideration criterion of selection of the amaranth (width and color of the leaves, lateness & time of cooking), cultivar AVRDC is always preferred by than all the others, is each time followed by Tamba7. Referring to farmers' classification, the best cultivars are respectively in decreasing order the cultivars AVRDC, Tamba7, Missira5 and Tamba4. (Figure 9). These results confirm those of DIOUF *& al*. (2004).

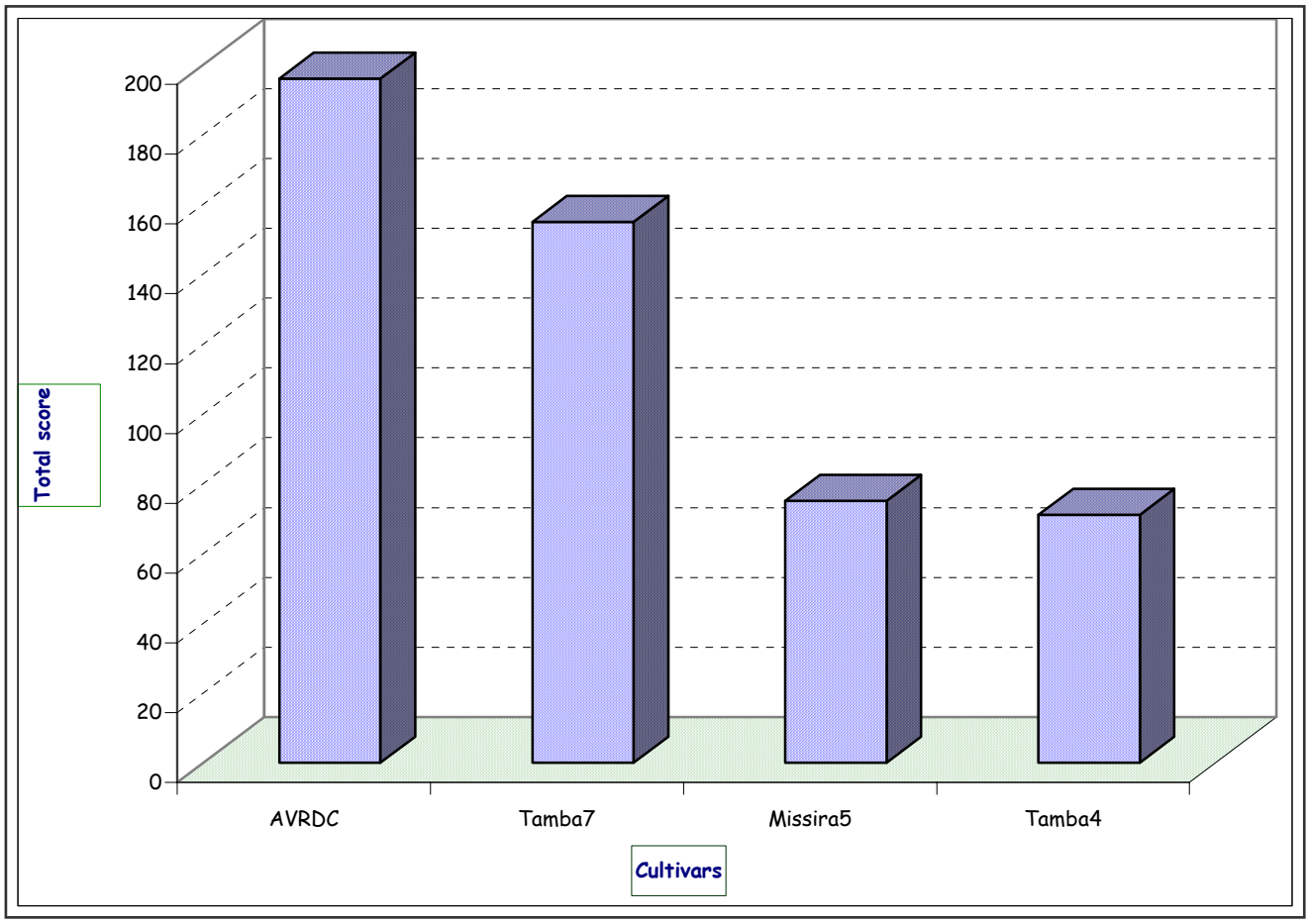

Figure 9: Ranking *Amaranthus* cultivars according to total score

Moringa (*Moringa oleifera* Lam)

By considering the four criteria (Width, color, productivity and time of cooking of the leaves) cultivar MAVRDC is the first. The classification of the cultivars according to the method of matric notation is established in a decreasing way as follows : MAVRDC, MCDH and MKOTHIARY (Figure 10). Over our previous surveys no criterion of varietal distinction or preferably was reported (DIOUF *& al*., 2004). It is why LABRADA (2000) reported that the farmers are main source of empirical and theoretical knowledges of the plants, seeds and potential production of each cultivar.

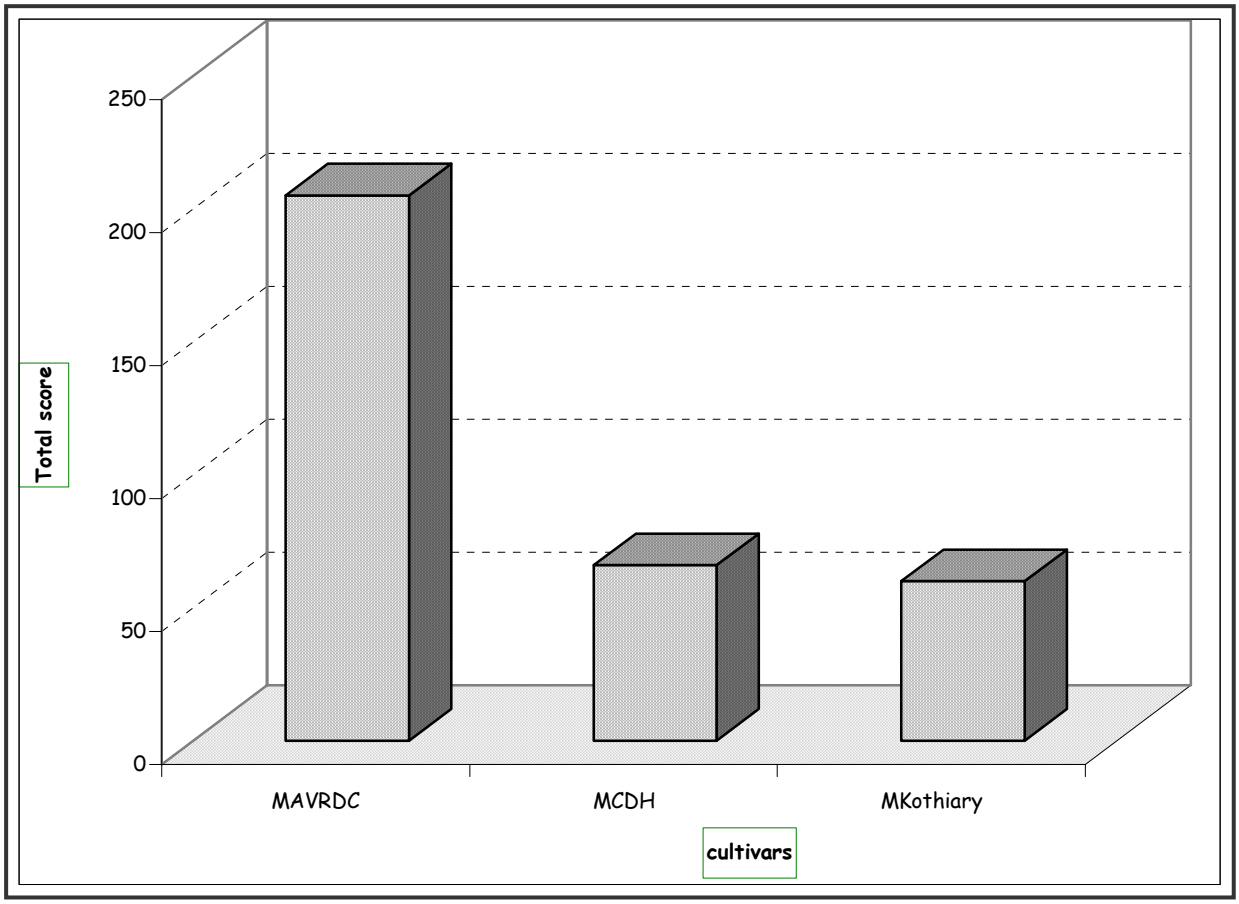

Figure 10 : Ranking *Moringaa* cultivars according to total score

3. CONCLUSION

The agro-morphological characterization of the accessions of the 4 traditional African leaf vegetables species has contributed to a better assessment of the phenotypes of the collected seed samples and an identification of the plants having the preference characteristics identified by the women producers. It has showed the existence of intra and inter-specific variability, but also the existence of gene movement of seeds within and between localities on a distance reaching sometimes 800 km. Markets remain the main exchange place for the seeds. The identification of duplication will lead to a good ex situ conservation strategy of the accessions. However, it is important that the agro-morphological characterization be completed by more sophisticated studies such as biochemical and molecular in order to validate or not our results. The enhancement of the number of accessions of the different species and the uses of different modes of characterizations can contribute to the implementation of a core collection which the best strategy of conserving germ plasm.

All the new cultivars are ranked by the users and the level of adoption should be high. Farmers at Mboro village start to increase their income when selling the best cultivar of Bissap (L24). Dissemination of these new cultivars & conservation strategies through use is starting using homegardens in pilots sites located at Fatick, Kaolack, Thies & Diourbel regions of Senegal.

4. REFERENCES BIBLIOGRAPHIQUES

AVRDC, 2002. AVRDC-GRSU *Collecting record sheet of Moringa oleifera* Lam AVRDC-ARP. P O Box 10 Duliti, Arhusha, Tanzanie. 2 pages.

BENIEST J. (ed.), 1987. *Guide pratique du maraîchage au Sénégal*. CDH-ISRA. BP 3120. Dakar Sénégal. 144 pages.

BRICAGE, P., 1978. Le bissap (*Hibiscus sabdariffa* L.), Malvacées. Aspects biologiques. *Bulletin AASNS*, décembre 1978, n°64 : Pages 9-23.

CERAAS, 2002. Les méthodes de recherche utilisées pour l'évaluation participative des technologies. *CERAAS* Thiès, Dakar Sénégal. 8 pages.

CHADHA M. L., ENGLE L. M. et OLUOCH M. O. 2000. *Vegetable germplasm - conservation and management.* A compilation of lecture contents of training course held at AVRDC-ARP 26-April 1, 2000. Organized by AVRDC-ARP SADC Plant genetic Resources Center, Zambia. AVRDC-African Regional Program. P. O. Box 10, Duliti, Arusha, Tanzania. 207 pages.

CGIAR (Consultative Group on Intenational Agricultural Research), **1994**. Le programme du GCRAI sur les ressources phytogénétiques. Les hommes et les plantes: programme pour le développement. 12 pages.

DIOUF M., DIOP M., LO C., DRAME K.A., SENE E., BA C. O., GUEYE, M. et FAYE, B. 1999. Prospection de légumes feuilles traditionnels de type africain au Sénégal. *In Biodiversity of traditional leafy vegetables in Africa*. Editors J.A CHWEYA and P. EYZAGUIRE, International Plant Genetic Institute (IPGRI) Via delle sette Chiese 14200145 Rome Italie. Pp111-150.

DIOUF, M., GUEYE M., FAYE B., DIEME O., LO C., GNINGUE D.,. BA, C. O. BA T., NIANG Y., BA. DIAO M., TAMBA A.,. MBAYE A. A. et FALL C. A. 2004. Gestion du germoplasme des légumes-feuilles traditionnels de type africain au Sénégal. *In Proceeding of regional workshop on Plant genetic resources for food and security in west and central Africa*. 22-30 avril 2004 Ibadan Nigéria.

GENTLEMAN R et IHAKA R., 1996. Statistics Department of the University of Auckland. *In Simple R-Using R for Introductory Statistics*. Editor VERZANI, J. (2002). IPGRI-SSA Nairobi Kenya. 109 pages.

IPGRI, 2001. *Design and analysis of evaluation trials of genetic resources collections*. IPGRI, Via dei treenari, 472/a, 00057, Maccarese, Rome, Italy. 53 pages.

ISRA-CNRA, 1987. Synthèse des travaux de recherche sur le niébé (*Vigna unguiculata* L.). ISRA-CNRA, BAMBEY, Sénégal. 12 pages.

LABRADA, H. R, 2002. Sélection végétale participative : cultiver le savoir. Point de vue. *Spore* N°102. page 16.

ROUSSEl, J., 1995. Pépinières et plantations forestières en Afrique tropicale sèche. ISRA-CNRF BP 3120 Dakar Sénégal 435 pages.

VAN DE PLAS, G., 1980. *Techniques pour la production de semences de Baselle et d'Amarante au Sénégal.* Centre pour le Développement de l'Horticulture (CDH), CDH / ISRA, B. P. 3120 Cambérène, Dakar Sénégal. 9p.

ZAHOUR.,1992. Eléments d'amélioration génétique des plantes. Manuels scientifiques et techniques. *Editions Actes*.230 pages.