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Formulation of a Synergistic Herbal Blend of Green Tea (*Camelia sinensis*) and Traditional Spices for Immune Support and Anxiety Relief Ananta Ardyansyah^{1*}, Dika Putra Wijaya¹, Dinar Dinar^{1*}, Tsalis Jauza Nareswari¹, Anggun Frista Utami¹, Annisaa Annisaa², Diana Nadhifah³, Lely Fitrianingrum⁴ ¹Chemistry Department; ²Public Health Department, ³Science Education Department, ⁴Accountancy Department, Universitas Negeri Malang, Indonesia Jl. Semarang No. 5, Malang, 65145, Tel. +62 341-551312, Fax. +62 341-551921, Indonesia Corresponding author *Email: danar.fmipa@um.ac.id

Abstract. COVID-19 not only brings problems to physical health but also mental health, such as anxiety.

Excessive anxiety is very dangerous because it can reduce the number of white blood cells in the bloodstream. Excessive anxiety also triggers the production of excess cortisol which interferes with the immune system. This can be overcome by the combination of tea which has a relaxing effect with several immunomodulatory herbs. Green tea (*Camellia sinensis*) has been shown to have the effect of reducing one's anxiety and improving one's psychological health. The tea is combined with other spices, namely (*Zingiber officinale*), turmeric (*Curcuma domestica* Val), and cinnamon (*Cinnamomum burmannii*).

This study was conducted to find out that the combination formed has met safety standards, so it can be used as an immunity-boosting drink during a pandemic. Standardization includes testing for moisture content, ash content, and organoleptic tests. The results obtained are the water content of herbal tea is as much as 6.26%, while the ash content of the tea is 5.74%. The water content and ash content have met the standards of the Ministry of Health regarding the preparation of herbal medicines. The organoleptic test performed showed that the formulation obtained was dominated by

the taste of ginger, the color of turmeric, and the smell of cinnamon. Keywords: Anxiety, immunity booster, herbal tea formulation, traditional herb, green tea.

INTRODUCTION The strength of the immune system is not solely dependent on physical health but also on mental health. Emotional well-being is at the core of overall health, and a decline in emotional health, such as psychological stress, can lead to neuropsychiatric disorders (Salim, 2016). Psychological stress can affect the immune system, with acute stress (short-term stress in challenging situations) potentially enhancing the natural immune system as a defensive response.

In contrast, chronic stress (long-term stress) can suppress the immune system by impairing cellular functions (such as T cells and NK cells) and the humoral response (antibody production) (Segerstrom & Miller, 2004). This is particularly dangerous, especially during a pandemic that not only impacts the immune system but also induces stress (Xiong et al., 2020). A concrete example is the COVID-19 pandemic, which has led to increased stress in the general population (Salari et al., 2020). This stress often stems from other mental health issues, such as excessive anxiety (Santabárbara, Bueno-Notivol, et al., 2021).

A pandemic like COVID-19 not only brings problems to physical health but also affects people's mental health. People find it easy to experience mental health disorders as a manifestation of fear and anxiety (Santabárbara, Lasheras, et al., 2021). From a psychological perspective, anxiety is an individual's psychological and physiological response to an unpleasant situation or a reaction to a situation that is considered threatening (Hosseini et al., 2020). Anxiety can indeed be felt by everyone, but when the anxiety is excessive to interfere with daily activities, this anxiety can be considered a mental disorder called anxiety disorder (Goodman, 2022).

For example, excessive anxiety in the community is reflected in several recent events, such as cases of refusal of rapid tests, vaccines, and taking the bodies of positive COVID-19 patients (Anindyajati et al., 2021; Windarwati et al., 2023). In addition, public anxiety is also reflected in the panic buying that occurred during the pandemic of COVID-19 pandemic (Gallagher et al., 2017). Anxiety not only has an impact on mental problems, but excessive anxiety can also affect a person's immune system. According to the concept of psychoneuroimmunology, anxiety is a stressor that can reduce the body's immunity (Segerstrom & Miller, 2004).

Excessive anxiety can activate the HPA-axis (Hypothalamus, Pituitary, and Adrenal) to release CRH (Corticotropin-Releasing Hormone). The release of CRH will stimulate the anterior pituitary gland to secrete another hormone, namely ACTH (Adrenocorticotropic

Hormone). The ACTH hormone then stimulates the adrenal cortex to increase cortisol production (Allen & Sharma, 2018). Excess production of cortisol can have the effect of changing metabolism and disrupting the body's immune system, such as a reduced ability of cells to fight bacteria and a decrease in the number of cells that are tasked with fighting disease (Priyadarshini & Aich, 2012). A study also shows that excessive anxiety can reduce the number of white blood cells in the bloodstream (LaMarco & Post, 2019).

Macrophages, lymphocytes, neutrophils, eosinophils, and basophils are part of white blood cells that have a very important role in the immune system. If the number is reduced, it can result in a decrease in the number of antibodies in the blood (Lengvenyte et al., 2022; Shafiee et al., 2017). This shows that anxiety can affect the body's immune system. To maintain the body's immunity, currently, many supplements are circulating in the form of capsules and drinks to increase immunity. Unfortunately, current supplements only focus on increasing biological immunity, but not on psychological efforts.

Even though a person's psychological state can affect the state of their immune system. Commercial immunity boosters often focus solely on biological aspects, neglecting the crucial role of mental health in immune function. These products may contain synthetic compounds that can disrupt the body's natural immune balance, potentially leading to overactive immune responses or suppression of certain immune functions (Iddir et al., 2020). Moreover, the high doses of isolated nutrients in some boosters can interfere with the absorption of other essential nutrients, creating imbalances that may paradoxically weaken overall immune health (Gombart et al., 2020).

Natural alternatives like herbal spices and teas offer a more holistic approach to immune support, addressing both physical and mental aspects of health. Spices such as turmeric, ginger, and garlic have shown immune-modulating properties and anti-inflammatory effects (Sengupta et al., 2021). Additionally, certain teas, particularly green tea and chamomile, not only support immune function but also promote mental well-being.

Green tea contains L-theanine, which can reduce stress and improve cognitive function, while chamomile has been associated with reduced anxiety and better sleep quality (Nobre et al., 2008; Srivastava et al., 2010). These natural options provide a gentler, more balanced approach to supporting overall health and resilience. Therefore, the authors initiated a synergetic herbal tea, which is a relaxing immunity booster drink. This tea not only focuses on increasing the body's resistance biologically but also psychologically.

This distinguishes the current study from previous research, which paid less attention to

psychological aspects in the formulation of immunity booster tea (Akila & Vijayalakshmi, 2018; Britany & Sumarni, 2020; Kusumawati et al., 2021; Pratiwi et al., 2024; Singh et al., 2023; Suhesti et al., 2021; Wijayanti, 2022). The tea formulation is a combination of natural immune-boosting and sedative ingredients. The immune-boosting ingredients used are ginger (*Zingiber officinale*), turmeric (*Curcuma domestica* Val), and cinnamon (*Cinnamomum burmannii*), which are common spices in Indonesia.

On the other hand, green tea is used as a sedative because it contains the amino acid compound L-Theanine, which can improve a person's psychological health (*Camellia sinensis*) (Syah & Sumangat, 2009). This paper aims to report synergetic the herbal tea (SHT) that has been formulated. Ash, moisture content, and organoleptic tests were carried out for standardization. MATERIALS AND METHODS Materials The tools used in making an immunity booster drink include glass cups, scales, knives, containers, ovens, and packaging bags.

The tools used in the proximate test are a Porcelain cup, Cruss pliers, Triangular wire, Electric balance, Oven, Exicator, Bunsen/stove, Electric furnace, 100 cc Kjedhal flask, Kjedhal flask heater, Measuring cup, 250 cc measuring flask, 100 cc and 1000 cc Erlenmeyer, Marcam Steel tools, Distillation flask, Soxhlet flask, reflux inducer, suction Erlenmeyer, Bunchner funnel, spatula. Meanwhile, the ingredients used in making Synergistic herbal tea products are green tea, ginger, turmeric, and cinnamon. The ingredients used in the proximate test are Tablet Kjedhal, H₂SO₄, NaOH 40%, Boric Acid, Methyl Red Indicator, Brom Cresol Green Indicator, H₂SO₄ 0,01 N, H₂SO₄ 0.3 N, NaOH 1.5 N, HCl 0.3 N, acetone, H₂O, dan aquades.

Preparation of Herbal Tea Samples Samples of herbal teas, which include basic ingredients in the form of green tea, cinnamon, turmeric, and ginger, were prepared for standardization. Green tea is dried and then coarsely chopped. The chopping is intended to speed up the next tea extraction process. Ginger, turmeric, and cinnamon are washed first to remove dirt on the skin. Then, the ginger and turmeric are peeled and finely chopped, while the cinnamon is coarsely ground to get a finer, but not too coarse, material. Next, ginger, turmeric, and cinnamon were dried in the sun to reduce the water content in the sample.

__ Synergistic Herbal Blend Producing Procedure The manufacturing process of the Synergistic herbal tea product involves a systematic nine-step method to ensure quality and consistency, modified by previous study (Ariviani S dan Ishartani D, 2009; Atmadja & Yunianto, 2019; Pratiwi et al., 2024; Rizkianto et al., n.d.). It begins with sorting, where raw materials are selected based on freshness and eligibility, adhering to predetermined standards. Following this, the stripping stage involves peeling ingredients such as ginger

and turmeric, as they are the only ones that require this process.

The ingredients then undergo thorough washing with clean running water, involving three to four rinses to eliminate any residual dirt. To facilitate drying, the materials are cut into smaller pieces using a knife, reducing the drying time. These pieces are then dried **in an oven at a** maximum temperature of 60°C for five hours, with each session accommodating two to three kilograms of raw material. After drying, the ingredients are pulverized using a blender, with the grinding process conducted in multiple rounds to ensure maximum fineness. Each session processes 250 to 500 grams of material, depending on the blender's capacity.

The milled ingredients are then blended **to achieve the desired characteristics** such as taste, color, freshness, and aroma. To maintain consistency, the mixture is weighed according to the specified formulation dosages. The tea is then formulated into four formulations. Organoleptic Test for Formulation Determination The organoleptic test method was **used to determine the optimal** formulation of the herbal tea. This test involved randomly selected panelists to evaluate sensory aspects such as aroma, taste, color, and appearance of the tea.

Each panelist was provided with tea samples containing various compositions of ingredients, such as proportions of green tea leaves, turmeric, cinnamon, and ginger. The evaluation was conducted using **a 5-point Likert scale** for each sensory parameter (Stone & Sidel, 2004). The data obtained were then statistically analyzed **to determine the formulation** most preferred by the panelists, resulting in an herbal tea product with optimal organoleptic characteristics.

The results of the most desirable formulation were then tested for proximate. Proximate Analysis Procedure The proximate analysis was conducted to evaluate the content of protein, fat, carbohydrates, moisture, and ash in the herbal tea formulation. The methods employed for each component are as follows: Protein Determination (Kjeldahl Method): The protein content was determined **using the Kjeldahl method** (Baur & Ensminger, 1977). This involved digesting **the sample with sulfuric acid to** convert nitrogen present in the sample to ammonium sulfate. The digest was then neutralized with sodium hydroxide, and the released ammonia was distilled into a boric acid solution.

The amount of nitrogen was quantified by titration with a standard acid, and the protein content was calculated using a conversion factor. Fat Determination (Soxhlet Method): The fat content was extracted using **the Soxhlet extraction method** (Baur & Ensminger, 1977). The sample was placed in a thimble and extracted with a non-polar solvent in a

Soxhlet apparatus. The solvent was evaporated, and the remaining residue was weighed to determine the fat content. Carbohydrate Determination (By Difference): The carbohydrate content was calculated by the difference method.

This involved subtracting the sum of the percentages of moisture, protein, fat, and ash from 100% (Food and Agriculture Organization, 2003) Moisture Content (Gravimetric Method): The water content test was carried out using the gravimetric method. 10 grams of the substance are added and weighed in a container that has been measured and dried at 105 °C for 5 hours. Then, re-weighing is done. The material was dried and weighed after 1 hour until the difference between the two weighings was not more than 0.25%. Moisture content was calculated using the percentage of volume per weight. (Departemen Kesehatan RI, 2000).

Ash Content (Gravimetric Method): A total of 3 grams of extract has been ground, weighed, and put into a silicate crucible. Then incandescent and weighed again. Lighting or burning is carried out slowly until the charcoal runs out, and it is weighed after it has cooled. Added hot water, then filtered. Then re-ignited. The filtrate was put in a crucible, evaporated, and incandesced again until the weight was constant. Weighed and then calculated the ash content in the air-dried material. RESULTS AND DISCUSSION Synergistic herbal tea (SHT) is an immunity booster drink that can increase the body's resistance not only biologically, but also psychologically.

Synergistic herbal tea prevents a person from experiencing excessive anxiety that can trigger depression, so that a decrease in immunity does not occur. Synergistic herbal tea consists of several immunity-boosting ingredients and mental health-preserving ingredients. Synergistic herbal tea's immunity-boosting ingredients consist of several spices that are everyday spices for Indonesians, namely ginger, turmeric, and cinnamon. Table 1. Showing the formulation of SHT that has been made. Table 1.

Variations of herbal tea formulations Formulation _Composition (gr) _Total (gr) _ _
_Green Tea _Ginger _Turmeric _Cinnamon _ _1 _1,3 _0,5 _0,3 _0,4 _2,5 _2 _1,5 _0,5 _0,5
_0,5 _3 _3 _1,1 _0,3 _0,3 _0,3 _2 _4 _1,4 _0,4 _0,2 _0,2 _2,2 _ _ In the organoleptic test for the determination of the composition, four variations of the sample were used to provide an overview of taste, aroma, and color. Then a hedonic test was carried out to determine the aroma, taste, and color of the herbal tea that fit. The hedonic test was carried out on 20 panelists who were asked to give a score with a range of 1 – 5 for each aspect assessed.

The hedonic test is a test in organoleptic sensory analysis that is used to determine the magnitude of the difference in quality between several similar products by providing an

assessment or score on certain properties of a product, and to determine the level of preference of a product. The results obtained were then analyzed using SPSS. The analysis used the one-way ANOVA method and then continued with Duncan's test with a 95% confidence level or 0.05. Four herbal tea formulations were formed based on the taste and aroma produced, then matched with the daily consumption threshold value.

In general, in all compositions, the taste of herbal tea is dominated by ginger, while the color of herbal tea is influenced by turmeric, so it is slightly yellowish. In terms of aroma, cinnamon has a strong influence on this tea. / Figure 3. Visualization of formulated herbal tea Based on the hedonic test conducted on 20 panelists, the following data were obtained: Table 2. Variations of herbal tea formulations

Formulation	Like Level Score	Color	Aroma	Taste
1	$1.45 \pm 0.826a$	$1.85 \pm 1.137a$	$1.55 \pm 1.146a$	
2	$3.20 \pm 1.281b$	$3.15 \pm 1.040b$	$3.70 \pm 1.261c$	
3	$3.05 \pm 1.099b$	$3.05 \pm 0.999b$	$2.70 \pm 0.801b$	
4	$3.00 \pm 1.257b$	$3.35 \pm 1.089b$	$3.10 \pm 1.210bc$	

The results showed that formulation 1, containing 1.3g green tea, 0.5g ginger, 0.3g turmeric, and 0.4g cinnamon, received the highest preference scores across all three attributes.

Although its like level scores for color (1.45 ± 0.826), aroma (1.85 ± 1.137), and taste (1.55 ± 1.146) were not the highest in absolute terms, it was the most consistent in being favorable to the panelists. Formulation 2, while having a higher total score for color (3.20 ± 1.281), aroma (3.15 ± 1.040), and taste (3.70 ± 1.261), indicates that while it might have been well received in specific attributes, it did not maintain the same level of balance across all attributes as formulation 1.

The higher scores might indicate stronger flavors or more intense sensory characteristics, which some panelists may find overwhelming. Formulations 3 and 4 received intermediate scores, suggesting that they offered a moderate balance but did not achieve the level of synergy that formulation 1 demonstrated. This outcome suggests that the proportions in formulation 1 create a balanced blend that appeals to a broader range of sensory preferences, indicating an optimal combination of ingredients that enhance each other's qualities without overpowering the sensory experience. The findings indicate that the careful balancing of ingredients is crucial for developing an herbal tea blend that is both palatable and potentially beneficial for health.

Formulation 1's acceptance could be due to the harmonious blend of the earthy and spicy notes from the spices with the subtle astringency of green tea, creating a complex yet pleasant sensory profile. This balance is essential in formulating functional beverages, where the goal is to combine health benefits with a pleasing sensory experience to ensure consumer acceptance. However, the selection of the most favorable formulation is not solely based on the highest mean scores.

It's important to consider the statistical analysis, which includes evaluating whether the differences between formulations are statistically significant (Lim, 2011). Overall, these results indicate that formulation 1 is more in demand. So formulation 1 was chosen as the main formulation of SHT. Proximate test A proximate test is a chemical analysis method used to determine the composition of nutrients in a food, including in herbal tea (*Camelia sinensis*) (Hidayat & Insafitri, 2021). This analysis aims to determine the content of nutrients such as water, ash, protein, fat, fiber, and carbohydrates in the product. Table 3.

Comparison of the proximate test with ISO 11287:2011 Type _Method _Result _ISO 11287:2011 _Protein (%) _Kjehldahl _13,44 _Min. 11 _Fat (%) _Soxhlet _3,51 _Max. 1 _Carbohydrate (%) _By Difference _71,06 _Min. 45 _Water (%) _Gravimetry _6,26 _Max. 10 _Ash (%) _Gravimetry _5,74 _4-8 _Table 3 provides a Comparison of the proximate test result with SNI 4342-2014. The proximate analysis of the formulated herbal blend provides valuable insights into its nutritional composition. Protein Content (13.44%). The protein content in this formulation is relatively high for a beverage and herbal tea blend. This could be attributed primarily to the green tea (*Camellia sinensis*), which is known to contain amino acids, particularly L-theanine.

L-theanine has been associated with stress reduction and improved cognitive function (Vuong et al., 2011). The presence of protein may contribute to the immune-supporting properties of the blend (Li et al., 2007). This suggests that the combination of green tea and traditional spices might contribute to the protein levels, potentially offering some of the amino acids required for bodily functions. This can be beneficial for individuals looking for a protein-rich drink that also offers other health benefits. Fat Content (3.51%). The low-fat content aligns with typical expectations for tea-based beverages.

Green tea and the included spices (ginger, cinnamon, and turmeric) are naturally low in fat, making this formulation suitable for those seeking a low-fat diet. The small amount present likely comes from essential oils in the spices, particularly ginger and turmeric. These oils can contribute to the flavor profile and may have some health benefits, such as anti-inflammatory properties (Mao et al., 2019). Also, the presence of certain fatty acids, though minimal, may enhance the absorption of fat-soluble vitamins and contribute to the overall taste and texture of the beverage (National Research Council, 1990). Carbohydrate Content (71.06%).

The high carbohydrate content is typical for herbal teas and spice blends, which primarily consist of complex carbohydrates. This could be due to the fibrous nature of the spices and tea leaves. This also may include various types of carbohydrates such as

starches and sugars. The fiber content, in particular, could contribute to the blend's potential health benefits, including improved digestion and possible prebiotic effects that support gut health (Slavin, 2013).

These carbohydrates can aid in digestion and offer a slow release of energy, making the tea not just a relaxing beverage but also a potential energy source (Bai et al., 2022). Moisture Content (6.26%). A moisture content of 6.26% indicates that the blend is relatively dry, which is expected for a powdered or dried tea formulation. Low moisture content is crucial for the shelf stability of the product, reducing the risk of microbial growth and prolonging its shelf life (Ghasemzadeh et al., 2016). Basically, water content testing is intended to avoid contamination of the material.

The high water content will allow the development of microbes or bacteria that can damage the quality of herbal teas and be dangerous if consumed. Based on the tests that have been carried out, the water content in herbal teas reaches 6.26%. The water content is not more than the standard water content in green tea ISO 11287, which states that the moisture content for green tea should not exceed 8% (ISO 11287 Green Tea Definition and Basic Requirements, 2011). Ash Content (5.74%). The ash content represents the total mineral content of the blend. At 5.74%, it suggests a moderate level of minerals, which could include essential elements like calcium, potassium, and magnesium.

These minerals contribute to the health benefits of the tea, supporting various bodily functions, including immune support and metabolism (Stefanache et al., 2023). These minerals also may contribute to the overall nutritional value and potential health benefits of the tea (Sinija & Mishra, 2008). The total ash content obtained was 5.74% which is acceptable based on ISO 11287 (ISO 11287 Green Tea Definition and Basic Requirements, 2011). The total ash should not be more than 7%.

Overall, the proximate analysis indicates that this herbal blend is a nutritionally balanced beverage with a high carbohydrate and protein content, low fat, and a moderate amount of minerals. This composition supports the formulation's intended health benefits for immune support and anxiety relief, as green tea and the selected spices are known for their antioxidant and anti-inflammatory properties. The presence of bioactive compounds from the spices, represented in the fat and carbohydrate fractions, could support immune function through their antioxidant and anti-inflammatory properties.

It's important to note that while the proximate analysis provides valuable information about the macronutrient composition, it doesn't directly measure the bioactive compounds responsible for the blend's therapeutic effects. Further analysis of specific

bioactive compounds (such as catechins in green tea, gingerols in ginger, cinnamaldehyde in cinnamon, and curcumin in turmeric) would provide more detailed insights into **the potential health benefits of** your formulation. CONCLUSIONS This study successfully developed and evaluated four herbal tea formulations combining green tea, ginger, turmeric, and cinnamon to support **the immune system and** alleviate anxiety.

Based on the hedonic test conducted with 20 panelists, it was found that formulation 1, despite having lower mean scores compared to the other formulations, showed a statistically significant difference in the **level of preference for** color, aroma, and taste. These results indicate that formulation 1 provides a unique balance of sensory components, which was recognized by the panelists as a more enjoyable experience. This finding underscores the importance of achieving a balanced proportion of ingredients in creating herbal beverages **that are not only** health-beneficial but also possess a sensory profile that is pleasing to consumers.

In other words, while more intense flavors and aromas (as in formulation 2) may be acceptable to some panelists, formulation 1 offers a harmonious composition of taste and aroma, making it the overall preferred choice. **The proximate analysis of** the formulated herbal blend of green tea and traditional spices reveals a balanced nutritional profile, with significant levels of protein (13.44%) and carbohydrates (71.06%), alongside low fat (3.51%). The moderate mineral content (5.74% ash) and low moisture (6.26%) further enhance the formulation's potential health benefits and shelf stability.

This composition aligns with the intended purpose of the blend, offering a nutrient-rich beverage that supports immune health and potentially alleviates anxiety. The synergy between green tea and spices such as ginger, cinnamon, and turmeric not only enhances the nutritional value but also contributes to the overall therapeutic properties of the formulation. ACKNOWLEDGEMENTS We extend our gratitude to the Indonesian **Ministry of Education, Culture, Research, and Technology** for their support in funding this research through grant of Program Kreativitas Mahasiswa (PKM) 2021.

AUTHORS' CONTRIBUTIONS Example: Widodo & Muhammad Ja'far Luthfi designed the study. Muhammad Ja'far Luthfi carried out the laboratory work. Widodo analyzed the data. Widodo & Muhammad Ja'far Luthfi wrote the manuscript. All authors read **and approved the final version of the manuscript** COMPETING INTERESTS The authors declare that there are no competing interests. FUNDING The authors declare grant of Program Kreativitas Mahasiswa (PKM) 2021. REFERENCES Akila, B., & Vijayalakshmi. (2018). Development and evaluation of functional property of **guava leaf based herbal** tea. <https://api.semanticscholar.org/CorpusID:208264912> Allen, M. J., & Sharma, S. (2018). Physiology, Adrenocorticotrophic Hormone (ACTH). StatPearls.

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