



Journal of Chemical, Biological and Physical Sciences

An International Peer Review E-3 Journal of Sciences

Available online at www.jcbpsc.org

Section B: Biological Sciences

CODEN (USA): JCBPAT

Research Article

An Experiment Comparing the Effect of Tea (*Camellia sinensis*) Types as an Arab-habit on Individual and Pathogenic-bacteria, at High-altitude Area, Taif, KSA, "VISION 2030"

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Received: 05 May 2018; **Revised:** 01 June 2018; **Accepted:** 06 June 2018

Abstract: This paper was carried out for "An experiment comparing the effect of tea (T) (*Camellia sinensis*) types as an Arab-habit (AH) on individual and pathogenic-bacteria (PB) at high-altitude (HA) area, Taif, KSA, "VISION 2030"". Boiling water T extract (BWTE) created benefits on digestive system (DS) as arranged (black, green, red, white and Matcha T); (B, G, R, W and MT). (BT and GT) caused constipation only, (RT, WT and MT) caused (nausea, hotness, gaseous and constipation). Gut microbiota (GM) was improved by T polyphenols (TP), BWTE supported GM as (BT, GT, WT, RT and MT). All pathogenic flora (PF) was eliminated and was reached to Zero percent. (Gt and RT) were the fastest and most powerful BWTE eliminated turbidity, which decreased to 25% / hr. BWTE components had bactericidal effect on *Escherichia coli* (*E. coli*); at first hr, all T types eradicated 25%. At third hr (BT and GT) eliminated 50%, (RT, WT and MT) killed 25%. At fifth hr, (BT and GT) eliminated 75%, RT killed 50% and (WT and MT) killed 25%. At seventh hr 100% was

eliminated by BT, 75% was by (GT and RT), 50% were by (WT and MT). That concluded the residents and visitors at HA area "Taif" were using (BT and GT) as essential drinking daily regularly as AH often because of that had qualities and benefits to DS. That recommend drinking (BT and GT) regularly daily, because for its preservations and positive effects on DS as reduction DS disturbances at HA area.

Key words: *High-altitude area, Digestive system, Boiling water tea extract, Gut microbiota, Escherichia coli.*

Abbreviation List:

Abbreviation	Meaning	Abbreviation	Meaning	Abbreviation	Meaning
AH	Arab habit	GM	Gut microbiota	PB	Pathogenic bacteria
AR	Arab Region	GT	Green tea	PF	Pathogenic flora
BT	Black tea	HA	High altitude	RT	Red tea
BWTE	Boiling water tea extract	hr	Hour	T	Tea
C. s	Camellia sinensis	MT	Matcha tea	TP	Tea polyphenols
DS	Digestive system	NA	Normal altitude	WT	White tea
E. coli	Escherichia coli	NF	Normal flora	yr	year

1. INTRODUCTION

T; "C. s" is drinking usually as leaves BWTE it is the most generally beverage as AH in AR¹. The common T content is caffeine 3%, in dry weight (30-90) mg / 250 ml depend on kind, product², and processing method³. The caffeine content is in 1 gm of BT (22-28) mg and 1 gm of GT (1-20) mg⁴. The most item in TP is the greatest leaves abundant compounds (30-40%)⁵. T has minor quantities of (theobromine, theophylline, stimulants, and xanthines) are similar to caffeine⁶; it also has Fluoride and Aluminium occasionally⁷. BT and GT contain nutritional mineral, Manganese 0.5 mg / mug for 26% of Daily Value⁸. T has varied polyphenols, counting (flavonoids, epigallocatechin gallate and other atechins)⁹, GT and BT may protect from cancer¹⁰, obesity or Alzheimer's disease¹¹.

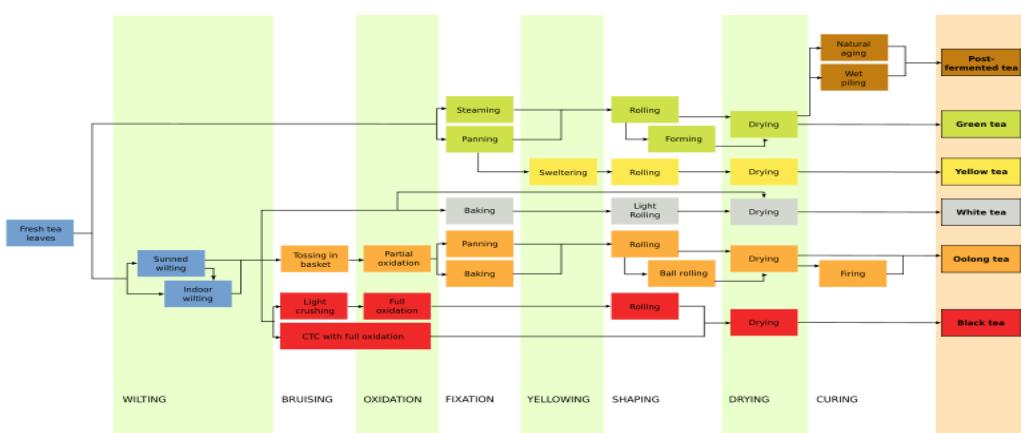


Figure 1: Common processing methods of tea Leaves¹²

The effect of TP on DS that promoted favorable GM growth as *Lactic acid* bacteria, inhibited PB proliferation ¹³⁻¹⁴, and maintained DS health by progress *Bifido-bacterium* growth ¹⁴. TP had anti-inflammatory effects to adjust macronutrient digestion represent a dose-limiting un-favorable effect ¹⁵. TP acted as a prebiotic provide favorable growth conditions for beneficial bacteria. TP supported GM composition and structure to maintain DS haemeostasis ¹⁶. TP protected GM, which ligated with host's health conditions, as in case of (obesity, allergy, and other diseases) ¹⁷. TP helped with changes GM composition, which helped GM in running suppressed the body weight and blood lipid increased with high-fat diet ¹⁸.

TP affected on PB as *E. coli*, that was common dangerous cause of food-borne outbreak, that due to food contaminations; the most dangerous on public health was *E. coli O157:H7* ¹⁹. TP had anti-microbial effects admitted for food preservation from PB contamination as *E. coli* ²⁰. TP had anti-bacterial effects against Gram-negative bacteria as *E. coli* ²¹⁻²². The anti-microbial effects were in BWTE contents (bacteriocins, nisin and natamycin), that were applied for foodstuffs preservative from PB ²³. The anti-microbial effects on PB were controlled meat and dairy products ²⁴. BWTE eradicated *E. coli* due to present of (nisin, natamycin, and their combinations), that were natural anti-microbial, had highly reducing effects on PB growth ²⁵.

The aim of experiment was studying in HA area "Taif" for the basic T components and that was work of BWTE for different T types which available in KSA, that for comparison. That was happened through the experiment of humans and were followed-up DS behavior and its impact on DS-GM and PB for 7 days. That was confirmed the comparison of BWTE for T types by experiment on PB as *E. coli* to show the extent of T bactericidal effect during (1-7) hr. That supported the role of different BWTE types on DS and its different flora, that demonstrated the importance of different T types contents, where T types were still usually using as AH in KSA which considered the first common AR.

2. MATERIALS AND METHODS

-Search area: "Taif" area had been selected as HA area in KSA and is frequented receiving many Arab Visitors throughout the year. The research was conducted because the T drinking was an old AH in KSA, which are still present daily and regularly. There might be were a differences in HA results from NA area due to differences in climates and situation which individuals were exposed ²⁶.

-Samples collection: T samples were brought and collected, that available in "Taif" area included (BT, GT, RT, WT and MT), that traditional consumed drinking using as AH in KSA ²⁷.

-Sample preparation: BWTE was made by weight from T (1.6-2.0) g in (250-300) ml distal water and was boiled for (3-5) minutes, was stayed for 10 minutes. Then small pores sterile gauze was used to obtain BWTE, after that was waited for 5 minutes to get all TBWE contents. BWTE was placed in sterile screw capped bottles with data label and were kept in refrigerator ²⁵.

-Experiment internally on humans as *in-vivo*:

- The volunteers consent, purpose notice and methods explanation were obtained; they were healthy individuals, did not have any disease and did not take any medicines or herbs, their ages of (30-50) yr. They are able to make BWTE at home. T bags were weighted for volunteer's uses and were brought to them for the experiment duration ¹⁴.
- Experiment Method: Faecal tasters remained placid before the experiment as Zero and were sent to the Bacterial Laboratory. BWTE was started by one cup without any additives three times / day for a week. DS behavior was recorded during the experiment and T types were compared. Faecal tasters remained

placid after the experiment at eight day and were sent to Bacterial Laboratory, the DS flora were compared at Zero and eight day¹⁴.

-Experiment externally on PB "E. coli" as *in-vitro*:

- Identified *E. coli* as PB to DS was brought from research center¹⁴.
- Experiment Method: Zero growth were quantified for *E. coli* suspension, and then mixture was made from equal amount of *E. coli* suspension and each BWTE in a separate sterile Wizerman tubes with data. The turbidity degree at (1, 3, 5 and 7) hr were recoded. As well 5 micron of mixture was taken using disposable plastic loop to culture at (1, 3, 5, and 7) hr on Molar Hinton agar with data, then were kept for (24-48) hr at degree of 37°C. *E. coli* growth quantities were recorded, and were compared at Zero, all hrs and between BWTE types¹⁴.

-Data analysis: All data and results were collected and were analyzed by "Simple Excel Program" to get the mean and to be establish in tables and graphs²⁸.

3. RESULTS AND DISCUSSION

*Table and graph 1: Prevalence of *DS behavior during experiment internally in-vivo*

Signs	*BT	*GT	*RT	*WT	*MT
<i>Nausea</i>	00%	00%	100%	100%	100%
<i>Hotness</i>	00%	00%	100%	100%	100%
<i>Gaseous</i>	00%	00%	100%	100%	100%
<i>*R. digestion</i>	00%	00%	00%	00%	00%
<i>Constipation</i>	100%	100%	100%	100%	100%
<i>*R. defecation</i>	00%	00%	00%	00%	00%

*DS: Digestive system, *BT: Black tea, *GT: Green tea, *RT: Red tea, *WT: White tea, *MT: Matcha tea, *R: Regular

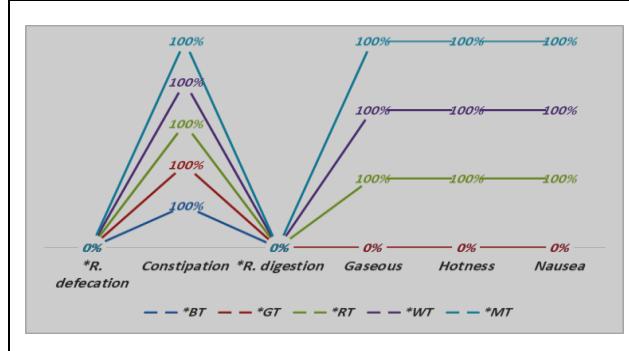


Table and graph 1 revealed prevalence of DS behavior during experiment internally *in-vivo*, BWTE created benefits behaviour also caused some effects on DS, which appeared as signs. The benefits on DS were arranged T types as (BT, GT, RT, WT and MT) respectively. The effects of (BT and GT) which caused constipation only, while (RT, WT and MT) caused (nausea, hotness, gaseous and constipation). Therefore, from the volunteers under the experiment used (BT and GT) as AH, that were considered the main drinking of them for their beneficial. While other T types did not show benefits for DS, but showed some effects and tired of DS¹³⁻¹⁸. That revealed the equivalent dose of T bags had a multiplier effect on DS, as it might be enough to more than one cup might be was enough to (3-4) T cups. That was indicated it was in high concentration of TP so was produced some side effects on DS. Therefore, the signs were obvious because an individual did not organize to the quantity of T related to the big quality of T contents. According to AH, both (BT and GT) were considered the most useful drinking for the DS and therefore it is still an AH in KSA at HA area as "Taif"¹⁻⁹.

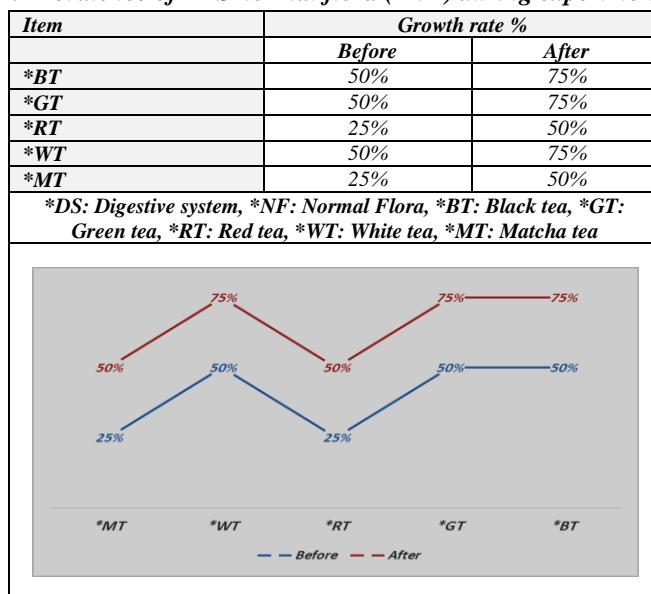
Table and graph 2: Prevalence of *DS normal flora (*NF) during experiment internally in-vivo

Table and graph 2 revealed prevalence of DS-NF during experiment internally *in-vivo*, NF called GM was investigated through faecal analysis, and was vary according to the person. Most importantly, during week of experiment was showed as well GM was improved and increased by an indication of TP effect, which improving GM, thus improving function and digestion. As well, GM supplied the body with vitamins. The results indicated the importance of TP to modify performance in DS through support and help GM. BWTE were arranged according to the support effect on GM as (BT, GT, WT, RT and MT) respectively¹³⁻¹⁸.

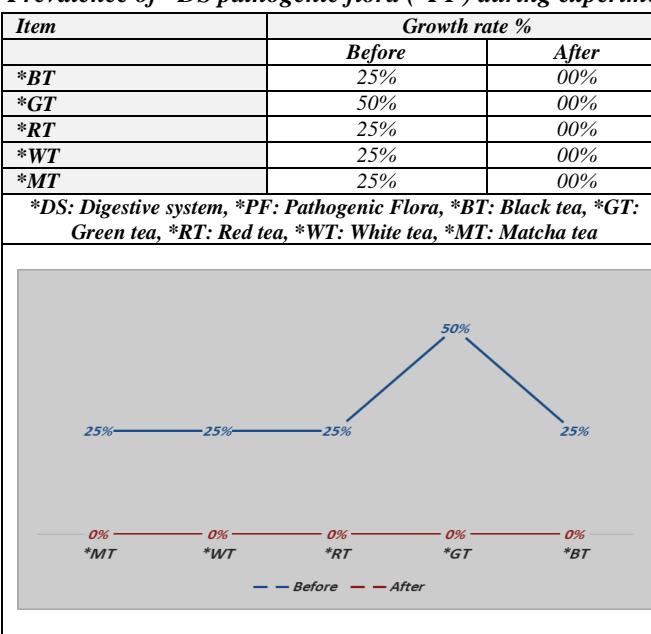
Table and graph 3: Prevalence of *DS pathogenic flora (*PF) during experiment internally in-vivo

Table and graph 3 revealed prevalence of DS-PF during experiment internally *in-vivo*, through the experiment found the DS-PF ranging from (25-50%) and this created failure of DS functions, killed GM and were consequent to affect digestion and absorption. Therefore, it might be lead to lack of vitamins production in addition disease symptoms. After the experiment, it was found that all PF was eliminated and the ratio was reached to Zero percent. This indicated the effect of TP on PF, which had antibacterial effects. This was a proof of results comparison before and after the experiment, it indicated TP had antibacterial effects against PF, as well nourished and helped GM function, also cleaned DS from PF¹⁹⁻²⁵. The use of (BT and GT) in the AR as AH helped GM presence and PF reduced, which showed it as DS treatment and that it was more in (BT and GT). As well as at "Taif" as HA area the visitors confirmed the use of T still in the AR as AH because of its GM benefits and reduced PB¹⁻⁹.

Table and graph 4: Prevalence of turbidity during experiment externally *in-vitro*

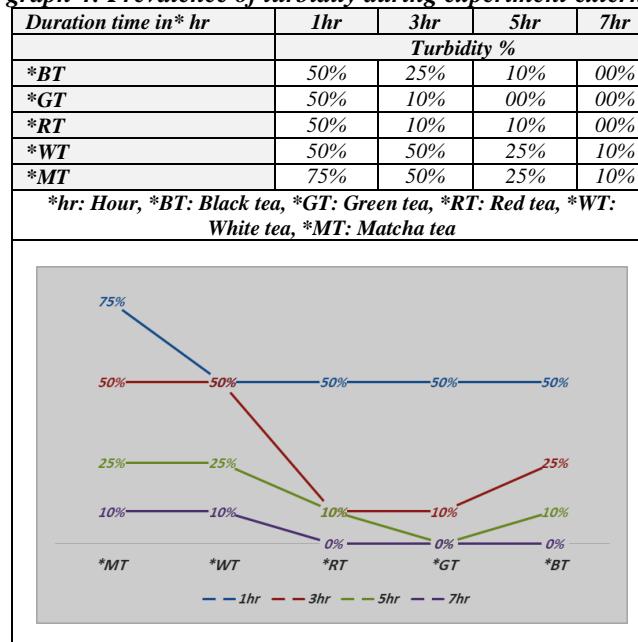


Table and graph 4 revealed prevalence of turbidity during experiment externally *in-vitro*, turbidity was estimated in mixture of BWTE and *E. coli*, which was found turbidity ranged between (50-75%). (Gt and RT) were the fastest and most powerful to eliminate turbidity, which decreased to 25% / hr. (GT and RT) were more in turbidity elimination than others T in the first hr. TP was reducing the turbidity by the basic materials externally. That indicated TP ability to eliminate bacteria, included *E. coli* externally and internally. Turbidity was erased which minded elimination of *E. coli* in varying proportions according to the type of BWTE. The strongest were (GT and RT) followed by (BT, WT and MT). This was another evidence of BWTE ability suppressed intestinal infections by PB. The elimination of turbidity was equaled to kill PB as completely so lead to clear solution was resulted from elimination of PB as *E. coli*. These benefits were still in the mind of AR persons, that were supported by drinking T in AR as AH specially in KSA. Also was still in mind the support of persons DS by drinking T regularly daily¹⁹⁻²⁵.

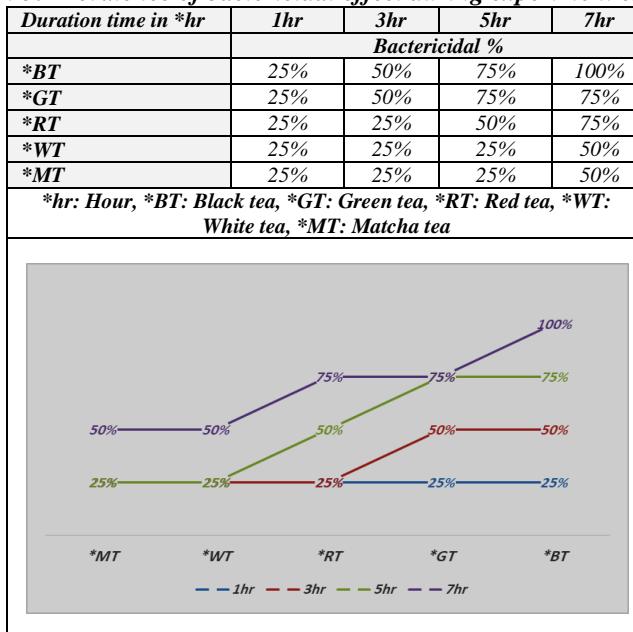
Table and graph 5: Prevalence of bactericidal effect during experiment externally *in-vitro*

Table and graph 5 revealed prevalence of bactericidal effect during experiment externally *in-vitro*, BWTE components had bactericidal outcome for *E. coli*; as well during the first hr, all T types killed 25%. While the difference was in the third hr as (BT and GT) eliminated 50%, but (RT, WT and MT) killed only 25%. At the fifth hr, (BT and GT) eliminated 75%; while RT killed about 50% and (WT and MT) killed 25%. At seventh hr, 100% was eliminated by BT, 75% was by (GT and RT), 50% was by (WT and MT) ¹⁹⁻²⁵. It was explained that BT was a strongest T types of *E. coli* eradication, and then were (GT, RT, WT and MT). It was found that the presence of TP and T contents showed the ability to kill PB as *E. coli*, which was used in the experiment, as it was the most important example of DS disease events. TP and contents was able to kill PB internally and externally to DS, which helped DS to reduce or to eliminate the bacterial infection, which was led to clean and restore DS functions ¹⁹⁻²⁵.

Through the DS behavior followed up, BWTE found to be as AH helping DS for digestion, absorption and defecation without causing any DS side effect. That indicated the importance using of (BT and GT) in AR more than others T as its usually continuous drinking and still as an AH ¹³⁻¹⁸. It was found that BWTE effect was help preserved GM and increased in DS, which saved it to DS and was styed DS in its functions so continued and increased uses of T in AR as a regular daily drink as AH ¹³⁻¹⁸. The DS-PF had eliminated by BWTE and thus helped to preserve DS functions and reduced infections, which associated side effects on DS. T drinking still using in AR for DS treatment diseases as it reduced or eliminated PB so this AH for resulted in province for state of healthy DS ¹⁹⁻²⁵. When tested BWTE for PB, it was found gradually eliminated either by turbidity or *E. coli* growth. It had showed that, BWTE had benefits for person as it had healthy benefits to maintain DS healthy, eliminated PB multiply and keep on GM ¹⁹⁻²⁵. Drinking regularly BWTE regularly daily as AH that improved DS by TP and contents, which were able to regulate DS functions. The effects of T contents stayed in the body for at least (4-6) hr, so drink T regularly such as herbal remedies ¹⁻¹¹. BWTE of (BT and GT) was found to be more commonly used in AR as AH. They had shown to be more active on DS than others T types that used in experiment internally and externally ¹⁻¹¹. The experiment was carried out in HA area, where as the known side effect of HA area was DS disturbance and

disorders. T drinking was used to stop side effect of HA area on DS and reactivated DS functions regularity. Therefore, individuals living at HA area still drinking (BT and GT) daily regularly about (3- 4) cups / day as AH had proved to be important and healthy for the HA individuals ¹⁻¹¹.

CONCLUSIONS

Through the results, that found the residents and visitors of AR at HA area "Taif" were using (BT and GT) as essential drink daily regularly as an AH often because of qualities and benefits to DS. The other types (RT, WT and MT), although they were available in KSA but they were less used than (BT and GT) as they led to some side effects on DS so they used others T (RT, WT and MT) in a very small amount compared to (BT and GT) as AH in AR.

RECOMMENDATIONS

The drinking (BT and GT) daily regularly had preservations and positive effects on DS as reduction DS disturbances at HA area as in AR as AH.

ACKNOWLEDGMENTS

Many thanks and grateful were sent to all persons who participated in this work also in the practical experiments and producing this research paper for its success.

FUTURE WORK

Use the five T types for experiment on individuals from NA area to observe the effects and comparison with HA area, also noting the daily habits of NA individuals for T drinking.

CONFLICT OF INTEREST

There was not conflict of interest related to this paper or to its practical work. That was cleared in this paper from the practical experiments, which were done on different T samples in AR; "KSA", which was carried especially in HA area "Taif".

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Online publication Date: 06.06.2018