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Anti-Allergic Compounds from Red Tomato Peel

I. Hossin[1], G. Talukder[1,] Nitai Roy[1] and Ranajit Kumar Shaha[1]*

**Faculty of Agro-Industry and Natural Resources, University Malaysia Kelantan, Jeli Campus, Beg Berkunci No. 100, 17600 Jeli, Kelantan, Malaysia.*
[1]Department of Biochemistry & Molecular Biology, University of Rajshahi, Rajshahi-6205, Bangladesh.

Abstract: Daily food habit with anti-allergic activities is expected to prevent the onset of allergic diseases and ameliorate allergic symptoms. The red color of ripe tomato fruit is due mainly to the accumulation of the carotenoid all-trans-lycopene, which is produced during fruit ripening. Red tomato peel extract (RTPE) contain lycopene (450mg/ kg) and has been found to have anti-allergic effects on man compare its flesh extract and yellow tomato peel extract (YTPE). RTPE could possibly inhibit histamine release and relieve the symptoms of all types of allergy including cedar pollinosis. To evaluate the anti-allergic effect of RTPE, we performed a research study randomized, on three groups in 50 perennial allergy patients with red peel extract; yellow peel extract; tomato flesh and Dextrin (20 healthy university students as control). All patients using oral administration of red tomato peel extract (RTPE); yellow/pink tomato peel extract (YTPE) and tomato flesh extract (TFE) 30mg per day /patient each group and Dextrin as control (30mg per day /patient each group) for 56 days. We found that the skin test for allergy score significantly decreased in the RTPE group only at the end of the trial compared to the beginning and treated with YTPE and TFE groups. Besides this, we also found that sneezing score decreases significantly at the end of experiment time compare to the beginning ($p<0.03$). There were decreasing tendencies of rhinorrhea and nasal obstruction in the RTPE group. The patients quality of life was significantly improved in the RTPE group after 56 days of treatment ($p=0.02$) but not in YTPE; tomato flesh extract (TFE) and dextrin group. A significant improvement in total symptom scores, combining sneezing, rhinorrhea and nasal obstruction, was observed after oral administration of RTPE for 56 days ($p=0.01$). Thus, this evidence will be helpful for the development of low molecular compounds for allergic diseases and it is expected that a dietary menu including an appropriate intake of carotenoids (fruits & vegetables) may provide a form of complementary and alternative medicine and a preventative strategy for allergic diseases.

Keywords: Lycopene, Eosinophils, Histamine release, RTPE (red colored tomato peel extract), YTPE (yellow/pink colored tomato peel extract), Tomato flesh extract (TFE).

1. Introduction

Plants produce pigments to provide color to their flowers and fruit for attracting pollinators and seed dispersers. In flowers and fruit, these colors are due mainly to carotenoid and flavonoid pigments. Carotenoids are lipid-soluble 40-carbon isoprenoids, and more than 700 naturally occurring carotenoids have been identified (Britton *et al*., 2004). Carotenoids are essential for plant life since they provide important photoprotective functions during photosynthesis in chloroplasts and serve as precursors for the phytohormone abscisic acid (Grotewold, 2006). Beyond their essential biological activities, carotenoids also accumulate in chromoplasts of flowers and fruit, where they function as yellow- to red-colored pigments (Young and Frank, 1996; Tanaka *et al*., 2008). Carotenoids play important roles in human nutrition and health since they are the precursors for vitamin A, are lipophilic antioxidants, and have anti-cancer properties (Basuand Imrhan, 2007; Rao and Rao, 2007; Singh and Goyal, 2008). Lycopene, a fat-soluble

*Corresponding author:
E-mail: ranajit@umk.edu.my; ranajit57@yahoo.com.

carotenoid, is a precursor of β-carotene (Sandmann, 1994) and has at least twice the antioxidant capacity of β-carotene (Di Mascio *et al*., 1989). Epidemiological studies have indicated positive health benefits in consumption of diets high in lycopene (Gerster, 1997).

Tomato (*Lycopersicon esculentum* Miller) is termed as "the most popular vegetable fruit". The tomato is cooked as vegetable alone or in combination with potato besides eating raw when ripe. It is a fruit of good nutritive value as it is fairly rich in vitamins (Vitamin C), and other minerals like calcium, phosphorus and iron. Considering its low cost, it qualifies for inclusion in the daily diet of young and growing children. The red color of ripe tomato fruit is due mainly to the accumulation of the carotenoid all-trans-lycopene, which is produced during fruit ripening. In addition to lycopene, tomato fruit contains significant levels of other carotenoids, such as β-carotene, phytoene, violaxanthin, and lutein. Mutants in the carotenoid pathway have an altered carotenoid composition, resulting indifferent fruit colors, such as orange (tangerine, beta) or yellow(r) fruit (Lewinsohn *et al*., 2005). Besides carotenoids, flavonoids play a role in determining the color of tomato fruit (Schijlen *et al*., 2008; Bovy *et al*., 2010). One of the most abundant flavonoids in the tomato fruit peel is the yellow-colored "naringenin chalcone"**.** It accumulates in the cuticle upon ripening and is responsible for the yellow color that develops in the peel at breaker stage, preceding the production of lycopene (Hunt and Baker, 1980). Lycopene is a vibrant red carotenoid that serves as an intermediate in the biosynthesis of other carotenoids and is found in moderate to high concentrations during tomato maturation, watermelon, red grapefruit and Brazilian guava (Stahl and Sies, 1996). These results suggest that "naringenin chalcone" is required to give tomato its typical orange-red color and that a lack of **"**naringenin chalcone" leads to pink-colored fruit. Tomatoes also contain small amounts of flavonoids in their peel (5–10mg/kg fresh weight), mainly naringenin chalcone and the flavonol rutin, a quercetin glycoside. Flavonols are very potent antioxidants, and an increasing body of epidemiological data suggests that high flavonoid intake is correlated with a decreased risk of cardiovascular disease. Allergic diseases, such as allergic rhinitis, bronchial asthma and atopic dermatitis, have increased dramatically in the past decades. Perennial Allergic Rhinitis is one of the representative allergic diseases. Its prevalence reached 18.7% in the Japanese population, which is higher than that of cedar pollinosis (16.2%) [Baba, K. *et al*., 2002]. The therapeutic strategy is usually focused on removal and evasion of antigens by environmental maintenance and pharmacotherapy. Inhibitors of chemical mediators, antihistamines and topical steroids are used widely, but their potential side-effects are of concern with long-term application. Recently, there is a growing interest especially in regard to the anti-allergic effects of plant polyphenols, some of which is also contained in red color tomatoes. Yamamoto, T. *et al*., (2004) reported that tomato extract (TE), extracted from the tomato flesh with 60% aqueous ethyl alcohol, could inhibit histamine release from rat peritoneal mast cells stimulated by a 40/80 compound and mouse ear swelling responses. Moreover, it is reported that TE can relieve the symptom of Japanese cedar pollinosis (Ryu Y. *et al*., 2003). These findings collectively indicate that TE may have therapeutic efficacy in allergic rhinitis. Now a day there is a growing interest in natural healing methods and particular efforts have been devoted to elucidating medicinal effects of natural agents in the plant and the typical methods of plant extract preparation (Saieed, P. *et al*., 2006). In this study, we confirmed the anti-allergic activity of red tomato peel extract (RTPE) which contents carotenoids compound is more effective than yellow color tomato peel and tomato flesh extract (TE). The experiment was done in a randomized, placebo-controlled clinical trial in patients with perennial allergy patients and skin test method.

2. Materials

2.1 Sample preparation

For biochemical and molecular analyses, Bangladesh Fruits Research center (Rajshahi) provided tomato fruits from individual genotypes of two breeding populations segregating for the pink phenotype (population 35025, two pinks- and one red-fruited genotype; population 35037, one pink- and one red-fruited genotype) and from an additional set of five pink varieties.

Samples consisted of a pool of at least six fruits per genotype, collected at three stages of ripening. Chemical constituents of the new variety tomato shown in Table-1. Samples were separated into the peel and flesh tissues, immediately frozen in liquid nitrogen, ground, and stored at –80°C until analysis.

2.2 Measurement of Lycopene

2.2.1 Conventional Lycopene Assay

Total lycopene was measured in duplicate 2gm samples of each puree by the method of Sadler *et al*., (1990) modified as described by Perkins-Veazie *et al*., (2001). This assay requires 25mL of 0.05% (w/v) butylated hydroxytoluene (BHT) in acetone, 25mL of 95% ethanol, and 50mL hexane per sample assayed.

2.2.2 Reduced Volume Lycopene Assay

To each amber vial were added: 5mL of 0.05% (w/v) BHT in acetone, 5mL of 95% ethanol, and 10mL of hexane. The hexane was delivered with a volumetric pipette calibrated to deliver 10mL at 20^0C. Later assays employed volume dispensers to deliver the organic solvents (Dispensette TM organic dispenser, BrandTech

Scientific, Essex, CT). Both dispensing techniques yielded results comparable in precision and accuracy. Each puree was assayed in triplicate.

A given volume of the sample was removed from the stirred puree and pipette into its corresponding assay vial whose tare weight had been determined. The amount of sample used in the reduced volume assay can range from 0.4 to 0.6gm. The vials were laid on their sides in a rectangular container, covered with a second container that contained ice and placed on an orbital shaker (Lab-Line Instrument Co., Melrose Park, IL) to mix at 180 rpm for 15 minutes. After 15 minutes of shaking, 3mL of deionized water were added to each vial, and the samples were shaken for another 5 min. shaking was stopped, and vials were left at room temperature for 5 min. to allow for phase separation. The absorbance of the hexane (upper) layer was measured in a 1cm path length quartz cuvette at 503nm versus a blank of hexane solvent. The lycopene content of tissue was then estimated by one of the relations:

Lycopene (mmol) = kg tissue ¼ A503

2.3 HPLC-analysis of flavonoid and carotenoid

2.3.1 Flavonoid and Carotenoid Extraction and HPLC Analysis

Flavonoid extraction and HPLC-PDA and liquid chromatography quadrupole time-of-flight mass spectrometry analyses were carried out according to Bino *et al*., (2005). The detected flavonoid compounds were identified using authentic standards and accurate mass liquid chromatography-mass spectrometry analysis (Moco *et al*., 2006). Carotenoids were extracted as described previously by López-Ráez *et al*., (2008) and analyzed by HPLC-PDA according to Bino *et al*., (2005).

2.3.2 Pathological test subjects

Fifty adult allergic patients (20 men, 30 women, and range 18-56 years of age) were enrolled in the clinical study. The positive results of the following any two tests confirm allergenic (Baba, K. *et al*., 2002).

(a) The allergen skin test or serum allergen-specific IgE against mango flower pollen protein;
(b) Eosinophil count in blood serum and
(c) The nasal provocation test. Each patient had a history of allergenic for more than three years.

Detailed characterizations of recruiting patients are described in Table-3. Among 50 patients, 27 patients (54%) were diagnosed as mild, 15 cases (30%) were moderate, and 8 cases (16%) were served according to the scores of three main teats, on the basis of the guidelines.

3. Methods

Subjects were randomly divided into three groups. One group of patients received an oral administration of tablets containing 30mg of RTPE (red tomato peel extract) per day (group-A; n=20); another group received an oral administration of tablets containing 30mg of PTFE (red tomato flesh extracts) per day (group-B; n=20) and the control group received 30mg (dextrin group; n=10). Each subject received two tablets per day for a course of 8 weeks. The clinical characteristics of patients in both groups are shown in Table-2. During the study period, each patient visited a doctor three times; at the beginning of the trial, after week 4 and the end of the trial (week 8). Skin test and serum allergen-specific IgE (mango pollen protein, Japanese cedar pollen and Artemisia pollen) and eosinophil cationic protein (ECP), and eosinophil count in nasal discharge were also measured.

3.1 Skin prick test

The skin tests were performed on patients suffering from nasobronchial allergy as well as healthy volunteers at the Rajshahi Medical College and the Rajshahi University students. Each patient was tested by placing 10µl of each allergen; at least 5cm apart on the volar surface of his/ her forearm and each site was then pricked with a disposable hypodermic needle. Negative and positive controls were also performed. The negative control was the buffer saline in which the allergen was re-suspended and the positive control was histamine acid phosphate injection diluted with buffered saline to 1:10,000 i.e. 1µg of histamine acid phosphate. The patients were prohibited from using antihistamine, steroid and ephedrine for 48 hrs before the skin prick tests. The skin reactions were read after 15 to 20 min. from the commencement of the test. The test was quantified on the basis of the wheel diameter and graded 1^+ to 4^+. The skin tests were conducted at the Rajshahi University Medical center and Rajshahi Medical College Hospital, Bangladesh. The patients were selected on the basis of their suffering from respiratory allergic disorders.

3.2 Skin Tests (Practical, Picture-1)

A diluted extract of each kind of pollen is applied to a scratch or puncture made on the patient's arm or back or injected under the patient's skin. With a positive reaction, a small, raised, reddened area with a surrounding flush (called a wheal and flare) will appear at the test site. The size of the wheal can provide the physician with an important reaction diagnostic clue. Skin testing remains the most sensitive and least costly diagnostic tool.

4. Results

Tomato (*Lycopersicon esculentum* Miller) is termed as "the most popular vegetable fruit". The tomato is cooked as vegetable alone or in combination with potato besides eating raw when ripe. It is a fruit of good nutritive value as it is fairly rich in carbohydrate,

proteins, fats, vitamins (vitamin C, B_1, B_2), and other minerals like calcium, phosphorus and iron (Table-1). Chemical analysis like percentages of moisture and lycopene contents in two experimental varieties of tomatoes peels and fleshes at different maturity stages are shown in Table-2 & 3. The Red tomato peel extract contains the maximum amount of flavonoids (lycopene) than others tested materials (Picture-2).

Picture 1. Skin Test.

Picture 2. Picture of experimental tomatoes.

4.1 Biochemical Analysis of Flavonoids and Carotenoids in Pink Tomatoes

HPLC-photodiode array (PDA) analysis of peel extracts of red-colored ripe fruit peel and flesh revealed the presence of the main tomato carotenoids are lycopene, β-carotene and lutein (Muir *et al*., 2001). In the peel of Moneyberg red fruit, carotenoids levels increased upon ripening and peaked at turning stage, reaching a maximum concentration of approximately 580mg kg^{-1} fresh weight. Lutein levels in peel remained fairly constant during ripening or slightly decreased in control fruit. The flesh of these fruits contained only trace amounts of carotenoids and flavonoids (Fig. 1-2), in agreement with earlier observations (Bovy *et al*., 2002). In contrast to Moneyberg fruit, peel extracts of the pink IL1b fruit did not show the ripening-dependent increase in naringenin chalcone, while levels of lutein were decreases comparable to those observed in Moneyberg red fruit. These results confirm that red tomato peel carotenoids play an important to relive histamine releases on the other hand to cure allergen city.

4.2 Skin prick test

The skin prick test shows (Picture-1) a great improvements of allergic patients treated with red tomato peel extract (RTPE) than patients treated with yellow tomato peel extract (YTPE); tomato flesh extracts (TFE) and dextrin as control at different time of the interval, which are shown in Table-4(A); (B) and (C).

4.3 Nasal Symptoms and Signs

During the study We found that, the rate of change in patients score of nasal symptoms, quality of living standard (QOL) and the sneezing score of the red peel tomato (RTPE) group significantly decreased at the end of the trial (56 days) compared to the beginning of the trial ($p<0.03$).

Table 1. 100 grams of Tomato contain.

Variety	Condition	Grams Carbohydrate	Grams Fats	Grams Protein	mg of Fe	Carotene µgm	Vit-B_1 µgm	Vit-B_2 µgm	Vit-C mg
Moneyberg (Red colour)	Green	3.7±1.8	.097±1.1	1.13±2.1	1.8	194±2.7	767±2.1	157±1.2	32±0.06
	Mature	3.9±2.2	0.19±0.9	1.17±2.4	0.98	280±2.9	107±2.3	557±1.04	28±0.08
	Ripen	3.6±2.1	0.27±1.1	0.98±1.9	0.4	358±2.1	125±1.9	657±1.0	24±0.09
IL 1b (TKM5U0436) (Yellow colour)	Green	3.5±1.8	0.08±1.6	1.17±2.1	1.6	198±3.1	727±3.1	117±1.7	31±0.09
	Mature	3.3±1.9	0.97±1.9	1.07±1.9	0.89	228±2.1	100±2.6	301±1.8	29±0.08
	Ripen	3.7±2.4	0.21±1.1	0.97±1.2	0.43	306±2.4	120±1.6	340±1.2	22±0.09

Table 2. Moisture and Lycopene content at different maturity stages of tomato peel.

Variety	Moisture (weight %)		Lycopene content (mg/kg dw)
Moneyberg (red colour)	Green	18.5±2.9	250.0±5.6
	Mature	29.0±3.4	340.0±7.4
	Ripen	35.0±4.3	405.0±3.4
IL 1b (TKM5U0436 (Yellow colour)	Green	17.0±2.8	120.0±5.3
	Mature	29.5±5.5	150.0±4.5
	Ripen	33.0±2.2	163.5±3.5

Table 3. Moisture and Lycopene content at different maturity stages of tomato flesh.

Variety	Moisture (weight %)		Lycopene content (mg/kg dw)
Moneyberg (red colour)	Green	68.5±1.9	50.0±3.6
	Mature	79.0±3.4	90.0±4.4
	Ripen	91.0±4.3	99.0±2.4
IL 1b (TKM5U0436 (Yellow colour)	Green	57.0±2.1	40.0±2.3
	Mature	79.5±3.5	50.0±3.5
	Ripen	83.0±2.9	63.5±2.5

Table 4(A). Results of skin test of patients of nasobronchial allergy and controls (Rajshahi University students).

Total number of tests	Patients					Control People		
	-ve	1+	2+	3+	4+	-ve	1+	2+
80 (patients) + 20 (control)	40 (50%)	25 (31.25%)	12 (15%)	3 (3.75%)	0 (0%)	18 (90%)	2 (10%)	0 (0%)

The test was quantified on the basis of the wheel diameter and graded 1^+ to 4^+.

Table 4(B). Results of skin test in patients with nasobronchial allergy after 4 weeks treatment with red tomato peel extract (RTPE); yellow tomato peel (YTPE) and tomato flesh extract (TFE).

Total number of tests	Patients (treatment after 4 weeks) 30mg+30mg=60mg/day					Control People (n=20) 60mg/day		
	-ve	1+	2+	3+	4+	-ve	1+	2+
40 (red tomato peel extract) treatment	22 (55%)	14 (35%)	3 (7.5%)	1 (2.5%)	0 (0%)	17 (85%)	3 (15%)	0 (0%)
40 (yellow tomato peel extract)	20 (50%)	15 (37.5%)	4 (10%)	1 (2.5%)	0 (0%)	18 (90%)	2 (10%)	0 (0%)
40 (tomato flesh extracts) treatment	16 (40%)	12 (30%)	9 (22.5%)	2 (5%)	0 (0%)	17 (85%)	3 (15%)	0 (0%)

The test was quantified on the basis of the wheel diameter and graded 1^+ to 4^+.

Table 4(C). Results of skin test in patients with nasobronchial allergy after 8 weeks treatment with red tomato peel extract (RTPE); yellow tomato peel extract (YTPE) and tomato flesh extract (TFE).

Total number of tests	Patients (treatment after 8 weeks) 30mg+30mg=60mg/day					Control People (n=20) 60mg/day		
	-ve	1+	2+	3+	4+	-ve	1+	2+
40 (red tomato peel extract) treatment	32 (80%)	6 (15%)	2 (5%)	1 (2.5%)	0 (0%)	17 (85%)	3 (15%)	0 (0%)
40 (yellow tomato peel extract)	20 (50%)	15 (37.5%)	4 (10%)	1 (2.5%)	0 (0%)	18 (90%)	2 (10%)	0 (0%)
40 (tomato flesh extracts) treatment	16 (40%)	12 (30%)	9 (22.5%)	2 (5%)	0 (0%)	17 (85%)	3 (15%)	0 (0%)

The test was quantified on the basis of the wheel diameter and graded 1^+ to 4^+.

5. Discussion

Now a day pollinosis caused by wild/garden flower pollen remains a public health problem throughout the world. However, the prevalence of PAR, mainly caused by house dust and mites, is higher than that of Japanese cedar pollinosis (Baba *et al*., 2002). PAR attacks in all seasons and also dominate in younger children, which makes it harder to be cured. Now, many kinds of medicine have been developed and applied clinically. Patients typically need to use these drugs for an entire year with consideration of their side-effects. Additionally, there are only a few kinds of medicine that are appropriate for younger children.

With this background, it is interesting to find food with anti-allergic effects, which could be expected to reduce the dependence on drugs to some extent. Recently, many experimental and clinical studies have been carried out for this purpose in Japan, but reports on anti-allergic foods were mainly against cedar pollinosis, such as lactic acid bacterium (Enomoto, T. *et al*., 2000; Shimada, T. *et al*., 2003; Shimada, T. *et al*., 2004; Shinada, T. *et al*., 2004; Xiao, J.Z. *et al*., 2006; Shimizu, K. 2005), and plant extract such as sweet tea (Ukai, K. *et al*., 1995; Ukai, K. *et al*., 1999), Perilla leaf and seed (Tamura, K. *et al*., 1998; Takano, H. *et al*., 2004) and persimmon leaf (Kotani, M. *et al*., 2000), suggesting an anti-allergic activity of plants with polyphenols. We have searched for anti-allergic components in various kinds of fruits and vegetables and found that tomato extract has activity in inhibiting histamine release from mast cells stimulated by a 40/80 compound (Yamamoto, T. *et al*., 2004). The main active component of TPE is naringenin chalcone, which is one kind of polyphenol and exists in red tomatoes, but not in pink tomatoes (Yamamoto, T., 2003).

Chlorogenic acid is the main phenylpropanoid in tomato fruit. In both peel and flesh of Moneyberg and IL1b fruit, chlorogenic acid levels were comparable and showed the same developmental pattern (Fig. 1), indicating that chlorogenic acid levels were not affected in pink tomato fruit. The predominant carotenoids in red-colored ripe Moneyberg tomatoes were lycopene, β-carotene, and lutein. Levels of lycopene and β-carotene increased during ripening, while lutein levels decreased slightly. At later stages of ripening, lycopene

levels were approximately 3.5-fold higher in peel than in the flesh, while lutein and β-carotene levels were similar in both tissues at all developmental stages. Interestingly, peel and flesh extracts of IL1b tomatoes revealed no major differences in the levels and distribution of these three major carotenoids compared with the Moneyberg control (Fig. 2). These results suggest that the pink phenotype is due to the absence of ripening-induced accumulation of the yellow-colored flavonoid "naringenin chalcone" in the fruit peel rather than to a change in carotenoid composition. This idea was confirmed by the analysis of a collection of commercial tomato materials, consisting of red and pink fruit of two segregating breeding populations and five pink accessions (data not shown).

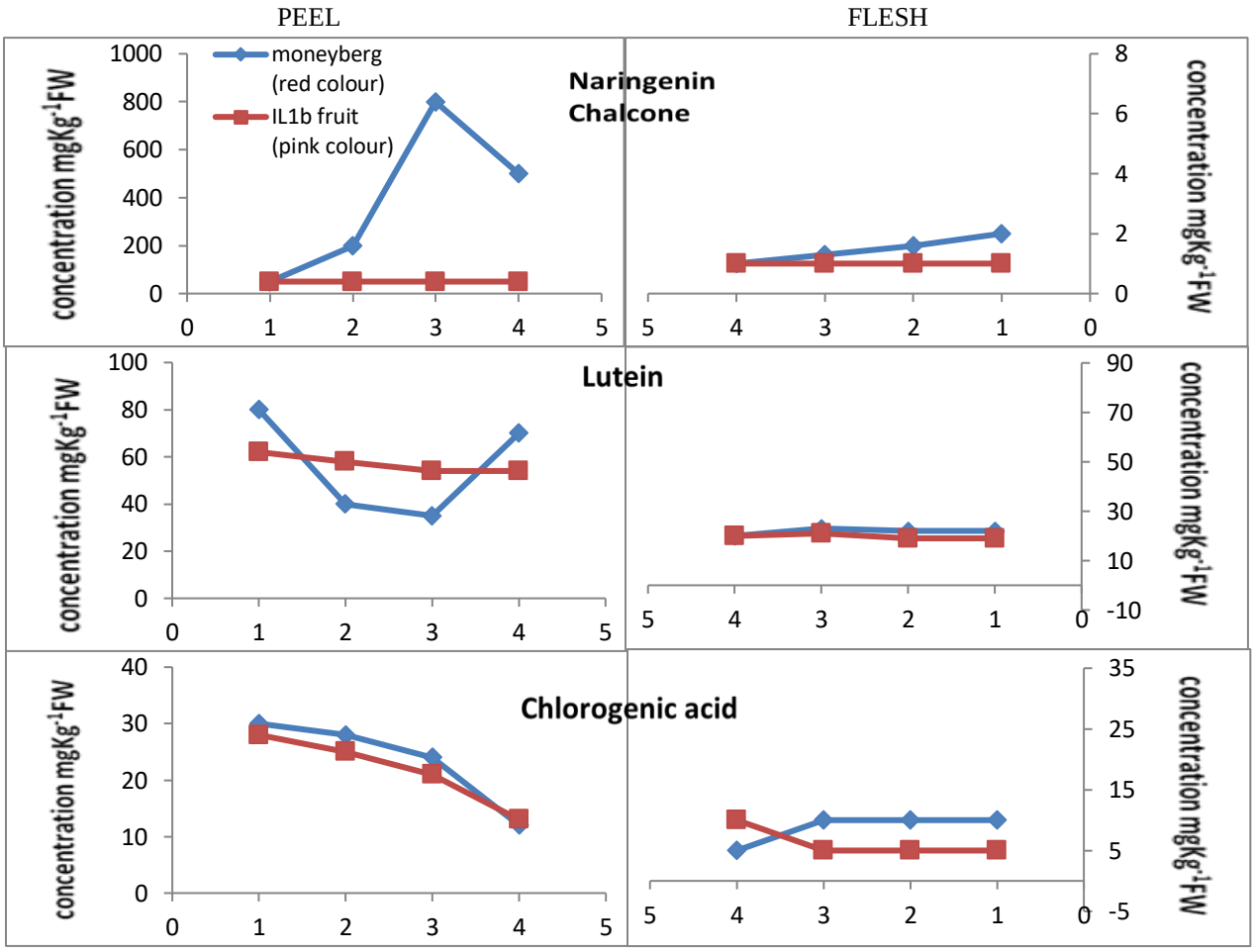

Fig. 1. Phenylpropanoid and flavonoid contents in the peel and flesh of the fruit from tomato CV Moneyberg (black squares) and IL1b (white circles) at four different stages of ripening represent averages of three biological replicates FW, fresh weight.

Table 5. Characteristic of Patients enrolled in the study.

n	Tomato peel extract (red)-20	Tomato flesh extracts-20	Placebo-10
Age (mean)	29.9 ± 2.6	28.8± 2.8	30.2±3.1
		Sex	
Male	9	10	5
Female	11	10	5
		Severity	
Mild	8	9	4
Moderate	8	7	5
Severe	4	4	1
		Symptom scores	
Sneezing	1.31 ±0.10	1.01±0.16	0.98±0.17
Rhinorrhea	1.53±0.19	0.91±0.18	1.01±0.15
Nasal obstruction	1.32 ±0.14	0.98±0.18	1.10±0.16
QOL scores	0.88± 0.11	0.52 ± 0.14	0.43± 0.13

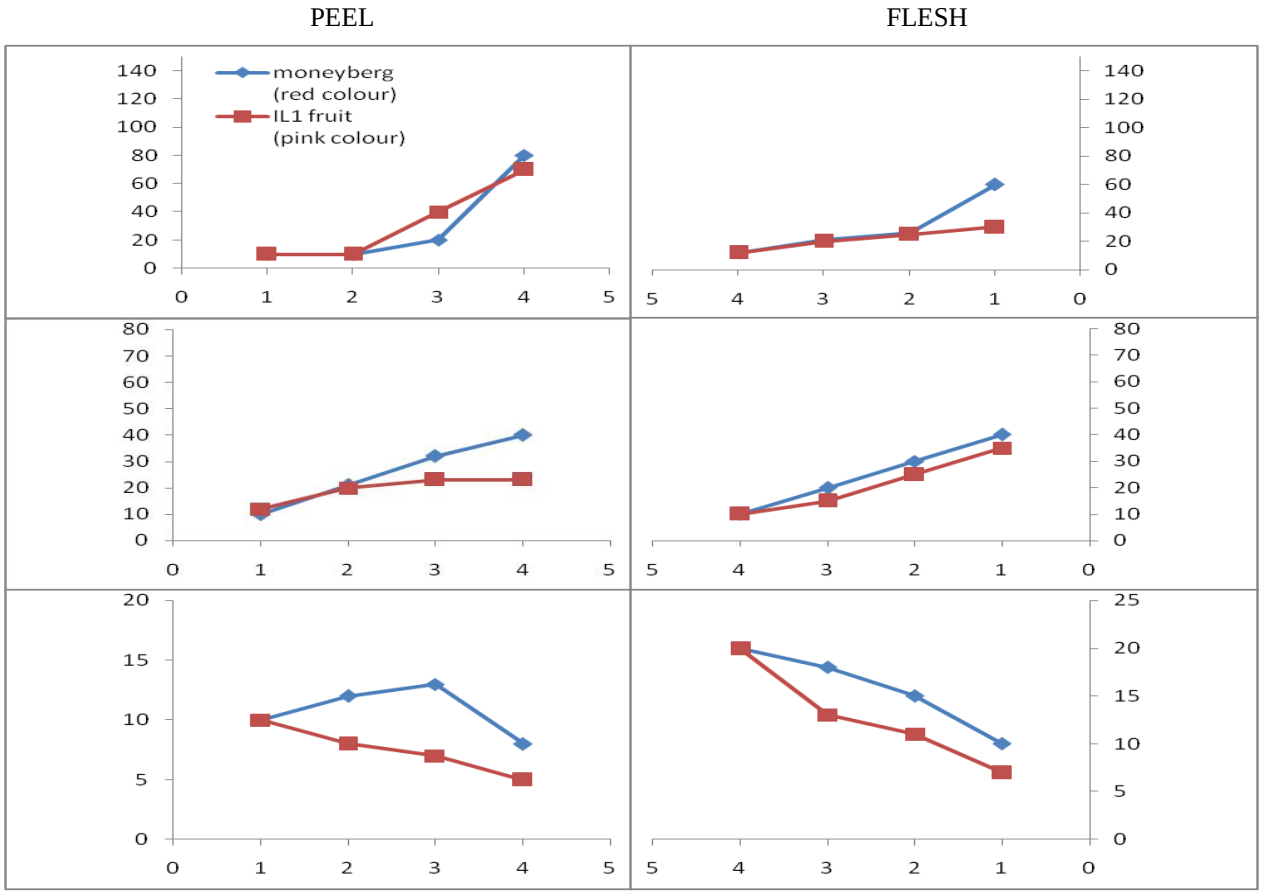

Fig. 2. Carotenoid contents in peel and flesh of fruit from tomato CV Moneyberg (black squares) and IL1b (white circles) at four different stages of ripening. Values represent averages of three biological replicates FW, fresh weight.

Ryu *et al*., have reported that 360mg of Tomato extract per day significantly improved 8 of 11 subjective allergic symptoms and 1 of 5 contents of QOL in 24 volunteer patients with Japanese cedar pollinosis during the peak of pollen scattering (Ryu, Y. *et al*., 2003). This fact indicates that a tomato extract may improve allergic symptoms of pollinosis. So our research findings confirm that Red Tomato Peel Extract which contains lycopene (RTPE) might improve allergic symptoms not only in cedar pollinosis but also in PAR.

Considering tomato is at first a type of food, we performed this study on mild-to-moderate, but not severe patients. In addition, this study is designed as a randomized, and a relatively long test period of 8 weeks was set (Table-5). This result suggested that RTPE is useful in controlling PAR; however, it likely does not act so quickly in the case of cedar pollinosis (Ryu, Y. *et al*., 2003). It is known that type I allergy has two phases of allergic response, i.e. an early phase and late phase. The early phase response is caused by chemical mediators, such as histamine, thromboxanes and leukotrienes released from mast cells after stimulation of antigen, and are the main mechanism in the onset of cedar pollinosis. The late phase response is caused by chemical mediators, such as LTs and ECP, and causes PAR generally (Enomato, T. *et al*., 2000). The anti-allergic function of RTPE is attributed more to inhibition of histamine release from mast cells than an antihistamine effect. Therefore, RTPE acts more quickly in the case of cedar pollinosis than in PAR. This clinical study showed a decreasing tendency of ECP concentration. ECP is one of the chemical mediators released from eosinophils and its quantity is controlled by the activity and quantity of eosinophils. (Wang, D.Y. *et al*., 2000; Naclerio, R.M., 1997). RTPE might inhibit release of chemical mediators, such as ECP, by decreasing eosinophil (Table-6) and, as a result, might inhibit allergic reactions. So we concluded that our finding results treated by RTPE gives better results compared to previous workers (Yoshimura *et al*., 2007) treated with tomato extract. The detailed pharmacological mechanisms and clinical outcomes of TPE need to be investigated further. In conclusion, the oral administration of RTPE can be expected to safely improve the nasal symptoms of PAR. The RTPE dosage and administrative period were fixed in the present trial. Further studies are needed to establish the number of doses for all types of allergic diseases such as asthma, atopic dermatitis and allergic rhinitis.

Table 6. Eosinophil count in nasal discharge (20 patients).

	3+	2+	1+	±	-
Red Tomato peel extract					
0 week	0	8	11	0	1
4 weeks	0	6	10	0	4
8 weeks	0	3	6	0	11
Yellow/Pink TPE					
0 weeks	0	8	11	0	1
4 weeks	0	8	10	0	2
8 weeks	0	5	13	0	2
Red tomato flesh extracts					
0 weeks	0	9	10	0	1
4 weeks	0	8	10	0	2
8 weeks	0	8	9	0	3
Placebo					
0 week	0	9	11	0	0
4 weeks	0	8	10	0	2
8 weeks	0	6	13	0	1

Acknowledgment

For our clinical experiment, we follow same procedure of clinical experiment as Yoshimura *et al*., (2007) and obtained a little different result compare to them. This may be due to Climate or soil fertility condition.

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