

Manufacturing of Low Haze Instant Tea Extracts Using Sri Lankan Broken Mixed Fannings (BMF) as Raw Materials

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ABSTRACT

Haze or tea cream in instant tea significantly affects tea-based product manufacturing due to its impact in preparing unclouded tea infusions. The effect of pre-processing temperature of spray drying and cultivar elevation with haze development of instant tea production from Broken Mixed Fannings (BMF) was studied first time by analyzing Total polyphenol (TPC), caffeine content, yield and haze value in instant tea. Significant differences in haze and TPC percentages were observed across elevation categories of upcountry (86.1 NTU & 16.33%), mid country (49.6 NTU & 13.25%), and low country (29.7 NTU & 11.94%) respectively. A strong positive correlation (0.992) was observed between TPC and haze level while a negative correlation showed between Caffeine and haze level (-0.967). However, there were no significant differences for haze, TPC and caffeine contents among tea estates in the same elevation category. The highest yield (27.17%) and lowest haze value (48.75 NTU) of instant tea were reported at the pre-process spray drying temperature of 70 °C when compared with 40, 50, 60, 80 and 90 °C. The method was validated in large-scale production and could be recommended as a proven methodology to reduce haze development and acquire a high yield of instant tea.

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INTRODUCTION

Tea [*Camellia sinensis* (L.) O. Kuntze] is one of the most popular non-alcoholic and aromatic beverages which has unique organoleptic characteristics (taste, aroma, and colour), and health benefits. Many studies have established the therapeutic effect of tea against cardiovascular diseases (Fang *et al.*, 2019), type 2 diabetes (Asbaghi *et al.*, 2020), and cancer (Sur & Panda, 2017). Tea is well known for its antioxidant property (Benzie *et al.*, 2000; Fang *et al.*, 2019).

“Ceylon tea” produced in Sri Lanka is acclaimed as the best tea in the world which has a high demand in international trade (Kottawa-Arachchi *et al.*, 2012) and currently Sri Lanka is the third largest tea exporter (Ceylon Tea Brokers PLC Annual Report 2019/20). Sri Lanka produces black tea in the form of Orthodox and Cut Tear and Curl (CTC), Green tea, Flavored tea, and Instant tea. Among the different tea products, instant tea has emerged as a modest and highly developing sector in the tea-producing countries. The changes of eating habits and lifestyle of the people, the global market for ready-to-drink tea, including instant tea is expected to grow further in the years to come.

Instant tea is generally made from fermented black tea (Kraujalyte *et al.*, 2016), green (Zhang *et al.*, 2020) and pu-erh (Du *et al.*, 2019) teas, however, Broken Mixed Fannings (BMF) is commonly used because of the low price and it is considered as a low-quality product in the industry. Instant tea suffers from an inherent haze problem or tea cream formation which causes the loss of transparency, colour, taste and cup quality (Powell *et al.*, 1993) which reduces consumer preference and affects physical and sensory attributes and biological activities (Youngmok & Stephen, 2012). The complexation of polyphenols by the association of their galloyl groups with limited solubility is said to be the cause for the tea cream formation (Argyle & Bird, 2015). Studies have shown that phenolic acids, tea catechins, and flavanol glycosides are involved in tea creaming and potassium and calcium ions activated the process. (Kim

& Talcott, 2012). Yin *et al.*, (2008) found that the main constituents of black tea cream were thearubigins (TRs), theaflavins (TFs), and caffeine. Moreover, in tea cream formation, polyphenol- caffeine complexation is influenced by several gallate and hydroxyl groups of the polyphenols with peptide groups in proteins and keto groups in lipids oxidized polyphenolics that are formed during the fermentation (Seshadri & Nagalakshmi, 1988; Jöbstl *et al.*, 2005).

During the manufacturing of cold soluble instant tea, as the tea infusion cools down, it becomes turbid and with strong infusion, coloured precipitate in a form of a suspension is formed. Tea creaming results in the blockage of nozzles in the spray drying step of instant tea manufacturing (Sivapalan & Herath, 1986). Thus, tea cream formation in the instant tea significantly affects the tea industry because it impacts producers who need to prepare tea infusions with high solid transparent concentrations and health beneficial effect which is mainly given by the phenolic compounds.

Therefore, in order to produce a good quality instant tea product, the tea cream portion is separated through filtration and centrifugation. However, complete separation is not achieved because tea cream will be gradually formed in filtered clear tea liquor during storage (Murugesu *et al.*, 2017). The addition of tannase, viscozymes enzymes to tea liquor in low pH followed by polymeric micro membrane filtration is also practiced by the industry. Tannase treatments hydrolyzing gallated polyphenolics produced in tea cream in black tea resulted in the loss of astringency and overall quality of the tea extract (Write, 2005). The enzyme treatment process is further limited as consumers do not prefer to drink enzyme or chemical-added tea, since tea beverage is considered as a healthy drink. Frequent changing of microfilters in commercial level production leads to higher cost of production. Therefore, reducing haze using existing methods while fulfilling consumer preferences will be a challenging task in the commercial manufacturing of instant tea.

Hence, it is important to develop a processing technique to control the tea cream formation naturally without losing organoleptic characteristics and bioactive compounds in tea extracts. Limited studies have been carried out to evaluate the effect of controlling the pre-spray drying processing temperatures, on tea cream formation however, details have not been reported. Therefore, the objectives of the present study were to investigate the chemical characteristics and polyphenolic contents of raw materials (BMF) collected from tea estates in different elevation categories in Sri Lanka and the effects of processing temperatures of pre-spray drying brewed tea in producing natural instant tea with low haze levels.

METHODOLOGY

Selection of estate and collection of BMF as raw material

Two representative tea estates from each elevation category of upcountry (elevation more than 600 m mean above sea level-MASL), mid-country (elevation 300-600 m MASL), and low country (0-300 m MASL) of Sri Lanka were selected for the study. When selecting the estates, priority was given to the tea estates which were having higher extents of the cultivar TRI 2025 and following more or less similar agricultural practices with the same manufacturing practice of the Orthodox/Rotorvane type. Estates of Ingestre and Robgill for the upcountry; Deniyaya and Sogama for the Mid country and Halgolla and Kelani for the Low country were selected for sample collection. Composite samples of 5 kg of BMF were collected during the April in 2020 for the tea liquor extraction and chemical analysis. Instant tea used for the analysis was produced in a commercial scale production system using 3000kg BMF blended according to the experimental design presented in Table 1.

Preparation of the tea extract

The method used for producing tea extract was a laboratory-scale equivalent to the commercial-scale procedure followed by

Hayleys Global Beverages (Pvt) Ltd, Dickoya, Sri Lanka. In commercial production to avoid the under-capacity of the machinery utilization, different BMF combinations from each elevation were taken into production and the same protocol was followed in the experiment. Approximately 500 g of nine different combinations of BMF from each elevation were taken as 50:50%, 60:40%, and 40:60% from the respective two estates as per Table 1. The BMF was brewed with 4.5 L of boiled distilled water (tea leaf to water ratio of 1:9 (w/w)) for 15 minutes while maintaining the temperature around 95 °C.

The extract obtained was filtered through a nylon mesh (aperture size of 100 µm) to remove fine insoluble solids in the extract. Filter solution was centrifuged (Z206 A, HERMLE, USA) at 4500 rpm for 3 minutes. The extract was then concentrated to a total soluble solid content of approximately 25 g/100 mL using a rotary evaporator (Witeg vapor, Germany) while maintaining the liquor temperature at approximately 75 °C. The tea extract of each combination was cooled down to 40 °C which is taken as a standard temperature equal to the holding tank where tea concentration was kept before spray drying and the same is used for the haze, TPC and caffeine analysis.

Validation of the experimental findings in a commercial setting

In a commercial setting, the tea liquor was extracted from Broken Mixed Fannings using heated process water and the spent leaves were filtered and decanted. Then the tea liquor was concentrated in a vacuum evaporator. The concentrated tea liquor thus obtained was passed through a heated jacketed tank maintained at 40, 50, 60, 70, 80 and 90°C to expose the tea concentrate to the experimental process temperature. The tea concentrate was then passed to a commercial scale spray dryer and instant tea powder was collected. The yield percentage was calculated as per the given equation below. The percentage yield of instant tea was calculated as follows:

Percentage yield = (net instant tea powder production / total BMF input) x 100%

Table 1: Preparation of combinations from each elevation

| Elevation | Estates | Combinations | | |
|---|----------|--------------|----------|----------|
| | | 1 | 2 | 3 |
| Upcountry (elevation > 600 m MASL) | Ingestre | 50% | 60% | 40% |
| | Robgill | 50% | 40% | 60% |
| | | 4 | 5 | 6 |
| Mid country (elevation 300-600 m MASL) | Deniyaya | 50% | 60% | 40% |
| | Sogama | 50% | 40% | 60% |
| | | 7 | 8 | 9 |
| Low country (elevation 0- 300 m MASL) | Halgolla | 50% | 60% | 40% |
| | Kelani | 50% | 40% | 60% |

Analysis of the haze value, polyphenol content and caffeine content of tea extracts

The above tea extract combinations were exposed to pre-process spray drying temperature of 40, 50, 60, 70, 80, and 90 °C. Haze value of tea extracts and instant tea samples and tea liquor were analyzed. In addition, tea extracts obtained from each combination is presented in Table 1 were analyzed for total polyphenols and caffeine contents.

Analysis of Haze value

The haze value was obtained as a measurement of turbidity in the infusion according to the method followed by Hayleys Global Beverages. Briefly, 0.3 g of sample of the extract was mixed with 50 mL of deionized water and 50 mL of citrate buffer solution (pH 3.7). Then the haze of this solution was measured using a turbidity meter (TURB 555 IR, Weilheim, Germany).

Analysis of Total Polyphenol Content (TPC) in tea extract

The determination of TPC was performed using the Folin Ciocalteu method according to ISO 14502-1 (ISO, 2005).

Analysis of Caffeine content in tea extract

The caffeine content of each combination was analyzed according to a standard procedure used by Hayleys Global Beverages (Pvt.) Limited, Dikoya, as follows: a 0.20 g (known volume of extract evaporated in a water bath and total dry solid measured by the

gravimetric method) equivalent volume of tea extract from each combination was taken to the 250 mL of volumetric flask and top up with distilled water. An aliquot (10 mL) of this solution and 20 mL of chloroform were transferred into a separating funnel. After mixing and allowing to settle into two layers, the chloroform layer was separated. The absorbance of the solution was read at 276 nm with a UV/Visible spectrophotometer (Evolution 201: Thermo Scientific, Waltham, MA) and the caffeine content was calculated using a standard curve prepared with pure caffeine.

Statistical Analysis

All experiments were performed in three replicates for each combination to obtain data on haze value, TPC and caffeine contents. ANOVA followed by mean separation (least-square means difference), was performed using SAS (Statistical Analysis System, Version No 9.4) to evaluate the effect of elevation factor, combination effect, and the temperature on haze level of above tea extract. The association among the yield, TPC, Caffeine, and haze value in tea extracts was assessed by Pearson's correlation analysis. Significant differences were defined at $p < 0.05$.

RESULTS AND DISCUSSION

Variation of haze values, polyphenols and caffeine of tea extracts with elevation categories

In the present study the haze value, TPC and caffeine contents analysis of tea extracted using BMF obtained from three different elevational categories are shown in Figure 1.

Results showed that there is a significant difference ($p>0.05$) in the haze level, TPC and caffeine contents of BMF among elevation categories, but no difference observed in same elevation levels. The lowest haze value (27.54 NTU) in tea extract was recorded in low country BMF while the highest (118.98 NTU) was recorded in the upcountry. Similar trend was observed for TPC, is the lowest value (11.94%) is reported from low country BMF while the highest TPC (16.33%) reported from up country BMF. However, Caffeine content variation showed an opposite trend as the highest caffeine content (3.97mg/L) is reported from low country BMF. Results revealed that higher clarity of instant tea can be obtained from the BMF of the low country, however, with the high caffeine content, there may be higher after tea taste.

Many factors such as region of cultivation and processing parameters influence in the tea

quality (Owuor *et al.*, 2008). Land elevation is one of the key factors determining the polyphenols in tea which is responsible for total antioxidant capacity (Zhang *et al.*, 2018). It has been reported that the higher growing elevation subjects the plant to a lower environmental temperature which in turn slows down the growth of the plant and helps to accumulate more TPC in tea leaves make tea is flavorful and rich with antioxidants (Kamal *et al.*, 2008). Moreover, in an upcountry where having an average of 16 °C mean temperature and an average rainfall of 155 mm/month will not be a favourable condition for the growth of the shoots. The misty weather conditions in higher elevations also reduce the number of sunshine hours which again slow down the shoot growth. Due to the redundancy in the shoot growth, the accumulation of total polyphenol is higher in upcountry and this resulted to produce a high amount of polyphenols in tea leaves.

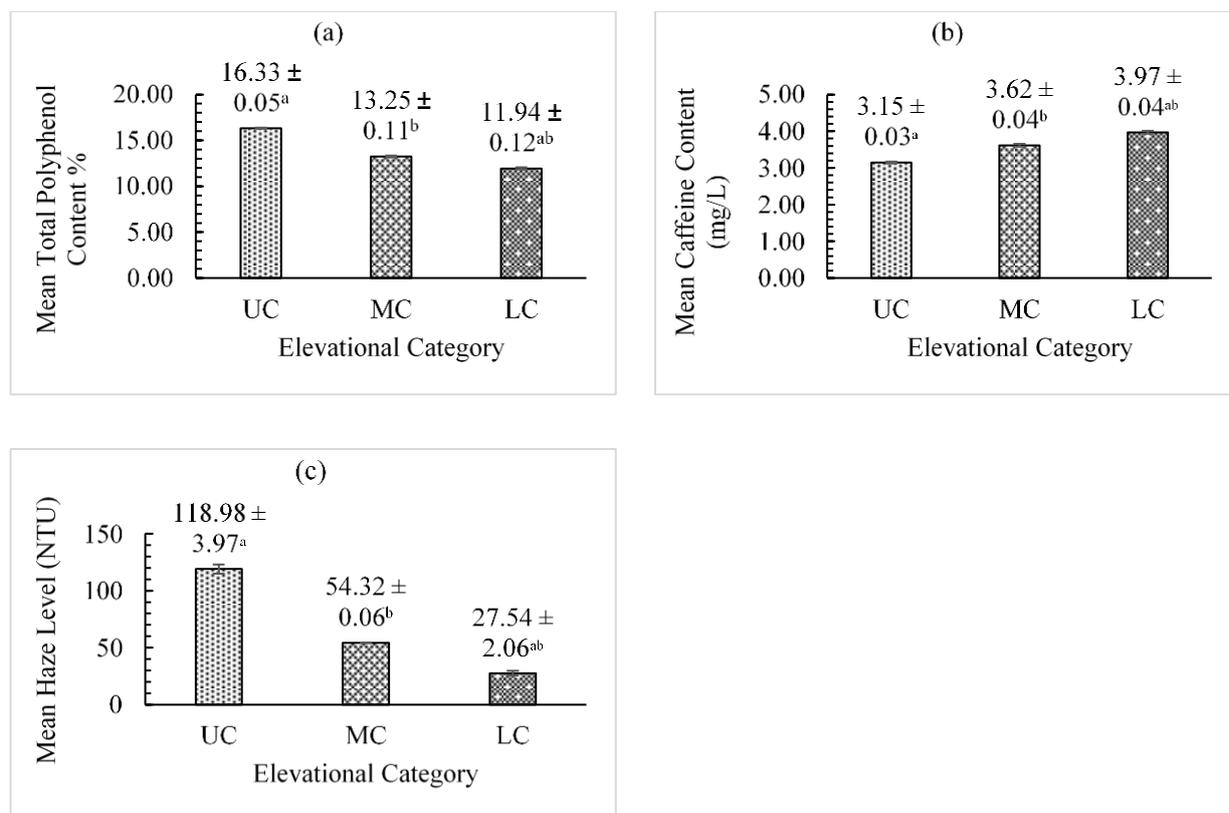


Figure 1: Variation of mean. (a) TPC % (b) caffeine content (c) haze value (mg/L) of tea extract at 40°C obtained from BMF of different elevational categories. Data obtained for combination 1, (50:50 w/w) of each elevational category. (UC- Up country- Mid country, LC- Low country)

Moreover, Catechin is the main polyphenols of the tea and theaflavins is the main oxidation products of catechins (Engelhardt, 2013) which are responsible for the haze level of tea. Smith, (1968) has determined that cream formation occurs in tea if the strength of the infusion exceeds that of a certain threshold, and hence, tea infusions that are high in theaflavins, caffeine and thearubigins have shown higher creaming power. Arachchilage *et al.*, 2015 reported that thearubigins/theaflavin ratio of upcountry instant tea is high compared to the other elevational levels was the reason for the higher haze in the upcountry instant tea infusions. Tea from different regions has different capacities to oxidize catechins due to variations in the levels of polyphenol oxidase. The temperature of processing has an influence on oxidation as the uptake of oxygen increases up to 29 °C, where the enzyme activity reaches a peak (Shitandi *et al.*, 2013) and the development of tea cream (haze) due to the complexing polyphenols with proteins and oxidized lipids formed during fermentation.

The present study further shows that low level of caffeine in upcountry BMF showing high haze values confirmed the Jöbstl *et al.*, (2005) findings that the contribution of caffeine on tea cream formation is happened only in the presence of sufficient substances with galloyl groups i.e., gallated tea polyphenols which further confirmed by the information presented in Figure 2.

Pearson's correlation analysis was performed to determine the relationship between the caffeine content Vs haze value and TPC content Vs haze value to understand the effectiveness of BMF combination, TPC, and caffeine contents to the cream formation of instant tea (Figure 2).

A positive correlation observed between haze value and TPC ($r=0.992$) confirmed the effect of TPC in developing haze value while strong negative correlation ($r=-0.967$) of haze value and caffeine content shows that caffeine is not highly interfere the haze value development in instant tea. Therefore, results can be further elaborated that high TPC concentration in higher elevations may form

more oxidized polyphenols at fermentation and that can be contributed for high haze development at up country tea.

In addition, there was no significant difference ($p<0.05$) reported for TPC and caffeine contents among the combinations used within the same elevation category. Therefore, in a practical situation, combining of BMF (mostly from the similar tea cultivar) from different estates in the same elevation category (agro-climatic region), is not causing any quality variations but could facilitate meeting the capacity requirement of the manufacturing plant.

Caffeine content affects tea creaming by binding with the galloyl groups and tea cream particles which increases their mass and density (Argyle & Bird, 2015). Therefore, even having high caffeine in low elevations has negatively affected the formation of haze in samples. This may be due to the fewer chances of having caffeine-polyphenol complexation compared to polyphenol-polyphenol molecular interactions.

Best processing temperature range to reduce the haze level in tea extracts.

Even though the tea cream could be removed by cooling and centrifugation, precipitation, or filtration in instant tea manufacturing, the haze will be gradually formed in a filtered clear tea liquor while temporarily storing them during the processing (Murugesu, 2017). The situation directly affected the quality and market value of instant tea. Therefore, different tea extracts obtained from different BMF combinations of three elevation categories ($n=27$) were exposed to six different pre-processing temperature levels in commercial scale instant tea production ($n=162$) and observed their haze formation pattern (Figure 3).

There was a significant difference ($p<0.05$) observed in means of haze when the processing temperature at 70 °C and 80 °C compared to 40, 50, 60, and 90 °C temperature levels. The lowest haze level (48.75 NTU) was reported at the pre-spray drying holding tank temperature level of 70 °C, and there was no significant difference

($p < 0.05$) of means values of haze between temperature levels of 70 °C and 80 °C compared to other temperature levels. In addition, a similar trend was observed for all elevations irrespectively to the combination when tea extract was exposed to

temperatures of 70 °C to 80 °C range, which reduced the formation of haze in instant tea.

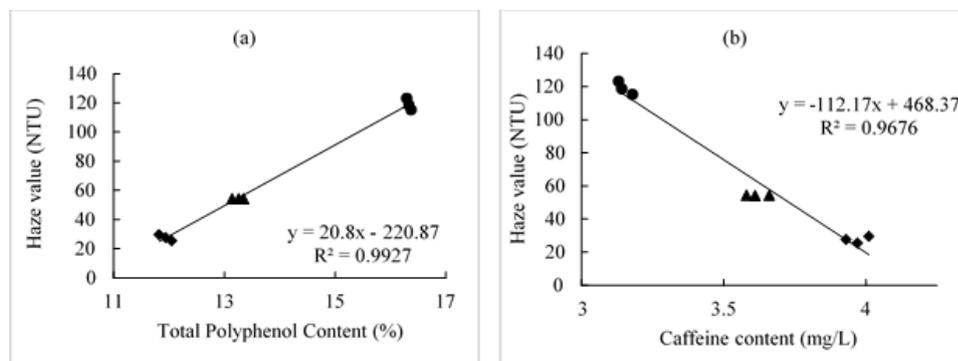


Figure 2: Relationship between (a) TPC contents and haze levels of tea extracts; (b) caffeine content and haze levels of tea extracts (● - Up Country, ▲ - Mid Country, ◆ - Low Country).

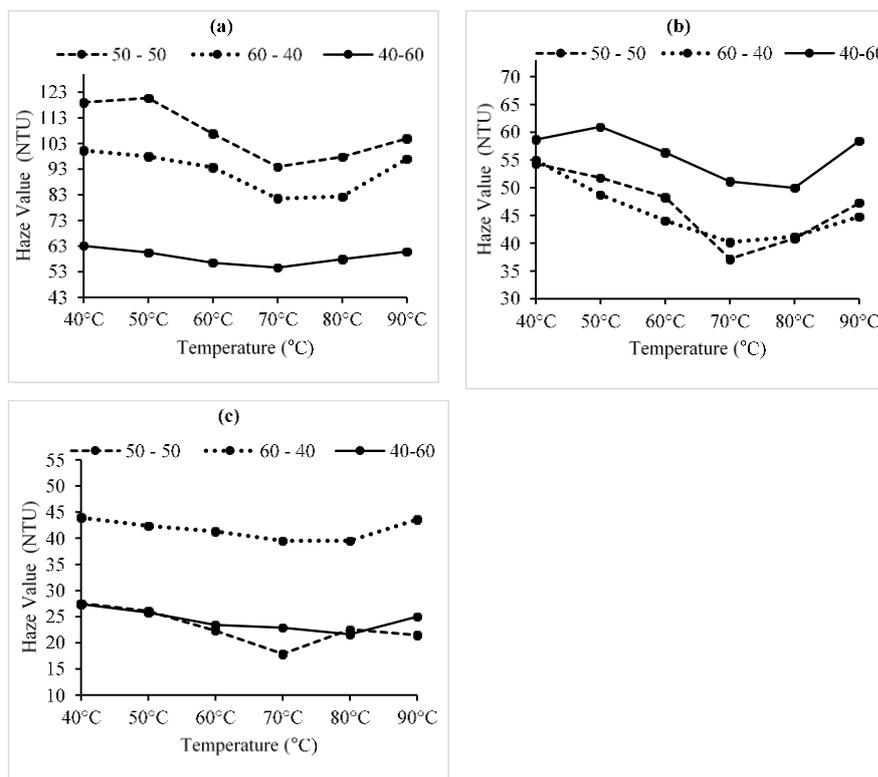


Figure 3: Variation of mean haze value of three combination of BMF obtained (a) Ingestre BMF: Robgil BMF (Up Country) (b) Deniyaya BMF: Sogama BMF (Mid Country) (c) Halgolla BMF: Kelani BMF (Low Country) blending ratios with different processing temperatures

At higher temperatures, the strength of the hydrogen bond may become lower and less energy to formulate protein and oxidized lipid complexes, and this may be the reason to get low haze value at higher temperatures like 70 °C and 80 °C. A similar finding has been reported by Liang, Y. & Xu, Y. (2003) that the tea cream which forms as of haze, has highly temperature-dependent. Therefore, at the processing or at holding tank before spray drying step when the extract is exposed to 70 °C to 80 °C, the bonds between the molecular complex get weaker, and more insoluble will come to the soluble phase. But in 90 °C there was a slight increase in haze can be seen in all three elevations and combinations (Figure 3), may due to the denaturing of protein of the tea cream and those particles may be accumulated at an insoluble portion of the tea extract. Similar reasons also described in the same study (Liang, Y. & Xu, Y., 2003), as there were compositional differences that have been noted at 4 °C of green tea creams which is having more catechin and caffeine whereas more rutin and protein complexes common in tea cream formed at a higher temperature of 25 °C and 40 °C.

The process described in present study in first time of exposing tea extract into 70 °C

just before the spray drying was validated in a large-scale production plant where approximately 3,000 kg of instant tea powder was produced daily for about one month period and similar results of lower haze levels in instant tea during this one month were obtained. Figure 4 showed that percentage yield of the instant tea is also higher when maintain the processing temperature at 70 - 80 °C compared to the other temperature levels and confirmed that this finding is very important in commercial level instant tea manufacturers in worldwide.

Therefore, this information presented herewith can be used by the food companies that wish to develop beverages using instant tea extract, in powder or liquid form, especially to cater to the ready-to-drink (RTD) market to obtain low haze levels. Hence, by the processing of tea liquor at the temperature range of 70 °C to 80 °C, just before the spray drying, the resulting instant tea produces less haze with high yield, and this will help to produce a low haze instant tea-based beverage product ultimately without harming natural health beneficial components as well as the overall quality of the final product.

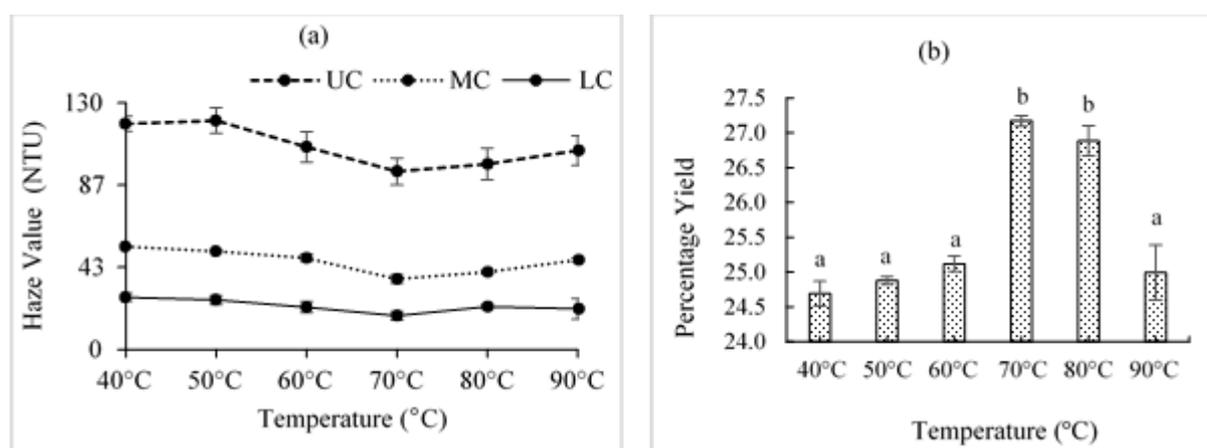


Figure 4: Variation of (a) mean haze value (b) percentage yield with different processing temperatures. ^{a-b} Means with different letters are significantly different ($p < 0.05$)

CONCLUSIONS

The study revealed that there were significant differences in haze levels observed among the three elevation categories up country, mid country, and low country in Sri Lanka. The lowest haze value was recorded from the BMF extract of the low country while the highest was recorded in the up country. The elevation groups had a significant influence on TPC, with upcountry having the highest and low country having the lowest. Similarly, elevation had an effect on the caffeine content of tea; however, the lowest caffeine content was found in the low country, while the highest caffeine content was found in the high country.

A strong positive correlation was observed between the haze value and TPC, while strong negative correlation was observed between the haze and the caffeine level. There were not any significant differences in TPC, caffeine, or haze level among the extracts obtained from BMF combinations of estates in the same elevation category. The haze level was significantly affected by the processing temperature. The temperature that resulted in the least amount of haze, as well as the highest yield of instant tea powder, was 70 °C. The changes carried out in the process by exposing the extract to 70 °C were validated in a large-scale production plant for one month and obtained the same result confirming the possibility of recommending the 70 °C processing temperature at the commercial level manufacturing of instant tea to obtain lower haze in the product.

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