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## **Result of Breeding and Selecting High Amino Acid and Reduced Sugar Content-Tea Varieties for High Quality Green Tea Processing in Vietnam**

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Green tea quality mainly depends on internal quality of tea leaves, in which the most important criteria are amino acid and sugar content. The tea varieties that used for green tea processing in Japan, China and Taiwan have higher content of amino acids, and reduced sugars, and lower content of tannin than those used for green tea processing in Vietnam. Basing on the amino acid, reduced sugar and tannin content of parental couples, some tea-hybrid combinations were identified and the crossing between Vietnam originated tea varieties with the tea group of Japan, China and Taiwan, respectively, were conducted. From the F1 hybrid populations, three lines e.g. 209, 218 and 224, which had high content of amino acids (more than 3%), low content of tannin (less than 30%) and good sensory evaluation score of green tea, were identified; 3 lines e.g. 210, 218 and 224 were determined as tea genotypes which denoted high reduced sugar content (more than 3%), low tannin content (less than 30%), and giving good sensory evaluation score of green tea, in which line 224 was the special one, with very high reduced sugar content (4.13%). Among the F1 hybrid population, line 212 was determine as the most prominent tea genotype, with strong growth (yielded 4.1 ton/ha/year at the second age), high content of amino acid (3.67%), rather high content of reduced sugar (2.93%), low content of tannin (27.69%) and giving good result of green tea sensory evaluation (18.2 scores).

**Keywords:** Selection, hybrid combinations, green tea, high quality, amino acid, reduced sugar content, tannin.

### **Introduction**

Up until now, the total area under tea in Vietnam is estimated to cover 130,000 ha. The average yield of 7.3 tonnes of tea buds/ha equivalent to the average yield of tea in the world. Prices average about 1661 USD/ton, just 60-70% compare to the average price in the world. One of the basic reasons that we do not have enough good vareties, especially high-quality vareties to give producers. Forecasts from now until 2020, green tea consumption demand will

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increase more than black tea. So, so we need to focus on improving the quality of green tea to increase the competitiveness of Vietnam's tea on the market.

Tea quality material depends on two factors: Characteristics outside are the mechanical composition and morphology of the material; Characteristic inside is the endoplasmic that is reflected by the indicators primarily chemical composition of tea leaves as tannins, catechins, sugar, amino acids and other substances. The two most important indicators are amino acids and sugar, their transformation during processing leads to the formation of water color, flavor characteristics for each type of tea products.

The varieties for processing green tea, Oolong tea with high quality requires high amino acids content. So, in breeding varieties require amino acid content as high as possible. High quality tea varieties of Japanese popular amino acid content 2.5 - 3%, while our tea varieties only about 1,0- 2,0%. There are many varieties imported from China, Taiwan and Japan into Vietnam. However, the varieties for green tea and Oolong tea with quality high, mostly weak growth, more pests and could not develop. That is why, in recent years we have conducted sexual hybrids between domestic imported varieties with high amino acid, reducing sugar content with Vietnam varieties to create varieties which have high amino acids and reducing sugars content, resistance to external conditions in Vietnam, meeting the demand of raw materials for processing high quality green tea and Oolong tea.

## **Materials and methods**

### ***Material***

Subjects of study: includes varieties representing for variants tea in the gene bank garden at The Northern Mountains Agriculture and Forestry Science Institute (NOMAFSI), the prospective tea line, the individuals were created by sexual hybrid method.

- Group of individuals hybrid: 1087 individuals of 15 pairs of hybrid following:

<b>No.</b>	<b>Hybrid pairs</b>
1	Long Van 2000/Trung Du
2	Kim Tuyen/Trung Du
3	Kim Tuyen/ TRI777
4	TRI777/Trung Du
5	Bat Tien/Trung Du
6	Kanayamidori/Chat Tien
7	Long Van 2000/PH1

8	Okumidori/PH1
9	Saemidori/Cu De Phung
10	Tham Ve/Trung Du
11	Bat Tien/Trung Du
12	Asatsuyu/Trung Du
13	Ho Nam 3/Trung Du
14	Meiryoku//Chat Tien
15	Meiryoku/Trung Du

- 15 lines selected from the hybrid pairs:

Line 204; 205; 207; 209 is a offspring of cross: Kim Tuyen / TRI777 (KT / TRI777).

Line 210, 212; 218 and 224 is a offspring of cross: Kim Tuyen / Midland (KT / TD)

Line 238; 241 is a offspring of cross: Kanayamidori / Quality Money (Kanayamidori / CT)

Line 248, 252 pairs of hybrid crosses :Okumdori / Midland (Okumdori / TD)

Line 253 is a offspring of cross: Asatsuyu / PH1 (Asatsuyu / PH1)

Line 262 is a offspring of cross: Long Van 2000 / PH1 (LV2000 / PH1)

Line 234 is a offspring of cross: Bat Tien / Midland (BT / TD)

### ***Research Methods***

- Group of representative tea varieties was planted in the gene bank garden at NOMAFSI. Each sample includes 3 plants with spacing of 1.5 m; rows 2.0 m; the varieties planted in the sequential method, not repeated.

- Compare varieties experiment was arranged in completely randomized design, repeated 3 times, each replication consisting of two rows, each row consisting of 10 trees.

Tracking methods: The samples of varieties in the gene bank garden and offsprings were assessed for indicators of morphological, biological properties of NOMAFSI method and method of monitoring DUS testing field in 2005 for the tea.

- Data analysis and processing were performed with Excel and IRRISTAT 5.0

### **Results and Discussion**

***Reviewed biochemical composition of some varieties representing tea variants.***

Tea products are harvested by buds and young leaves, so the characteristics and properties of buds and tea leaves directly related growth capacity, ability for productivity, quality of materials and quality of tea products. Assess the levels of chemicals in the tea leaves show in Table 1:

**Table 1.** Composition of certain biochemical main tea varieties representing variants of tea

No	Variaty	Tanin %	CHT %	Catechin TS Mg/gck	Cafein %	Reducing sugarse %	Amino acids %
small leaf China							
1	KT	26,61	42,65	146,00	2,80	3,22	2,42
2	Lv 2000	26,95	42,27	157,05	2,45	3,34	2,02
3	Kanayamidori	25,15	41,29	152,60	2,36	2,91	2,31
4	Saemidori	23,52	42,12	120,00	2,69	4,03	2,86
5	Asatsuyu	25,44	42,57	134,50	2,42	5,01	2,50
6	Bat Tien	30,35	48,51	145,00	2,67	4,53	2,17
7	Okumidori	23,85	42,86	113,6	2,34	3,72	2,74
	The average	25,984	43,18	138,39	2,53	3,82	2,43
big leaf China							
8	TD	30,15	43,30	157,20	2,37	3,26	2,31
9	PVT	27,39	42,84	146,90	2,66	3,03	2,15
10	PT95	27,66	42,23	148,60	2,84	3,19	1,85
	The average	28,4	42,79	150,90	2,62	3,16	2,10
Shan tea							
10	TRI777	33,30	41,7	143,52	3,76	2,91	1,43
12	Chat Tien	29,52	42,03	159,90	2,85	4,00	1,81
13	Tham Ve	32,89	44,68	165,80	2,38	4,35	1,76
14	Cu De Phung	33,22	43,40	128,40	2,65	4,63	1,92
	The average	32,23	42,95	149,40	2,91	3,97	1,73
Assam							
15	PH1	34,10	44,56	151,91	3,82	2,52	2,05
16	1A	33,60	44,15	152,40	3,28	2,98	1,38
	The average	33,85	44,36	152,16	3,55	2,75	1,72

Table 1 shows that the tannin content of varieties belong variants TQLN is lowest, averaged 25.98%, ranged from 23,52-26.95% (only Bat tien reached 30.35%) , then to the varieties of variants TQLT have medium tannin content with 28.4% (ranged from 27.66 to 30.15%), this is an important criterion that decide to quality of tea, the varieties of variants Shan tea with tannin content ranging from 29,52-33.3% which consisting with the processing of both green tea and black tea. The varieties of variants assamica have high tannin content ranging from 33.6 to 34.1% that consistent with processed black tea, yellow tea or Pu'er-type tea.

Although reducing sugar content and amino acid in the tea less, only about 2-3% by weight of dry matter, but they are very important to the flavor and taste

of tea. Under the effect of high temperatures during drying tea, sugar is caramelized sweet fragrance form. Some amino acids have important implications for the producing tea, when they react with tannin and sugar will form aldehyde which increasing the aroma of tea.

Sugar content of variant TQLN is highest, average of 3.82%, inside Asatsuyu variety reached 5.01%, then Bat Tien also 4.53%, the lowest is Kanayamidori variety 2.91%. Assam variant has the lowest sugar content averages only 2.75%. Sugar not only reacts with the amino acids forming products flavorings, but a significant portion (glucose, fructose and sucrose) soluble in water, in extracts of tea, making tea has a sweet taste.

Amino acid is the most important criteria determining the aroma of the tea, variant TQLN is the highest amino acid content ranged from 2.02 to 2.86%. Then, Saemidori variety reached 2.86%, thereafter to Asatsuyu variety reach 2.5%. The varieties of variants Shan tea, Assam has the lowest levels of the amino acid only 1,72- 1.73%.

Moreover, catechin concentration, caffeine, substances dissolved in tea leaves are an important indicator of quality form of tea products.

The figures show that the high quality tea varieties of China, Taiwan and Japan have amino acids and reducing sugars content higher than the tea varieties of Vietnam. If we want to the Vietnam tea have good varieties for processing Oolong tea and green tea, we should focus on domestic breeding with high amino acids and reducing sugar content.

### ***Results of the initial plants selective prospects, with a high reducing sugar content and amino acids***

With the goal of select varieties with high yield and good quality, especially interested in breeding to processing green tea, Oolong tea, high quality; from 2001-2010, we have conducted 15 cross between the varieties have good quality with high-yield. Results obtained 1087 hybrid seeds and planting. By individual selection method has selected 44 outstanding individuals, cuttings and planting cuttings to selected lines, from which selected 15 promising lines. Initially, assess the growth, yield and quality of the selected lines obtained the following results:

**Table 2:** The ability growth of prospects tea lines (Age 2)

No	Hybrid pairs	Line	survival rate (%)	height (cm)	stem diameter (cm)	Number of branch level 1 (number)
1	KT/TRI777	204	91,67	69,05	1,05	7,55
2	KT/TRI777	205	100,00	79,6	1,68	5,5
3	KT/TRI777	207	91,67	78,09	1,75	6,64
4	KT/TRI777	209	75,00	71,33	1,47	7,67
5	KT/ TD	210	93,7	76,05	1,18	8,9
6	KT/TD	212	100,00	80,25	1,65	6,75
7	KT/TD	218	66,67	61,19	1,16	7,88
8	KT/TD	224	50,00	58,67	1,2	10,0
9	BT/TD	234	90,00	58,67	1,37	7,89
10	Kanayamidori/CT	238	83,33	87,3	1,27	9,0
11	Kanayamidori/CT	241	91,67	76,05	1,6	10,82
12	Okumidori/TD	248	91,67	70,95	1,34	6,6
13	Okumidori/TD	252	100,00	68,82	1,21	9,64
14	Asatsuyu/PH1	253	90,00	71,33	1,65	7
15	LV 2000/PH1	262	80,00	58,63	1,34	11,75
16	Kim Tuyen (control)		70	66,7	1,14	7,5

Subscribe growth indicators show that the tea lines are selective high survival rate, which is typically lines 205; 212 and 252 have the survival rate was 100%, and then to the line 204; 207; 241 and 248 was 90%; the lowest was line 224 only 50%. In general, the selected lines were higher survival rate than Kim Tuyen control.

Subscribe plant height show that the lines 212 and 238 have the greatest plant height was over 80 cm, the line 224; 234 and 262 have the lowest height were only about 58 cm and lower than the Kim Tuyen control.

Evaluation of plant height, stem diameter, survival rate and number of branch level 1 shows that lines 205 and 212 have the largest growth indicators, the lines 218 and 224 have the weakest growth indicators.

**Table 3:** Yield and productivity elements of the promising tea line (age 2)

No	Line	The weight bud+2 leaves	buds/plants	Số g/cây	Yeild (Tons/ha)
1	Line 204	0.5	174.8	87.4	1,70
2	Line 205	0.47	158.6	74.542	1,40
3	Line 207	0.49	130.6	63.994	1,28
4	Line 209	0.56	123.8	69.328	1,39
5	Line 210	0,74	229,3	169,68	3,39
6	Line 212	0.73	280.8	204.984	4,10
7	Line 218	0.54	145.6	78.624	1,572
8	Line 224	0.63	325.8	205.254	3,24
9	Line 234	0.56	289.5	162.12	3,24
10	Line 238	0.58	268.9	155.962	3,12
11	Line 241	0.47	209.7	98.559	1,97
12	Line 248	0.5	197.9	98.95	1,98
13	Line 252	0.57	265.8	151.506	3,03
14	Line 253	0.51	118.1	60.231	1,20
15	Line 262	0.34	141.7	48.178	0,96
16	Kim Tuyen (control)	0,6	210	126,00	2,52

Products of tea tree are buds and young leaves, so the shape and weight of tea shoots have a huge impact on productivity, quality of products. The large weight of tea shoots will give high yields, but the product looks rough, tea shoots with small weight will affect the productivity of tea.

The results in Table 2 shows the average weight of tea lines are small, ranges from 0.34 to 0.74 g/shoots. The weight of tea shoots of lines 210, 212 and 224 has the largest, average 0.60 - 0,7g/shoots and it is equivalent to the Kim Tuyen control. The lines 262; 205 and 241 have small weight shoots was less than 0.5 g/shoots, these lines have good appearance but will not high yield.

Number of shoot is indicator correlated to yield, the varieties have large number of shoot on the tree will be highly productive. Line 224 have the largest number shoot on plant reached 325.8 shoots /plant, then to lines 212 and 234 to reach about 280 shoots /plant, lines 253 and 209 were the lowest only about 120 shoots /plant.

results show that productivity of line 212 was highest yield reached 4.1 tons / ha, and then to the lines 224, 234, 238, 252 and 210 from 3.03 to 3.39 tonnes/ha, the Kim Tuyen control reached 2.52 tonnes / ha. The remaining lines are less productive than Kim Tuyen controls.

In addition to tracking the growth and yield of tea lines, we have evaluated the quality of tea through biochemical composition analysis and tasting quality of green tea.

**Table 4:** Result analysis of biochemical indicators, point organoleptic about quality of the promising hybrid tea line

No	Line	<i>Indicator analysis</i>					Point tasting
		Tanin (%)	solubles (%)	Amino acids (%)	Catechin (mg/gck)	Sugar (%)	
1	204	29,36	44,54	2,32	154,0	3,00	18,0
2	205	28,00	44,36	2,35	145,7	2,91	17,0
3	207	21,91	42,72	2,90	105,7	2,05	17,0
4	209	25,87	43,61	3,56	137,2	3,15	17,0
5	210	23,64	46,66	2,98	143,4	3,42	18,2
6	212	27,69	45,54	3,67	135,9	2,92	17,4
7	218	23,74	42,56	3,24	122,3	3,17	17,5
8	224	27,39	44,33	2,54	142,5	4,13	16,9
9	234	31,71	45,53	2,56	162,4	3,30	16,9
10	238	30,43	45,25	2,48	158,4	2,58	17,0
11	241	29,84	44,18	3,00	155,4	2,77	17,3
12	248	22,52	41,27	3,74	133,5	3,48	17,3
13	252	27,98	44,05	2,55	143,8	2,23	17,9
14	253	32,87	45,75	2,39	168,4	2,92	17,6
15	262	23,62	41,01	2,52	122,3	2,15	17,7
16	KT control	26,61	42,65	2,32	143,0	2,28	17,3

Results from table 4 shows the tea lines are not high tannin content, ranging from 21,91- 32.87%, lines 207, 210, 218, 248; and 262 have very low tannin content only about 22-24%. For the above tannin content will conform to processing high quality green tea and specialty tea. The two lines 253 and 234 have high tannin content only 31- 33% suitable for black tea processing.

Amino acids is chemical compounds that have a strong influence on the quality of tea, normally tea varieties in Vietnam only have amino acid content ranged from 1,0- 2,0%, so the quality is not high. Assessment group with over 1000

individuals from sexual hybrids, we selected out 15 individuals with largest amino acid content, ranging from 2.32 to 3.74%, which excellence are lines 209, 212, 218 and 248 over 3.24%.

Compared with the varieties use as parents in hybrid pairs showed that the tea lines are selected on the mother varieties are Kim Tuyen and Okumidori that two varieties have high amino acid content 2,42- 2,74%. Thus, the probability to select hybrids with high levels should implement hybrid combinations with mother variety have high amino acid content.

Assess sugar content shows that all selected lines have high sugar content ranged from 2.05 to 4.13%, while the lines 210, 218; 224; 234 and 248 have the highest sugar content of 3.0-to 4.2%, the other lines have higher sugar content than or equal to Kim Tuyen control, the only line 207 is lower than the controls. Evaluation results quality green tea show that the tea lines have good quality green tea, typically line 210 with green tea is very good quality, aroma, flavor characteristics, reaching 18, 2 points are classified as good. Then, the lines 204; 205; 218; 248; 252; 253 and 262 with good quality green tea, aroma, its flavor is mild with a total score approximately 18 points.

From the above results show that the lines 210; 212; 205; 218; 248; 252; 253 and 262 have the strong growth indicators, high productivity and good quality raw materials, good quality tea product.

## Conclusions

- The high quality tea varieties from China, Taiwan and Japan have content of amino acids and reducing sugars higher than Vietnam's tea varieties.
- Results of the evaluation of amino acid content, reducing sugars and tannins combined with assessment growth capacity, yield of 15 promising lines of tea, have been identified:
  - + The lines 209, 218, 224 have amino acids content over 3% and less than 30% tannin content. Green tea tasting are achieved with good point.
  - + The lines 210, 218, and 224 have a reducing sugar content higher than 3% and less than 30% tannin content. Especially, line 224 very high reducing sugar content (4.13%). Green tea tasting are achieved with good point.
  - + Line 212 have strong growth (age 2 yields reached 4.1 tons/ha), high amino acid content (3.67%), high reducing sugar content (2.93%), low tannin content (27.69% ), green tea tasting with good point (18.2 points), and suitable for processing high-quality green tea.

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