High pigments and atroviolacea double mutant of tomato*(Solanum lycopersicum)*with high pigments content

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Abstract

Tomato is known to contain nutritional components with several types of health-promoting actions. Lycopene which is an antioxidant present in tomatoes has been identified as being responsible for the beneficial effect of tomatoes. With the aim of increasing the antioxidants contents of tomato, two dark fruited single recessive mutants were crossed to produce double mutant with higher antioxidants contents. The high pigment (*hp*) mutant of tomato characterized by dark green fruits, when it is immature and deep red mature fruit which is a monogenic mutant of tomato, was crossed with atroviolacea (*atv*) monogenic non-allelic mutant which had the same phenotype. Although it was easy to distinguish the *hpatv* segregating double mutant in the f2 generation, the conformation of the *hpatv* double mutant was made genetically. The double mutant showed darker phenotype in stem, leaves,and much darker fruits due to higher pigments content. The double mutant qualitatively showed higher pigments content which is of great nutritional value The detailed estimation of the quantitative differences in pigments of the wild type, singlemutants and constructed mutant is recommended.

Keywords: High pigment, mutant , tomato, double mutants, antioxidant.

Introduction

Tomato (*Solanum lycopersicum)* is an important vegetable all over the world and, in Yemen, its consumption is increased as its available in all seasons, it is also part of the three times limited daily menu in Yemen. Oxidative stress is now recognized as an important etiological factor that causes several chronic diseases including cancer, cardiovascular diseases, osteoporosis, and diabetes(4). Antioxidants play an important role in mitigating the damaging effects of oxidative stress on cells. Lycopene, a carotenoid antioxidant has received considerable scientific interest in recent years(4). It was foundthatlycopenecould decrease the low density lipoprotein (LDL) oxidation(1). Recently, it has also been found out that a diet high in olive oil and rich in lycopene may decrease the risk of coronary heart diseases by improving the serum lipid profile compared with a high-carbohydrate, low fat lycopene rich diet(1). At least 85%of our dietary lycopene comes from tomato fruits and tomato based products, the remainder being obtained from watermelon, pink grapefruits, guava, and papaya(3). Color is a defining factor of the quality of tomato fruits which is of great influence on its nutritional quality . There are many mutants of tomato described by hyper responsiveness to light and possess deep-red colored fruits. These mutant fruits are of high level of antioxidants. One of these mutantsis high pigment mutant (*hp*) which is caused by single recessive mutation, and atroviolacea(*atv*)is anther non allelic recessive mutant with the similar phenotype. At the ripening stage,the dark green fruits of the two mutants become dark red, this dark red color is mainlydue to elevated levels of carotenoids of which lycopene is the most important. There are many approaches to improve nutritional qualities of tomato such as genetic engineering (6,10) breeding for quantitative traits and breeding of qualitative traits using traditional methods (10) . In this study,the classical approachwas used breeding of qualitative traits to bring two recessive genes to ensure their traits stability, compared to quantitative traits breeding which mostly segregate and the genetic engineering which has many disadvantages. The objective is to increase the amount of flavonoids and carotenoids, Including lycopene and other pigments in daily consumed tomato.

Materials and methods:

The Ailsacraig cultivar of tomato (*Solanum lycopersicum*), high pigment (*hp*) mutant of tomato and atroviolacea (*atv*) monogenic non-allelic mutant were used in this study.

Growth condition:

Seeds were surface sterilized and sown on filter papers. After emergence of the radicle, seedlings they were grown on peat moss. After 2weeks, young plant were transferred to pots in green house and normal cultural practices were done in growing the plants.

Crossing, construction and isolation of double mutant :

The crosses were made between the monogenic mutants *hp* and *atv* and double mutant was scored in f2 and f3 generation of segregating population on the basis of the phenotype**.** Flower which did not open were emasculated before shedding pollens. Emasculated flowers were covered with cheesecloth or butter paper bags to protect them from insect pollination. After 24 hours of emasculation, pollen were collected from parents and applied to the receptivestigma.The pollinated flowers were recovered (G. Kalloo personal communication, Directorate of vegetable Research, Varanasi,India and G. and (7).To confirm the presence of the two alleles *hp* and *atv*in homozygous state, the double mutant *hpatv* was crossed with each single mutant which resulted inasingle mutant in phenotype in f1 generation. Observation was made on fruits grown under field conditions

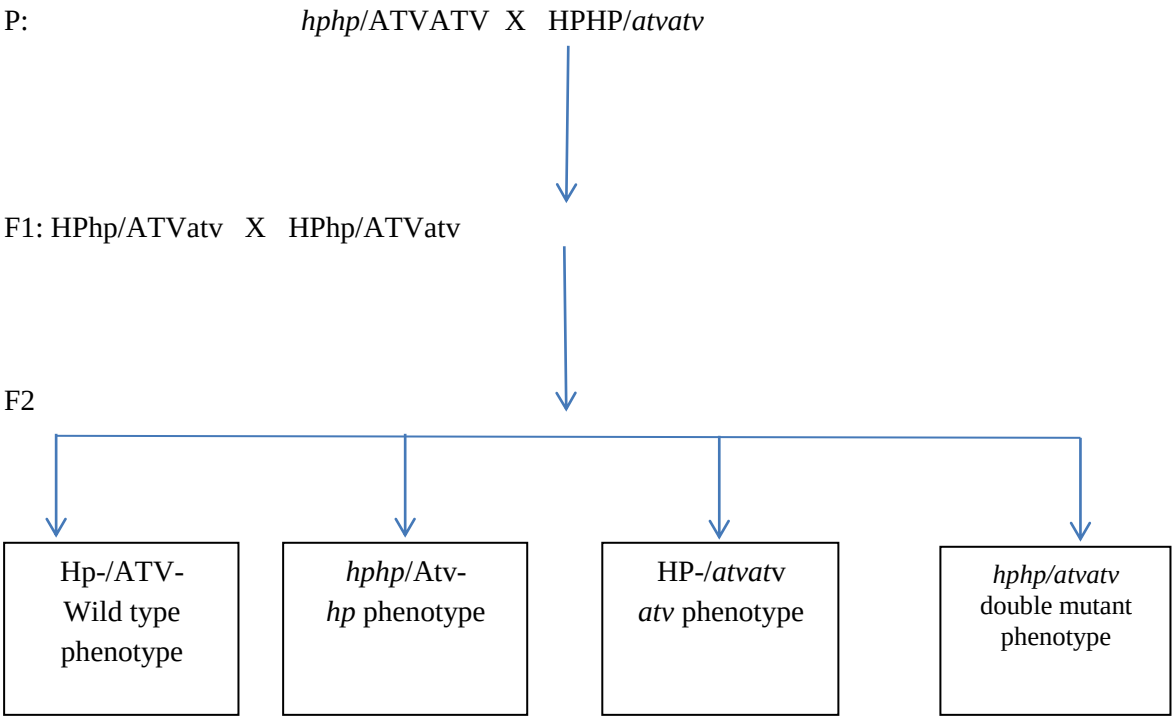


Fig.1 The crossing of the single mutants *hp* and *atv* and isolation of the double mutant *hpatv* in the f2 generation.

Results and discussion:

Color is one of the most important quality characteristic of tomato and other vegetables and fruits based on the European Union quality standards.I isolated the double mutant of the two monogenic recessive high pigments nonallelic High pigment (*hp*) and atroviolacea (*atv*) mutants of tomato which is named *hpatv*. The double mutant seedling was easily isolated in f2 generation at the early stage which is characterized by high pigmentation of its hypocotyl, cotyledons and leaves (Fig 2).

The pigmentation intensity was increased by the age of the plant. Approximately, 3 weeks old double mutant continue to be distinguished by its intense pigmentation (Fig3)

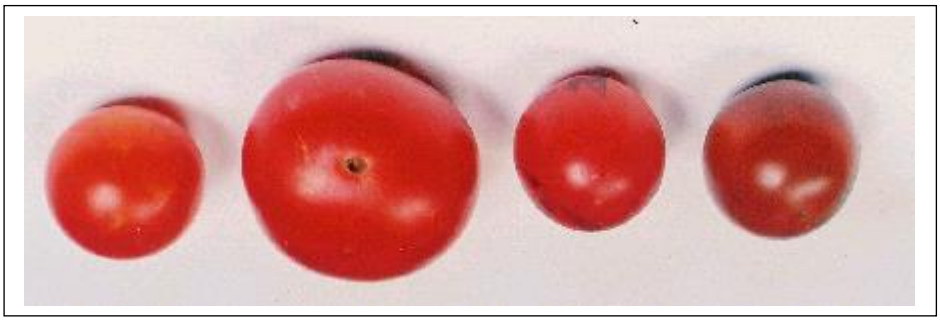
At the early fruit development stages, the double mutant fruits color was also characterized by intense deep green to purple color (Fig.4). The resultant double mutant showed that the manipulation of light signals transduction could be a promising approach to breed tomato for fruit color and nutritional quality. The mature fully ripened fruit of the double mutant also showed deep red color (Fig.5). Early studies indicated that there is correlation between fruit color and total antioxidant concentration with an increase in lycopene content from rose to red color.What was clearly seen is anthocyanin pigment which is contributed by*atv* mutant . It is known that these single mutants mainly *hp* are characterized by their exaggerated photo responsiveness and increased fruits pigments, mainly carotenoids (8).The double mutant showed additive effect for the two allele s*hp* and *atv* the anthocyanin pigmentsseen in the hypocotyl, cotyledons, leaves and stem were much greater than those seen in the single mutant as individuals.The dark red color of the fruit is also another indication of additive effect of the two alleles on the quality of the fruits regarding colors and their contents of flavonoids and carotenoids. The deep red color indicate higher lycopene content which is supported by a study done by Cox *et al*(5)who emphasized that the red- fruiting cultivars also have a higher lycopene content.

The double mutant fruits are definitely high in their carotenoid content, including lycopene which makes it of good nutritional quality. Therefore, it is recommended to study the antioxidant qualities of the *hpatv* double mutant and the amount of Lycopene, B- carotene, ascorbic acid, total sugar and nitrate. It is also recommended to study in details the morphologic, other metabolomic, and photomorphogenic phenotypes of this double mutant in comparison to its normal wild type counterpart and to evaluate its significance relative to other tomato mutants known for increased fruit and leaves and stems pigmentation.Many studies have already shown the increased nutritional qualities of tomato with high pigments (9), but also less work has been done on the involvement of flavonoids on many aspects of plant growth and development, like resistance to disease,uv light protection and other developmental traits.

Figure (2):The variation in green seedlings pigmentation of wild type Alisa Craig cultivar of tomato and *hp* and *atv* single mutants in addition to the double mutant *hpatv*.Note the pinkish darkcolor of the double mutant

Figure (3): the variation in leaves color pigmentation of tomato *hp* and *atv* single mutants in addition to the double mutant *hpatv*.Note the dark pigmentation color of the double mutant

Figure (4): the variation in green fruits pigmentation of wild type Alisa Craig cultivar of tomato and *hp* and *atv* single mutants in addition to the double mutant *hpatv*.Note the deep green color of the double mutant



Figure(5): the variation in fruits red color pigmentationof wild type Alisa Craig cultivar of tomato and *hp* and *atv* single mutants in addition to the double mutant *hpatv*. Note the deep red color of the double mutant

References

1. AhujaKD, PittawayJK, Ball MJ (2006) effect of olive oil and tomato lycopene combination on serum lycopene, lipidsprofile, and lipid oxidation Nutrition 22: 259-265
2. Agarwal S , Rao AV. (1998) Tomato lycopene and low density lipoprotein oxidation: a human dietary intervention study . Lipids. 33 :981-984.
3. Bramley P.M (2000).Is lycopene beneficial to human health? Phytochemistry54:233-236
4. Chen J, Song Y, Zhang L. (2013) Effect of lycopene supplementation on oxidative stress: an exploratory systematic review and meta-analysis of randomized controlled trials.Journal Med Food. 16: 361-374
5. Cox S. E., Stushnoff C., Sampson D. A. (2003). Relationship of fruit colourandlight exposure to lycopene content and antioxidant properties of tomato. CanadianJournal of Plant Science, 83(4): 913–919
6. Fraser, P.D., Enfissi, E.M.A., &Bramley, P.M. (2009) Genetic engineering of carotenoid formation in tomato fruit and the potential application of systems and synthetic biology approaches. Archives of Biochemistry and Biophysics, 483: 196–204. doi:10.1016/j.abb.2008.10.009
7. Kalloo, G. (1991) Genetic Improvement of Tomato, Springer, Berlin, Germany, pp:39-50.
8. Lavi N, Tadmor Y, Meir A, Bechar A, OrenShamir M,Ovadia R, Reuveni M,Nahon S,Shlomo H, Chen L, (2009) Characterization of the intense pigment tomato genotype emphasizing targeted fruit metabolites and chloroplast biogenesis. J Agric Food Chem **57**: 4818–4826
9. Liu Y, Roof S, Ye Z, Barry C, van Tuinen A, Vrebalov J, Bowler C, Giovannoni J. 2004b. Manipulation of light signal transduction as a means of modifying fruit nutritional quality in tomato. *Proceedings of the National Academy of Sciences, USA* **101**: 9897–9902
10. *Santos, C. S. , Noureddine Benkeblia2 and Marta W. Vasconcelos* (2017) Strategies for Enhancing Phytonutrient Content in Plant-Based Foods in *Phytonutritional Improvement of Crops*, First Edition. Edited by NoureddineBenkeblia. Published by John Wiley & Sons Ltd pp:203-232

hpatv طافر مزدوج للطماطم *(Solanum lycopersicum)* ذو محتوى عال من الأصباغ

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الملخص

تعرف الطماطم باحتوائها على عدة مركبات غذائية معززة لصحة الإنسان، واليكوبينوهوعبارة عن مضاد للأكسدة موجود في الطماطم يعد المسؤول عن الفوائد الصحية للطماطم. بهدف زيادة المواد المضادة للأكسدة في الطماطم استخدمت في هذه الدراسة طافرتين تعرف الأولى بزائدة الأصباغ *hp* والثانية *atv* وكلاهما تتميز بثمار خضراء وحمراء داكنة تم تهجينهما معا. وعلى الرغم من سهولة تمييز الطافر المزدوج في الجيل الثاني إلا أنَّ تأكيد ذلك تم وراثياً بالتهجين الاختباري. تميز الطافرالمزدوج بمظهر خارجي أشد دكونه في الساق والأوراق والثمار بسبب زيادة الاصباغ التي لها قيمة غذائية ووقائية عالية. توصي الدراسة بالتقدير الكمي والدراسة التفصيلية للفروق في الأصباغ ومضادات الأكسدة في النوع البري والطافرات المفردة والطافر المزدوج.

الكلمات المفتاحية: عالي الصبغة، طافر، طافر مزدوج، مضاد أكسدة، طماطم .