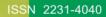
## green tea by LPPM STIKES NU TUBAN

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# Synthesis and characterization of green tea paste nanoparticles based on wet milling

Yuly Peristiowati, Zauhani Kusnul<sup>1</sup>

Department of Nursing, Surya Mitra Husada Health Institute, <sup>1</sup>Department of Nursing, Pamenang Health Institute, Kediri, East Java, Indonesia

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ABSTRACT

This study aims to analyze the synthesis of green tea herbs in the nanoform. Green tea is divided into three grades, namely Grade A, B, and C. Making pasta samples is done by wet milling technique. The tea paste sample was characterized by particle size analyzer and scanning electron microscope methods. Grade A green tea has a grain size of 77,014  $\pm$  50,759 nm. Grade B green tea has a particle size of 12,987  $\pm$  7,674 nm. Grade C green tea has a particle size of 4409  $\pm$  5379 nm. It was concluded that the difference in grade of green tea determines the structure and size of the particles formed. Thus, the wet milling technique can be an alternative in making green tea nanoparticles for industrial scale.

Key words: Green tea, morphology, nanoparticle, wet milling

#### INTRODUCTION

The World Health Organization states that 80% of the world's population had used traditional medicine for hundreds of years. The discovery of synthetic drugs caused the population to move from herbal medicines to synthetic drugs. Although herbal medicine is still seen as nonquantitative, nonscientific, primitive and is used by those who are unable to access real medicine, since 2000, the use of traditional medicines has begun to increase again.<sup>[1-4]</sup>

The function of herbs is based on the work of their active compounds in synergy to form pharmacological actions. Most herbs are not soluble due to low bioavailability and improved cleansing systems. This makes that herbal

Address for correspondence:					
Dr. Yuly Peristiowati, Jl. Manila, No. 37, Sumberece, Tosaren, Kec. Pesantren, Kota Kediri, Jawa Timur 64133, Indonesia. E-mail: yulystikes@gmail.com					
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medicine must be given in repetitive and high doses so that it becomes less potent in therapy.<sup>[5]</sup> The efficacy and absorption of herbal medicines can be increased through nanoformulation and through incorporation with nanocarriers. Nanoherbal form is used to overcome the weaknesses of herbal medicines, namely low solubility, low permeability through the intestine, quickly disappear by oral route, and the first stage of metabolism before reaching blood circulation.<sup>[6]</sup>

High-energy wet milling has proven to be a strong top-down approach to the production of nanoparticles. In this technique, material particles are suspended in aqueous solution and repeatedly pressed by colliding beads that are downsized to the nanometer scale, provided the grinding process is continued for a sufficient period. High-energy wet milling is used in many fields such as minerals, ceramics, pigments, microorganisms, and pharmaceuticals.<sup>[7,8]</sup> Previous studies have proven the benefits of wet milling on the formation of quercetin nanocrystals.<sup>[9]</sup>

Tea is the beverage most often consumed after water. Tea is consumed in the form of oolong, green, black, or

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white.<sup>[10,11]</sup>Green tea is a nonfermented form that is consumed between 3 and 10 cups per day. The benefits of green tea include anti-inflammatory, antioxidant,<sup>[12]</sup> anticancer,<sup>[13]</sup> antiobesity,<sup>[14]</sup> inhibiting metabolic syndrome,<sup>[15]</sup> and antitoxin.<sup>[16]</sup> To the best of our knowledge, until now, no study has made green tea in the form of nanoherbs. Therefore, this study aims to analyze the synthesis of green tea herbs in the nanoform.

#### MATERIALS AND METHODS

#### Classification of green tea

Before making pasta, tea leaves are classified into three groups. Grade A is a tea leaf from the first three strands. The characteristics of grade A leaves are small, green to blackish-green, fragrant, not musty, no foreign matter. This class comes from fine picks, medium (young shoots) young leaves, and young bird leaves. Grade A is a high quality tea leaf. Grade B is tea leaves in the fourth and fifth strands; the leaves are less curled, wide, yellowish green to blackish green, less fragrant but not musty. This type of quality comes from rough passages (old leaves) to the fifth leaf and young bird leaves. Grade C is a leaf powder widened, blackish green color, less fragrant aroma, but not musty. This type of quality comes from fine-picking, rough-picking destroyed by processing, young bird leaves and medium bird leaves.

#### Nanoparticle paste

After the tea leaf sorting process, the next step is to make nanoparticle paste using wet milling. Comparison of tea and aquades 1: 8 leaf samples consisting of Grade A samples (1: 8) 15 g of leaves: 120 ml of distilled water; Grade B samples (1: 8) 100 g of leaves: 800 ml of distilled water; and Grade C samples (1: 8) 30 g leaves: 240 ml of distilled water. The milling time used is 6.5 min; run time is 15 min, off time is 1 min.

#### Characterization

Paste sample characterization was done by particle size analyzer (PSA) and scanning electron microscope (SEM). The paste sample was filtered using filter paper (20 ml), the filter results formed filtrate and residue (tea leaf fiber), and the particle size was measured using PSA. For characterization with scanning electron microscopy analysis, the residue sample (tea leaf fiber) was dried 60°C for 30 min, then smoothed with mortar, and sieved 80 mesh. The sieve results are measured using SEM.

#### RESULTS

Figure 1 shows the wet milling results for green tea. Wet milling results show a brown paste shape and have a soft texture.

Particle size analysis can be seen in Table 1. By PSA, Grade A green tea has a particle size of 133.6 ± 32.6 nm; Grade B green



Figure 1: The green tea paste from wet milling process

tea has a particle size of  $118.4 \pm 30.4$  nm; Grade C green tea has a particle size of  $198.5 \pm 48.7$  nm. By SEM, Grade A green tea has a grain size of  $77.014 \pm 50.759$  nm; Grade B green tea has a particle size of  $12.987 \pm 7.674$  nm; Grade C green tea has a particle size of  $4.409 \pm 5.379$  nm.

Figure 2 shows the morphology of green teananoparticles. For Grade A, fiber is found without agglomeration. In Grade B, no fiber was found and was dominated by singlet particles. For Grade C, there is no fiber, but there are singlet particles which have agglomerated to form globular.

#### DISCUSSION

To the best of our knowledge, until now, no study has applied the wet milling technique in making herbal nanoparticles. Wet milling is intended to produce smaller particle sizes than dry milling.<sup>[17]</sup> In this study, the results of wet milling are in the form of soft paste. Previous studies have shown that wet milling is a method that can maintain ascorbic acid content, total phenolic, total flavonoid, and free radical activity.[18] Furthermore, measurements were made with a PSA obtained for Grade B green tea, having the smallest average particle size, followed by Grade A and Grade C. This proves that the characteristics of the raw material determine the size of the particles formed. This finding was confirmed by SEM that Grades B and C had comparable grain sizes. There are differences in the particle morphology of the three grades. For Grade A, fiber is found without agglomeration. In Grade B, no fiber was found and was dominated by singlet particles. For Grade C, there is no fiber, but there are singlet particles which have agglomerated to form globular. Previous studies have stated that ball milling for more than 120 min will trigger fiber breakdown and increase fine particles. These fine particles will form globular.<sup>[19]</sup> This study has proven that the formation of globular structures is caused by differences in the raw materials of tea.

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	Table	1:	The	size	of	nanoparticle green t	tea
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Method	Grade A	Grade B	Grade C
PSA	133.6±32.6 nm	118.4±30.4 nm	198.5±48.7 nm
SEM	77.014±50.759 nm	12.987±7.674 nm	4.409±5.379 nm

SEM: Scanning electron microscope, PSA: Particle size analyzer



Figure 2: The morphology analysis of green tea nanoparticles. For Grade A, fiber is found without agglomeration. In Grade B, no fiber was found and was dominated by singlet particles. For Grade C, there is no fiber, but there are singlet particles which have agglomerated to form globular. (SEM, ×250)

#### CONCLUSION

It was concluded that the difference in grade of green tea determines the structure and size of the particles formed. Thus, the wet milling technique can be an alternative in making green tea nanoparticles for industrial scale.

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Conflicts of interest

There are no conflicts of interest.

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