

# Response of Plain Black Tea Parameters, Individual Theaflavins and Yields Due to Location of Production and Clones within Lake Victoria Basin

P Okinda Owuor<sup>1</sup>, Peter O Ogola<sup>1</sup>, Samson M Kamunya<sup>2</sup>

## ABSTRACT

Tea (*Camellia sinensis*) is a major cash crop and leading foreign exchange earner, contributing to poverty alleviation by providing employment and livelihood to many stakeholders in the producing countries. Production has increased faster than consumption causing price stagnation, especially for CTC black teas. Kenya is the third-largest tea producer and Lake Victoria Basin produces over 60% of her tea. Selection of tea cultivars in Kenya has been centered in one location before the selected clones are introduced to other growth environments. This study evaluated if tea clones maintain their yield and plain black tea quality attributes when grown at different locations within Lake Victoria Basin. The basin produces mainly plain black teas whose quality is due to levels of polyphenolic compounds, especially green leaf flavan-3-ols that are oxidized to theaflavins and thearubigins during black tea processing. The theaflavins and thearubigins contribute to the color and brightness of black teas. The trials were done in two sites Timbilil and Kipkebe using twenty clones. All the plain tea quality parameters including individual theaflavins and yields varied ( $p < 0.05$ ) with clones, demonstrating diversity in the cultivars used. The levels of the parameters and yields also changed ( $p < 0.05$ ) with the location of production. These results demonstrated that clonal tea quality and yields vary depending on the geographical location of production. There were also significant interactions effects between the clones and location of production in the quality parameters and clones showing the extent of the changes varied from clone to clone. Indeed the relative ranking of the clones varied with location. No clone retained its relative superiority ranking at the two locations. Both the Spearman correlation coefficients (rs) and the Pearson correlation coefficients (r) between the individual parameters were positive but low and insignificant, except for theaflavin-3-gallate. These results demonstrate the need for location-specific evaluation of both new and old clones to establish clonal yield and quality potentials in new locations of cultivation.

**Keywords:** Tea, Individual theaflavins, Quality, Yields.

*International Journal of Tea Science* (2019); DOI: 10.20425/ijts1413

## INTRODUCTION

Tea beverages from *Camellia sinensis* (L) O. Kuntze are most widely consumed fluids after water,<sup>1</sup> and are gaining popularity as important "health drinks".<sup>2,3</sup> Their production has therefore risen fast in the last few decades.<sup>4</sup> The fast growth of the production subsector has, however, not been accompanied by a concomitant rise in consumption. Consequently, tea prices have stagnated or declined over time,<sup>5,6</sup> especially for the "cut, tear and curl" (CTC) black teas. Kenya, the third-largest producer of tea after China and India is the main producer of CTC black teas and the leading exporter of black tea.<sup>4</sup> Tea plants are economically produced in different parts of the world, between longitudes ranging from 45°N in the Carpathians and 35°S in Natal<sup>7</sup> at altitudes ranging from sea level in Japan and Sri Lanka<sup>8</sup> to 2,700 m above mean sea level Olenguruone (Kenya) and Gisovu (Rwanda).<sup>9</sup> In Kenya, approximately 60% of the tea is produced in the west of the Great Rift Valley in Lake Victoria Basin.

To improve tea productivity and production, continuous efforts are directed at developing appropriate agronomic,<sup>10-12</sup> and processing technologies.<sup>10,11</sup> Such efforts include development and identification of high yielding and high-quality tea cultivars.<sup>12,13</sup> Most of the breeding/selection research have, however, been done in a single location, on the assumption that plants maintain desirable traits wherever they are cultivated. Consequently, developed tea cultivars have been introduced in different ecological zones within Kenya and other African countries without further re-evaluation for suitability. But studies have demonstrated wide response ranges among tea genotypes to different environments.<sup>14-17</sup> The tea-growing environment

<sup>1</sup>Department of Chemistry, Maseno University, Maseno, Kenya

<sup>2</sup>Tea Research Institute, Kenya Agricultural and Livestock Research Organization, Kericho, Kenya

**Corresponding Author:** P Okinda Owuor, Department of Chemistry, Maseno University, Maseno, Kenya, e-mail: okindaowuor@maseno.ac.ke

**How to cite this article:** Owuor PO, Ogola PO, Kamunya SM. Response of Plain Black Tea Parameters, Individual Theaflavins and Yields Due to Location of Production and Clones within Lake Victoria Basin. *International Journal of Tea Science* 2019; 14(1):14-25

**Source of support:** Inter-University Council of East Africa, Lake Victoria Research Initiative (VicRes)

**Conflict of interest:** None

has profound effects on growth, productivity,<sup>14,15</sup> leaf chemical composition<sup>16-19</sup> and quality of tea.<sup>16,20</sup> Environmental factors such as soil moisture content<sup>21,22</sup> and temperatures<sup>23</sup> influence tea yields. Tea cultivars do not respond to variations in these environmental factors in the same patterns<sup>24</sup> causing unpredictable yield responses over different environments.<sup>25</sup> Unfortunately, tea growers traditionally import new cultivars based on information at the development site without re-evaluating suitability in new growing locations.

Black tea quality and yields vary in different environments even when the genetic make of the plants and agronomic inputs are the same. For example, for Kenya and Malawi black teas produced from the same clones under similar agronomic and processing conditions had varied chemical quality parameters.<sup>26</sup> Yields of same tea clone grown in Tanzania, Rwanda and Kenya but receiving similar agronomic inputs differed with the region of production.<sup>27</sup> Such

variations were attributed to large differences in environmental factors affecting growth and metabolism in the countries. Similar observations were made on yield and black tea quality produced within one country.<sup>16,20</sup> The variations in plain black tea quality parameters of tea clones with the location of production did not follow the same pattern.<sup>28</sup> Similarly, green leaf quality precursors varied in the same clone due to the location of production.<sup>18,19</sup> The yield and black tea quality variations, especially the new tea varieties in different locations within the Lake Victoria Basin are not documented.

Most Kenya black teas are classified as plain to medium flavoured teas. Such black teas are valued for the plain quality parameters, i.e., theaflavins, thearubigins, and caffeine.<sup>29-31</sup> Some studies demonstrated a relationship between black tea valuation and theaflavins content.<sup>32-36</sup> Such relationships were however not successful for Kenya<sup>37,38</sup> and Sri Lanka<sup>39</sup> black tea. Black tea contains four major theaflavins (theaflavin, theaflavin-3-gallate, theaflavin-3'-gallate and theaflavin-3, 3'-digallate (Fig. 1)).<sup>40</sup> The individual theaflavins contribute to astringency differently.<sup>41</sup> Theaflavin digallate is 6.4 times more astringent than theaflavin while theaflavin monogallates are 2.22 times more astringent. Tea cultivars that possess high levels of theaflavin digallate have high quality.<sup>38,42-46</sup> An astringency normalizing factor, theaflavin digallate equivalent was developed to cater to the differential contribution of individual theaflavins to black tea astringency<sup>47</sup> and later improved to measure astringency more correctly.<sup>45</sup> Good relationships exist between theaflavin digallate equivalent and sensory evaluation for Kenyan, Malawian and South African teas.<sup>38</sup> Thearubigins are responsible for the color and thickness of black tea.<sup>31</sup> Very high thearubigins levels reduce liquor brightness and hence tend to lower the sensory evaluation of black teas.<sup>38</sup> The mode of variations in theaflavin, the individual theaflavins compositions with a geographical area of production in clonal tea new cultivars within the Lake Victoria Basin is not documented. This research evaluated the variations of tea quality parameters and yields due to genotypes and locations of cultivation among some tea cultivars grown within Lake Victoria Basin.

## METHODOLOGY

### Site Description and Research Design

The trial comprising twenty clones, ten of which were popular cultivars (TRFK 6/8, TRFK 12/12, TRFK 12/19, TRFK 303/1199, TRFK 303/577,

TRFK 31/11, TRFK 7/3, TRFK 100/5, TRFK 11/4, TRFK 31/8) and ten new improved cultivars (TRFK 301/5, TRFK 430/90, TRFK 371/3, TRFK 6/10, EPK C12, TRFK 12/56, TRFK 306/1, BBK BB35, TRFK 301/4, TRFK 301/6) was established in two major tea growing locations within Lake Victoria Basin: Timbilil (0° 22' S, 35° 21' E, 2180M AMSL) and Kipkebe (0° 35' S, 35° 5' E, 1800M AMSL). Cultivar TRFK 6/8 was used as a control for quality and yield traits. The trial was set up in a randomized complete block design with three replications in plots of 30 plants spaced at 0.61 m within rows and 1.22 m between rows and received 150 Kg N per hectare per year in the form of NPKS 25:5:5:5 fertilizer. The trials in both sites were subjected to recommended agronomic management.<sup>48</sup>

Two leaves and a bud was harvested every 7 to 10 days and converted into made tea per hectare (mt/ha) by a conversion factor of 0.225.<sup>48</sup>

From each plot, 1200 g of leaf comprising of two leaves and a bud was collected for miniature black tea manufacturing using CTC procedure.<sup>26,37,49</sup> Total theaflavins were determined by Flavognost method.<sup>50</sup> The individual theaflavin ratios were determined by HPLC.<sup>51-53</sup> Liquors were prepared by adding 4 g of black tea to 195 mL deionized water that has just reached boiling and shaking done for 10 min in a 475 mL capacity thermos flask. Clean liquor was obtained by filtration through cotton wool. The hot liquor was cooled to room temperature by placing the flask containing the liquor under a cold water tap (1-3 min). The liquor was diluted (1:1) with water before HPLC analysis. The liquor was analyzed using a Hypersil 5 I ODS column (25 cm x 4.6 mm) and monitored at a UV wavelength of 365 nm and results recorded and analyzed using a JC600 Cecil data system. Solvent A was 1% aqueous acetic acid and Solvent B acetonitrile. A linear gradient from 8-31% solvent B over 60 min with a flow rate of 1.5 mL per minute was used.<sup>51,52</sup> The theaflavin ratios calculated from the HPLC data and the Flavognost (total) theaflavins data were used to calculate the amounts of the individual theaflavins, since the molar absorption coefficients of the four theaflavins are similar at 365 nm.<sup>53</sup> Astringency normalizing factor (Theaflavin digallate Equivalent) was calculated according to the improved equation.<sup>45</sup> Thearubigins total color and brightness were determined as described by Roberts and Smith.<sup>54</sup>

Data were analyzed using randomized complete block design in a factorial 2 arrangement, with sites as the main treatments and clones as sub treatments using GenStat statistical package.

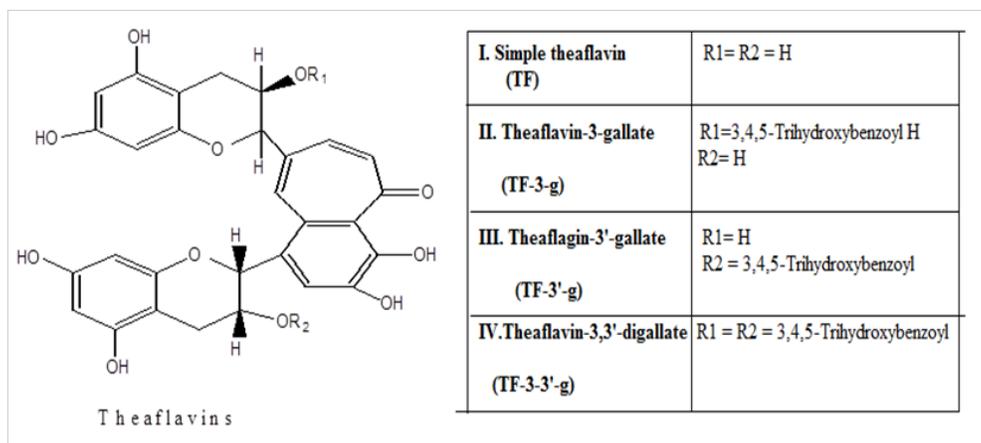


Fig. 1: The major individual theaflavins in black tea

## RESULTS AND DISCUSSION

## Effect of Clones and Locations of Production on Brightness of Black Tea

The changes in black tea brightness with clones and locations of cultivation are presented in Table 1. Brightness levels differed

( $p \leq 0.05$ ) between the clones, possibly due to genetic differences among the tea clones causing variations in the polyphenol content in tea shoots.<sup>55</sup> The clones used in the study belonged to three main varieties i.e., *Camellia sinensis* var. *assamica*, *Camellia sinensis* var. *sinensis* and *Camellia sinensis* sub-*spp* *Lasiocalyx* that are genetically different.<sup>19</sup> A comparison between the newly

Tables 1: Variations in theaflavins and brightness of clonal tea with region of production

Clone	Brightness			Ranking brightness			Total color			Ranking		
	Location		Mean clone	Location		Mean clone	Location		Mean clone	Location		Mean clone
	Kipkebe	Timbilil		Kipkebe	Timbilil		Kipkebe	Timbilil		Kipkebe	Timbilil	
TRFK 6/8	21.09	21.15	20.62	9	7	8	4.80	5.19	4.09	3	5	19
TRFK 12/12	35.67	22.82	29.24	1	4	1	5.24	5.05	5.15	2	7	3
TRFK 303/577	16.63	14.15	15.39	15	18	16	3.94	5.02	4.48	20	8	10
TRFK 303/1199	16.28	21.55	18.91	16	5	11	4.70	4.09	4.39	7	18	13
TRFK 7/3	19.26	18.23	18.74	11	13	13	4.56	5.08	4.82	9	6	6
TRFK 100/5	24.84	18.76	21.80	5	11	6	4.52	6.12	5.32	10	2	2
TRFK 11/4	20.12	18.65	19.38	10	12	10	4.80	4.76	4.78	4	9	8
TRFK 31/8	22.79	23.75	23.27	8	2	4	4.25	4.49	4.37	14	12	14
TRFK 12/19	10.80	19.62	15.21	20	9	17	4.31	4.42	4.37	13	14	15
TRFK 31/11	14.05	17.12	15.58	19	14	15	4.47	4.44	4.45	11	13	11
TRFK 301/5	18.28	19.50	18.89	13	10	12	4.32	5.30	4.81	12	4	7
TRFK 301/6	18.59	10.71	14.65	12	19	18	4.25	3.92	4.08	15	19	20
TRFK 430/90	23.08	15.52	20.80	7	16	7	4.79	5.52	4.91	6	3	4
TRFK 371/3	25.01	24.17	24.59	4	1	2	6.04	3.77	4.91	1	20	5
TRFK 6/10	25.06	20.09	22.57	3	8	5	4.20	4.60	4.40	17	10	12
EPK C12	17.56	23.34	10.45	14	3	20	4.64	6.48	5.56	8	1	1
TRFK 12/56	15.09	17.06	16.07	17	15	14	4.07	4.36	4.21	19	15	18
BBK BB35	26.27	21.35	23.81	2	6	3	4.23	4.24	4.24	16	17	17
TRFK 301/4	24.40	15.13	19.76	6	17	9	4.80	4.49	4.64	5	11	9
TRFK ST 306	14.55	8.99	11.77	18	20	19	4.20	4.32	4.26	18	16	16
Mean location	20.47	18.68					4.53	4.76				
CV (%)		21.7						15.00				
LSD, $p \leq 0.05$		1.55	4.89					NS				0.80
Interactions		6.91						1.14				



developed clones and the old popular clones revealed that the new clones produced brighter black teas than the old cultivars, although this did not reach a significant level. The new clones, therefore, have the potential to produce black tea whose brightness qualities are comparable to old popular ones. All clones produced black tea with quality comparable or better than clone TRFK 6/8 (quality standard clone) except clones TRFK 303/577, TRFK 12/19 and TRFK 306/1, and seedling stock TRFK ST306 which had lower ( $p \leq 0.05$ ). Clone TRFK 12/12 produced black tea with brightness higher ( $p \leq 0.05$ ) than TRFK 6/8.

The black tea brightness differed ( $p \leq 0.05$ ) with the location of production (Table 1). These results confirmed the previous findings<sup>26,28,38</sup> and showed that clonal teas do not retain their brightness attributes when grown in different environments. In other studies, tea plants responded differently to different growth environment<sup>24,25</sup> that caused variations in black tea quality parameters.<sup>16,20,26,28,37</sup> Since the two locations under study were within the same tea growing zone in the Lake Victoria Basin, the results confirmed previous findings that even within a radius of 10 km variations in black tea quality parameters occur.<sup>56,57</sup> When the clones are ranked in the two sites, it is observed that most of the clones did not retain their relative ranking in the two sites. Indeed both Spearman's rank correlation ( $r_s = 0.40$ ) and Pearson's correlation coefficient ( $r = 0.39$ ) (Table 2) were weak. Also, there was a significant ( $p \leq 0.05$ ) interactions effect between clones and the location of production. The low correlation coefficients and significant interactions effects demonstrate that the brightness of the tea with locations of production was not varying in the same patterns in the same clone at different sites. Thus black tea brightness of a clone at a specific location cannot be used to accurately predict the brightness of the same clone in another location.

**Variations in Black Tea Total Color due to Clones and Location of Production**

There were significant ( $p \leq 0.05$ ) total color variations due to clones and locations of cultivation (Table 1). The clonal differences were attributed to genetic variations among the tea plants,<sup>58</sup> as observed for black tea brightness (Table 1). These results confirmed previous findings where black tea color varied with clones.<sup>46,58</sup> The total color level of the newly developed clones and the popular

widely cultivated clones were similar. This implied that both the newly developed clones and the popular widely cultivated ones would produce tea infusions of almost similar color. When the individual clones were compared to quality standard clone TRFK 6/8, only clones TRFK 12/12, TRFK 100/5, TRFK 430/90 and EPK C12 had significantly ( $p \leq 0.05$ ) higher values. All other clones had TC values comparable to TRFK 6/8. Although in some markets, colory black teas may be preferred, usually black tea with high total color tends to be very thick but taste muddy and undesirable.<sup>31</sup>

Overall, the location of production did not have a significant influence on clonal black tea total color. This implied the total color was insensitive to the area of production. These findings contradicted previous findings where black tea total color changed with the region of production.<sup>37,59-64</sup> It is likely that the difference could be due to the fact that in the same basin. However, a closer data evaluation revealed that in terms of ranking, all the clones did not retain their relative rank positions at the two locations (Table 1). Moreover, there was a significant ( $p \leq 0.05$ ) interactions effect between clones and the location of production. The clones performed differently in the two locations of production, with most of them not retaining their rank in the two sites. The Pearson correlation coefficient ( $r = -0.05$ ) and Spearman's rank correlation ( $r_s = 0.270$ ) (Table 2) were very low. This suggested that response of the individual clones in production of total color varied with locations in an unpredictable manner.

**Changes in Black Tea Thearubigins due to Clones and Location of Production**

Thearubigins impart color, body, and thickness to black tea.<sup>29-31</sup> However high thearubigins levels reduce liquor brightness.<sup>29-31</sup> There were clonal significant differences ( $p \leq 0.05$ ) in thearubigins levels (Table 3). This was due to genetic differences among the tea clones, that resulted in the accumulation of varying green leaf polyphenols during the growth of the tea plant.<sup>55</sup> The clones assessed were from different varieties. Clones TRFK 12/19, TRFK 31/8, TRFK 12/12, TRFK 31/11, TRFK 31/11, TRFK 12/12, TRFK 306/1, BB 35, TRFK 430/90, TRFK 6/8, TRFK 7/3, TRFK 6/10, TRFK 371/3, TRFK 100/5, TRFK 11/4 and TRFK 12/56 were *Camellia sinensis* var. *assamica* species, while EPK C12 was *Camellia sinensis* var. *sinensis* variety, TRFK 301/4, TRFK 301/5, TRFK 301/6 were *Camellia sinensis* sub-*spp* *Lasiocalyx* clones and e.g., TRFK 303/577 and TRFK 303/1199

**Table 2:** Pearson correlation coefficients, regression coefficients and Spearman correlation coefficients of the individual parameters in clonal tea produced in different locations

	<i>Theaflavins</i>	<i>Brightness</i>	<i>Thearubigins</i>	<i>Total colour</i>
r	0.29	0.39	-0.09	-0.05
r <sup>2</sup>	0.08	0.15	0.01	0.003
r <sub>s</sub>	0.36	0.40	0.06	0.27
	<i>Simple theaflavins</i>	<i>Theaflavin-3-gallate</i>	<i>Theaflavin-3'-gallate</i>	<i>Theafavin-3,3;-digallate</i>
r	0.4389	0.62	0.04	0.30
r <sup>2</sup>	0.1926	0.39	0.002	0.09
r <sub>s</sub>	0.40	0.70	0.12	0.43
	<i>Theaflavin digallate equivalent</i>	<i>Yield</i>		
R	0.30	0.33		
r <sup>2</sup>	0.09	0.11		
r <sub>s</sub>	0.26	0.51		

are crosses between *Camellia sinensis* var. *assamica* and *Camellia sinensis* var. *sinensis*. Even the clones from the same variety had different parents and were genetically diverse. Previously, green tea catechins in these clones exhibited a significant difference ( $p \leq 0.05$ ).<sup>19</sup> This largely contributed to differences in the clonal black tea thearubigins levels. Similar clonal differences in thearubigins had been recorded.<sup>20,26,28,38</sup> The lowest thearubigins level was observed in TRFK 31/8 while the highest levels were recorded in TRFK 301/4 and TRFK 306/1. Both TRFK 301/4 and TRFK 306/1 were new clones. TRFK 301/4 produced thearubigins values higher ( $p \leq 0.05$ ) than TRFK 6/8, but the value of thearubigins in TRFK 306/1 was not different ( $p \leq 0.05$ ) from that of TRFK 6/8. However, the high concentrations of TR in clone TRFK 306/1 needs further investigation since 306/1 is low in catechins,<sup>65</sup> and is mainly rich in anthocyanin. The clone was developed for production of unfermented purple tea. There could have been other unrelated compounds that could have been classified as thearubigins. Usually, high amounts of flavonol glycosides are measured together with thearubigins fraction hence overstating the concentration of the thearubigins.<sup>51,52</sup> Some of the clones studied might have had high flavonol glycosides concentration. The clones under investigation had thearubigins levels comparable to TRFK 6/8 except for TRFK 6/10, TRFK 31/8 and TRFK 301/4. Comparison of the newly developed clones with old clones, the new clones had higher levels of thearubigins, though the difference did not reach not significant ( $p \leq 0.05$ ) level.

Thearubigins are responsible for color and thickness<sup>49</sup> of black tea. The thearubigins levels were expected to mirror those of total color, but this not the case. This discrepancy noted be due to the high amount of flavonol glycosides<sup>66</sup> measured together with thearubigins in the Robert and Smith method,<sup>67</sup> leading to an overstated value of thearubigins. The method does not discriminate between the flavonol glycosides and thearubigins.<sup>51,66</sup> Moreover, the correlation between color and thearubigins was weak and insignificant ( $r = 0.06$ ). The observation was against popular belief in the tea industry that thearubigins levels influence the total color.

There were significant ( $p \leq 0.05$ ) differences in thearubigins levels with the location of cultivation (Table 3). These were explained in terms of differences in the factors that affect growth in the two sites. These factors lead to different levels of polyphenols in tea leaves.<sup>17-20</sup> For teas grown in Kenya and Malawi, Kenyan teas contained higher levels of thearubigins than Malawi teas<sup>20,26</sup> due to differences in growth factors prevailing in the two countries. Conditions in Malawi favor faster growth rates than those in Kenya. A similar trend was noted for the two sites under investigation. The Kipkebe site experiences weather patterns that favor faster rates of growth than Timbilil.

The relative ranking of the clones (Table 3) from the two sites showed that most of the clones did not retain their ranks in the two sites. The Pearson correlation coefficient and Spearman's rank correlations between the thearubigins levels in clonal black tea from the two sites were weak (Table 2). The interactions effects between clones and location of production site were also significant ( $p \leq 0.05$ ) confirming that the levels of thearubigins were not changing in the same pattern in same clones at the two locations. The pattern was unpredictable.

### Clonal and Locational Variations of Plain Black Tea Total Theaflavins

Theaflavins levels are critical plain black tea quality parameter responsible for briskness, brightness, strength, and color of

black tea.<sup>29-31</sup> The total theaflavins (Table 3) varied with clones and location of production. The clonal black tea content of total theaflavins exceeded the minimum threshold UNCTAD recommended levels for export tea.<sup>68</sup> This demonstrates all clones were suitable for black tea production, even though a clone like TRFK 306/1 was developed for the production of purple-green tea. The total theaflavins levels changed ( $p \leq 0.05$ ) with the clone, similar to previous studies.<sup>26,28,37,38</sup> The popular old widely cultivated clones had slightly higher total theaflavins levels, which did not differ significantly from the new clones. These showed new cultivars have potential producing black tea with quality comparable to the old popular clones. Some clones (TRFK 6/10, EPK C12 and BBK BB35) had theaflavins amounts comparable to the quality standard clone (TRFK 6/8),<sup>13</sup> while TRFK 371/3 had total theaflavin amounts significantly higher than TRFK 6/8.

The overall effect of the location of production on the total theaflavin among the clones not significant. The results contradict the previous findings<sup>16,20,26,28</sup> that black tea plain quality parameters such as total theaflavins vary with a geographical area of production. This was possibly due to the clones being produced within one basin. The relative ranking of clones of the clone in respect to total theaflavins varied with location. For example, TRFK 371/3 was the best clone in Kipkebe but ranked twelfth in Timbilil. Similarly, the best-ranked clone in Timbilil was TRFK 301/5, but the tenth in Kipkebe. More strikingly, TRFK 100/5 which ranked third in Kipkebe was eighteenth in Timbilil and TRFK 430/90 which was sixth in Timbilil ranked fifteenth in Kipkebe. Both the Spearman's rank correlation and Pearson correlation coefficient for total theaflavins were also low (Table 2). Indeed, the effects of the interactions between clones and the location of production of total theaflavins levels were significant. These results showed that the change in total theaflavins levels of clonal tea was with the location was not systematic and predictable.

Tables 4, 5, and 6 present the variations in levels of individual theaflavins and theaflavin digallate equivalent (TFDGeq) with clones and location of cultivation. In most clones the order of concentration of the individual theaflavins was TF-3-g > TF-3'-g > TF-3,3'-dg. The order was slightly different from that observed for southern and central African teas,<sup>36</sup> where simple theaflavin was the dominant theaflavin. In some clones, of individual theaflavins in TRFK 6/8, TRFK 303/577, TRFK 11/4, TRFK 301/5 and TRFK 6/10 were, however, similar to the southern and central African.<sup>36,38</sup> Similar to an earlier study<sup>38</sup> dominant levels of theaflavin digallate was observed in clone 31/11. These results demonstrate the individual theaflavins may not be used as markers for the location of origin. These differences in the levels of individual theaflavins were attributed to genetic differences among the clones assessed leading to differences in the ability of the clones to form individual theaflavins and reaffirmed previous findings<sup>38,42</sup> that genetic variations in the tea plants are an important factor that affects the distribution of black tea theaflavins and hence tea quality. The significant difference in the individual theaflavins with clone ( $p \leq 0.05$ ) demonstrated the differences in the ability of the clones to produce black tea of different quality. Significant clonal differences in green leaf catechins had been recorded for these clones.<sup>19</sup> The observed differences in this study in the total and individual theaflavins arose from the catechins variations.

Theaflavin digallate equivalent, (TFDGeq) has been demonstrated as a superior black tea quality indicator than total theaflavins.<sup>42,44,46</sup> The clonal TFDGeq values were significantly ( $p \leq 0.05$ ) different. The mean TFDGeq levels for the old popular



**Table 3:** Changes in clonal black tea thearubigins and total color due to location of production

Clone	Thearubigins (%)			Ranking			Total theaflavins			Ranking total theaflavins		
	Location		Mean clone	Location		Mean clone	Location		Mean Clone	Location		Mean clone
	Kipkebe	Timbillil	clone	Kipkebe	Timbillil	clone	Kipkebe	Timbillil	Clone	Kipkebe	Timbillil	clone
TRFK 6/8	14.95	16.62	15.78	10	5	7	21.14	23.57	22.36	5	3	5
TRFK 12/12	12.13	15.17	13.74	20	12	18	32.89	22.37	27.63	1	5	1
TRFK 303/577	14.60	17.05	15.82	11	4	6	18.68	19.05	18.86	14	12	14
TRFK 303/1199	15.10	15.60	15.35	9	10	9	19.21	18.42	18.81	12	13	15
TRFK 7/3	15.85	16.40	16.12	4	6	3	19.29	21.40	20.34	11	7	9
TRFK 100/5	12.83	17.37	15.10	17	3	11	26.13	17.27	21.76	3	18	6
TRFK 11/4	14.35	14.44	14.39	13	15	17	20.75	17.43	19.09	8	17	13
TRFK 31/8	12.35	14.11	13.23	19	16	20	19.01	21.03	20.02	13	9	10
TRFK 12/19	16.22	13.95	15.08	2	18	12	14.74	18.14	16.44	17	15	17
TRFK 31/11	14.52	14.55	14.98	12	14	13	13.88	17.85	15.86	19	16	18
TRFK 301/5	14.29	15.94	14.86	15	9	14	19.72	25.67	22.70	10	1	4
TRFK 301/6	15.75	15.94	15.84	6	8	4	17.28	14.15	15.72	16	19	19
TRFK 430/90	14.32	16.11	15.21	14	7	10	17.59	22.16	19.88	15	6	11
TRFK 371/3	19.42	12.34	15.83	1	20	5	32.40	19.28	25.84	2	11	2
TRFK 6/10	12.80	13.77	13.28	18	19	19	20.91	20.39	20.65	7	10	8
EPK C12	16.18	15.03	15.60	3	13	8	22.34	23.70	23.02	4	2	3
TRFK 12/56	15.44	14.06	14.75	7	17	15	14.74	22.88	18.31	18	4	16
BBK BB35	13.40	15.53	14.46	16	11	16	21.12	21.30	21.21	6	8	7
TRFK 301/4	15.78	21.90	18.84	5	1	1	20.41	18.21	19.31	9	14	12
TRFK ST 306	15.26	17.56	16.41	8	2	2	11.11	13.23	12.18	20	20	20
Mean Location	14.83	15.64					20.17	19.86				
CV (%)		11.9						12.06				
LSD, p ≤0.05		0.66	2.09					NS	2.04			
Interactions		2.95						2.88				

**Table 4:** Response of clonal tea to production of simple theaflavin and theaflavin-3-gallate to location of production

Clone	Simple TF						TF-3-gallate						Ranking		
	Location		Mean	Location		Mean	Location		Mean	Location		Mean	Location		Mean
	Kipkebe	Timbilil	clone	Kipkebe	Timbilil	clone	Kipkebe	Timbilil	clone	Kipkebe	Timbilil	clone	Kipkebe	Timbilil	clone
TRFK 6/8	7.06	7.81	7.43	5	2	2	7.83	5.84	6.84	2	3	1			
TRFK 12/12	6.98	4.70	5.84	6	15	15	7.99	5.39	6.69	1	5	2			
TRFK 303/577	5.75	6.04	5.90	11	7	7	4.79	4.41	4.60	10	13	12			
TRFK 303/1199	4.05	4.79	4.42	16	14	14	4.59	4.85	4.72	12	10	11			
TRFK 7/3	5.78	6.45	6.11	10	3	3	4.50	5.08	4.78	13	8	10			
TRFK 100/5	7.13	4.62	5.87	4	16	16	7.12	4.49	5.80	4	12	4			
TRFK 11/4	10.41	3.47	6.94	1	18	18	2.84	4.28	3.56	18	15	17			
TRFK 31/8	5.58	5.02	5.30	12	12	12	5.56	6.32	5.44	6	1	6			
TRFK 12/19	3.03	4.84	4.39	18	13	13	3.73	4.23	3.79	14	16	15			
TRFK 31/11	2.22	2.16	2.19	20	20	20	4.99	3.62	4.30	9	17	14			
TRFK 301/5	9.09	11.96	10.50	2	1	1	3.16	4.40	3.78	16	14	16			
TRFK 301/6	5.32	4.34	4.83	14	17	17	2.69	2.22	2.47	19	20	20			
TRFK 430/90	5.45	6.17	5.81	13	4	4	4.77	5.52	4.90	11	4	9			
TRFK 371/3	8.51	5.25	6.68	3	11	11	7.74	4.75	6.25	3	11	3			
TRFK 6/10	6.33	6.08	3.21	7	5	5	5.47	5.11	5.29	7	7	8			
EPK C12	5.91	6.05	5.98	8	6	6	5.40	5.96	5.65	8	2	5			
TRFK 12/56	3.84	5.64	4.74	17	8	8	3.67	5.03	4.35	15	9	13			
BBK BB35	5.27	5.26	5.26	15	10	10	5.60	5.18	5.39	5	6	7			
TRFK 301/4	5.89	5.28	5.59	9	9	9	2.99	2.85	2.92	17	18	18			
TRFK ST 306	2.78	3.43	3.10	19	19	19	2.47	2.58	2.53	20	19	19			
Mean location	5.86	5.47					4.85	4.56							
CV (%)		28.70						23.60							
LSD, $p \leq 0.05$		0.30	0.94					NS	1.27						
Interactions		1.33						1.80							



**Table 5:** The changes in clonal tea and rankings of theaflavin-3'-gallate and theaflavin-3,3'-digallate due to location of production

Clone	TF-3'-Gallate				TF-3,3'-digallate				Ranking	
	Location		Mean	Ranking	Location		Mean	Ranking	Location	Ranking
	Kipkebe	Timbillil	clone	Location	Kipkebe	Timbillil	clone	Location	Kipkebe	Timbillil
TRFK 6/8	3.95	7.04	5.49	17	6	15	3.30	2.88	9	16
TRFK 12/12	10.53	7.36	8.94	1	5	1	7.39	4.92	1	2
TRFK 303/577	5.46	5.88	5.67	11	14	14	2.68	2.71	15	17
TRFK 303/1199	6.39	5.49	5.94	6	18	13	4.18	3.28	6	9
TRFK 7/3	6.23	6.82	6.53	7	8	5	2.78	3.05	14	14
TRFK 100/5	7.64	5.21	6.42	3	19	7	4.24	2.96	5	15
TRFK 11/4	4.39	5.62	5.01	16	16	17	3.11	4.06	11	6
TRFK 31/8	2.78	6.54	4.66	20	10	18	5.09	4.15	4	4
TRFK 12/19	4.88	5.90	5.39	13	13	16	2.59	3.16	17	10
TRFK 31/11	3.37	5.65	4.51	18	15	19	3.30	6.42	10	1
TRFK 301/5	6.13	7.61	6.87	9	3	4	1.35	1.70	20	20
TRFK 301/6	7.01	5.61	6.31	4	17	8	2.22	1.96	19	19
TRFK 430/90	5.27	7.03	6.15	12	7	10	2.60	3.45	16	8
TRFK 371/3	10.22	6.11	8.16	2	11	2	5.93	3.10	3	12
TRFK 6/10	6.01	6.08	6.05	10	12	11	3.10	3.11	12	11
EPK C12	6.91	7.42	7.16	5	4	3	4.12	4.28	7	3
TRFK 12/56	4.41	7.65	6.03	15	2	12	2.82	3.56	13	7
BBK BB35	6.23	6.76	6.47	8	9	6	4.02	4.10	8	5
TRFK 301/4	4.61	7.68	6.15	14	1	9	6.92	2.40	2	18
TRFK ST 306	3.37	4.18	3.77	19	20	20	2.49	3.07	18	13
Mean location	5.79	6.38					3.66	3.42		
CV (%)		20.60						33.40		
LSD, $p \leq 0.05$		0.46	1.44					NS	1.39	
Interactions		2.03						1.92		

**Table 6:** Response rankings of clonal tea theaflavin digallate equivalent (TFDGE) and yields to location of production

Clone	TFDGE			TFDGE ranking			Yields			Yields ranking		
	Location		Mean Clone	Location		Mean Clone	Location		Mean Clone	Location		Mean Clone
	Kipkebe	Timbillil		Kipkebe	Timbillil		Kipkebe	Timbillil		Kipkebe	Timbillil	
TRFK 6/8	7.49	8.57	8.43	10	8	9	2413	1914	2164	13	9	13
TRFK 12/12	14.90	10.08	12.49	1	1	1	2250	1384	1817	16	17	17
TRFK 303/577	7.13	4.23	7.18	13	20	16	2952	2158	2555	9	4	6
TRFK 303/1199	8.62	7.62	8.12	8	14	11	2278	2048	2163	15	6	14
TRFK 7/3	7.41	8.18	7.79	11	9	12	2033	1307	1670	19	20	19
TRFK 100/5	10.48	7.04	8.76	3	16	7	2063	1630	1847	18	13	16
TRFK 11/4	7.25	8.04	7.64	12	10	14	2370	1566	1943	14	14	15
TRFK 31/8	8.85	9.04	8.95	7	5	6	4226	1680	2953	2	11	3
TRFK 12/19	6.05	7.43	6.74	18	15	18	2243	1379	1811	17	18	18
TRFK 31/11	6.55	9.97	8.26	15	2	10	3070	1668	2369	7	12	9
TRFK 301/5	5.99	7.76	6.86	19	12	17	2627	2547	2587	10	1	5
TRFK 301/6	6.45	5.37	5.91	16	19	19	4395	2095	3245	1	5	1
TRFK 430/90	6.76	8.70	7.76	14	7	13	3312	1945	2629	4	8	4
TRFK 371/3	13.49	7.75	10.32	2	13	2	2953	1730	2342	8	10	10
TRFK 6/10	8.07	7.94	9.01	9	11	4	3151	1479	2315	6	16	11
EPK C12	9.31	9.86	9.59	5	3	3	3770	2259	3015	3	2	2
TRFK 12/56	6.22	8.84	7.53	17	6	15	2021	1313	1667	20	19	20
BBK BB35	8.95	9.06	9.00	6	4	5	3307	1548	2428	5	15	7
TRFK 301/4	10.47	6.87	8.67	4	17	8	2570	2248	2409	11	3	8
TRFK ST 306	4.95	5.95	5.45	20	18	20	2429	1948	2198	12	7	12
Mean Location	8.27	8.07					2819	1793				
CV (%)		21.10						12.6				
LSD, P≤0.05		NS	1.98					106	335			
Interactions		2.80						374				



clones were not significantly different from that of the new ones. This demonstrated the ability of the new clones to produce black tea whose quality is comparable to the old popular clones. All clones, except TRFK 306/1 and TRFK 301/6, had TFDGEq levels similar to clone TRFK 6/8, the quality standard clone.<sup>13</sup> The two clones had lower TFDGEq levels. Clone TRFK 306/1 is rich in anthocyanin and was specifically developed for production of unfermented purple tea and has low concentrations of theaflavins. The low value of TFDGEq in 301/6 could be due to low concentrations of theaflavin digallate and theflavin-3'-gallate, which are important components used to calculate TFDGEq most clones could produce high-quality black tea.

Except for TF-3'-g and TF-3,3'-g, the individual theaflavins varied ( $p \leq 0.05$ ) due to the location of production. These results were similar to previous results<sup>38</sup> where the individual theaflavins for Kenyan clonal teas and the Malawi/ South African clonal teas differed. When the clones were ranked for the different theaflavins in the two locations, most clones did not retain their ranks in the two sites. For instance, for simple theaflavins, TRFK 11/4 was the best clone in Kipkebe, but ranked eighteenth in Timbilil. Clone TRFK 12/56 was the best clone for theaflavin-3-gallate in Timbilil, while it was fifteenth in Kipkebe; TRFK 371/3 was the third clone in for theaflavin-3'-gallate in Kipkebe, but eleventh in Timbilil; Clone TRFK 31/11 best clone for theaflavin digallate in Timbilil but nineteenth in Kipkebe. The Spearman's rank coefficients ( $r_s$ ) (Table 2) were 0.40, 0.70, 0.12, 0.43 and 0.26 for simple theaflavin, TF-3-g, TF-3'-g, TF-3,3'-g and TFDGEq respectively. Thus, only Spearman's rank coefficient for TF-3'-g was significant ( $p \leq 0.05$ ). Pearson's correlation coefficients ( $r$ ) for the individual theaflavins and TFDGEq were all insignificant. The interaction between clone and site was also significantly different ( $p \leq 0.05$ ) for all the individual theaflavins and TFDGEq. These results confirm the previous results<sup>38</sup> and demonstrate the levels of individual theaflavins and TFDGEq of clonal tea vary with the location of production. Such variation did not follow a predictable order.

### Clonal and Locational Variation in Yield

The clonal tea yields (Table 6) changed ( $p \leq 0.05$ ) with clone and location of cultivation. The clonal yield differences showed that different clones have different abilities to produce leaf. The results affirmed previous results that tea yield is a polygenically controlled trait,<sup>69</sup> varying from one clone to another. Most of the new clones yielded higher ( $p \leq 0.05$ ) than the old popular clones. This was expected since the current tea breeding and clonal selection strategies aim at developing high yielding tea clones.<sup>69,70</sup> All new clones produced yield higher than or comparable to the clones used for yield check i.e. TRFK 31/8, TRFK 303/577 and TRFK 303/1199. Indeed, the clonal tea yield ranking revealed that clones TRFK 301/6 and EPK C12, both new clones ranked second and third respectively.

The clones produced significantly higher yields ( $p \leq 0.05$ ) in Kipkebe than in Timbilil. This is due to differences in environmental factors at the two sites. In previous studies,<sup>24,25</sup> environmental factors caused variations in clonal yield responses. These factors were identified as water stress, temperature<sup>14</sup> and altitude.<sup>15,56</sup> The factors were different at the two sites with those in Kipkebe favoring faster growth. Both Kipkebe and Timbilil fall within the same tea-growing block in the West of the Great Rift Valley. The wide variations in the yield noted in the two sites, therefore, affirmed the previous studies<sup>15,24</sup> that even within a radius of 10 Km variations in yield and yield-related components occur in tea. Indeed, within Kericho, for every 100M rise in altitude,

there is 1 Kg decline in yield.<sup>56</sup> The Spearman's rank correlation ( $r_s = 0.51$ ) and the Pearson's correlation coefficient ( $r = 0.33$ ) (Table 2) for yield between the two sites were, however. Indeed, there was also significant ( $p \leq 0.05$ ) interactions effects between the clones and location of production. These observations demonstrated that yields of the tea clones varied with the location of production unpredictably.

## CONCLUSION

There were variations in all the plain tea quality parameters including individual theaflavins, and yields with clones. The results demonstrated the need to evaluate tea clones for quality and yields before wide plantation for commercial exploitation. However, the responses in the plain tea quality parameters and yield of the tea clones changed with the location of production. Further, there were significant ( $p \leq 0.05$ ) interactions effects between clone and location of production for all parameters. Moreover, the Spearman's rank coefficients ( $r_s$ ) and Pearson's correlation coefficients ( $r$ ) were low and insignificant. These results demonstrated that changes in the quality parameters and yield of clonal tea with the location were not systematic and were unpredictable. There is, therefore, need to re-evaluate the tea clones, selected for quality or yield in one location, in the new location of intended commercial exploitation to ensure that farmers use cultivars that can produce high quality and yield in their regions.

## ACKNOWLEDGMENT

Authors want to thank the management of Kipkebe Tea Estate and Timbilil Estate for provision of experimental fields and management of the trials. This research was supported with funds from the Inter-University Council of East Africa (VicRes).

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