A THESIS FOR THE DEGREE OF MASTER OF SCIENCE

# Effect of roasting condition and ventilation on reduction of benzo(a)pyrene generation in sesame oil

볶음 조건과 배기가 참기름의 벤조피렌 저감화에 미치는 영향

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# ABSTRACT

Sesame oil is a preferable seasoning having lots of usage in Korea. However, highly carcinogenic benzo(a)pyrene (BaP) was often detected exceeding the regulation level for sesame oil. Therefore, there have been many tries attempted to reduce BaP in sesame oil. BaP increases through manufacturing process especially during roasting or pressing at high temperature by incomplete combustion of hydrocarbons. To reduce BaP production in the manufacture of sesame oil, improved equipment and optimized conditions are necessary. New equipment for production of sesame oil in this study consists of automatic washing machine, roaster, filter, cyclone, and oil press, and the process conditions to reduce BaP were explored. Water content of sesame seeds, roasting temperature, ventilation speed, and dust removal were the main investigated factors. Raw sesame seeds had 5% of water content but increased to 25% after washing. Water content was controlled to 30, 25, 15, and 10% by repeated washing and drying. As water content of sesame seeds increased, BaP content in oil also increased by having high chance of generating incomplete combustion during roasting. Meanwhile, roasting without washing resulted in slightly high concentration of BaP in oil, indicating that washing had an effect of removing dust which can burn and increase BaP content. In case of roasting, roasting temperature and ventilation

was important factors related to BaP generation. Roasting temperature is classified as heater temperature (220 and 250°C) and completing seed temperature (150, 180, and 210°C) which varies roasting time. Roasting at heater temperature 250℃ spent less than 40 min until seed temperature reaches 210°C, but roasting at 220℃ spent more than 1 hr to reach 150 and 180℃ and generated more BaP. It indicated that total roasting time was more important than temperature for BaP generation. Also, ventilation during roasting was another effective factor for reduction of BaP. The smoke was eliminated by diverse ventilation speed (0, 15, 30, 45, and 60 Hz). Higher ventilating speed tended to reduce more BaP generation, however, too high speed (60 Hz) of ventilation increased BaP making air circulate faster around sesame seeds causing cooling effect. Correspondingly, sesame seeds were heated longer, which led the part near the heater burnt more. Finally, the dust removal process also affected BaP concentration. Foreign materials which went through the roasting process would cause high BaP content in oil and need to be removed. The removing process was separated by size of the material. A filter was used for bigger material than sesame seed, and a cyclone was used for fine dusts to be removed. Between these two, cyclone was more effective to reduce BaP in oil, but operating together made synergetic effect

decreasing more BaP. In conclusion, low water content, short roasting time, adequate ventilating speed, and removing dust with a filter and a cyclone made 35, 59, 46, and 38% of BaP reduction in sesame oil, respectively. Throughout these process, BaP content was reduced by 67% in sesame oil. From this study, BaP reduced sesame oil can be manufactured by using the remodeled equipment and applying optimal condition of process, and it could be used for studies constructing production process for other types of pressed oil.

*Keywords*: benzo(a)pyrene, sesame oil, sesame seed, water content, roasting temperature, ventilation, cyclone

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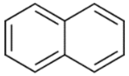
# I. INTRODUCTION

Among edible oils, national shipment of sesame oil is second-highest after soybean oil in Korea (KOSIS, 2009). Sesame oil is usually used as a flavor oil having nutty flavor that Koreans prefer. Because of the unique flavor of sesame oil, manufacturing process is important to differ the flavor. For this reason, small-scale production of sesame oil that has individual flavor is preferable than consistent process of large-scale manufacturing which brings constant flavor. However, the processing method of small-scale production is individually different, so it’s hard to manage their safety of the product. Inevitably, a carcinogen, benzo(a)pyrene, was detected in sesame oil exceeding the regulation level, 2 ppb.

BaP is one of the poly aromatic hydrocarbons (PAHs) which are composed of multiple aromatic rings (Figure 1). The EPA has classified seven PAH compounds as probable carcinogens: benz(a)anthracene, benzo(a)pyrene, benzo(b)fluoran-then, benzo(k)fluoranthene, chrysene, dibenz(a,h)anthracene, and indeno-(1,2,3-cd)pyrene. Especially, BaP is classified as group 1 carcinogen by IARC which means sufficient evidence in humans or sufficient evidence in animals and strong mechanistic data in humans. The mechanism of BaP is performed by enzymatic metabolism which metabolize BaP to a mutagen, bezo(a)pyrene diol epoxide. This molecule covalently binds with guanine nucleobases in DNA. X-ray crystallographic and nuclear magnetic resonance structure studies show that the binding distorts the DNA structure (Volk DE et al., 2003). This interrupts copying process of DNA and induces mutations which occur cancer. This mechanism is similar to that of aflatoxin which binds to the guanine (Eaton, D. L. et al., 1994). For this toxicity of BaP, exposure limit is set up to 2 ppb in U.S., EU, and Korea.

PAHs are neutral, nonpolar molecules; they are found in fossil fuels and in tar deposits, and are produced when insufficient oxygen or other factors result in incomplete combustion of organic matter. PAHs can also be found at high levels in cooked foods, for example, in meat cooked at high temperatures over open flame, and in smoked fish (Lijinsky, W. 1991). They are also found in various edible oils which is manufactured with high temperature (Shuguang, L., et al., 1994; Qu, Y.H., et al., 1992; Chiang, T.A., et al., 1997; Shields, P.G., et al., 1995; Roy, G. M. et al., 1995). Production process of sesame oil includes heating with high temperature, and unintended BaP could be generated. Because the processing method of small-scale production is different from each other, they occasionally produce sesame oil of high BaP content. For example, they roast excessively to get dark-colored sesame oil because of mistaken idea that dark-colored oil would have much nutty flavor. Also, they usually don't have ventilating system or do not use them, and have no sanitary standard to clean the equipment. Existing residues of burnt materials or oil sediment in the equipment may cause additional generation of BaP in oil product. However, just setting the regulation level cannot ensure the safety and have chance to be detected continuously. Indeed, 47 of 623 edible oil was recalled by exceeding the exposure limit of BaP in 2007. In this study, therefore, newly remodeled equipment and optimized process conditions were suggested for production of BaP reduced sesame oil. Most of the earlier studies were dealt with relation between roasting temperature and BaP content (Park, C. H., et al., 2011; Kim, H.Y., et al., 2008; Seo, I. W., et al., 2009), but in this study other factors were also evaluated as causative material. In addition to roasting temperature, water content after washing and drying, ventilation speed during roasting, and dust removing effects are also considered as subject.

Most PAHs are fluorescent, emitting characteristic wavelengths of light when they are excited. BaP is a relatively intense fluorescent compound. However, the absorption and luminescence profiles of the BaP and their isomers overlap, and therefore would require isolation of the individual compounds prior to analysis. Analytical methods for the determination of BaP are based on chromatographic techniques, both GC and HPLC (Grimer et al., 1983). Because of the low molar absorption coefficient of molecules, direct UV detection without any analyte enrichment lacks the sensitivity to determine the relatively low amount of BaP. A sensitive HPLC method for the determination of BaP could be performed with fluorescence detection (Chen, S. H., et al., 1991; Gundel, J. and Angerer, J., 2000; Ramesh, A., 2001; Strickland, P. T., et al., 1994).



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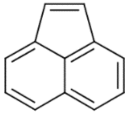

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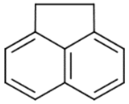

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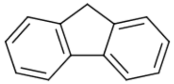

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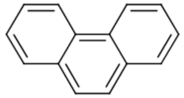

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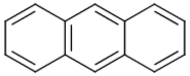

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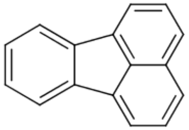

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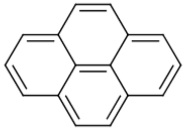

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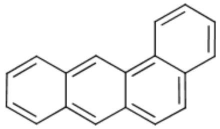

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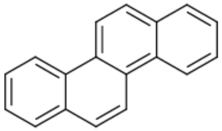

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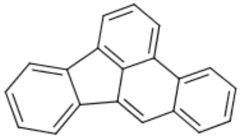
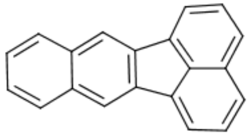


11. Benzo(b)fluoranthene



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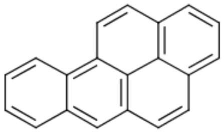

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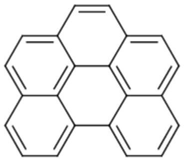
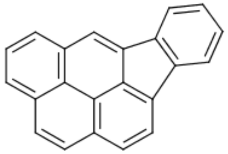
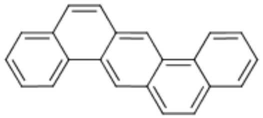


14. Benzo(g,h,i)perylene



15. Indeno(1,2,3,c,d)pyrene



16. Dibenz(a,h)anthracene

Figure 1. Molecule structure of PAHs and BaP

# II. MATERIALS AND METHODS

## 2.1. Chemicals and reagents

Benzo(a)pyrene was purchased from WAKO (Osaka, Japan) to be used as standard for HPLC analysis. Acetonitrile (ACN), methanol, and *n*-hexane were purchased from J. T. Baker (Philipsburg, NJ, USA), and all reagents were HPLC grade. Dichloromethane (DCM), N,N-dimethylformamide (N,N-DMF), ethanol, and anhydrous sodium sulfate ($Na_2SO_4$) were purchased from Sigma Aldrich (St. Louis, MO, USA). Potassium hydroxide (KOH) for saponification was purchased from Showa (Tokyo, Japan). Sep-Pak florisil cartridge for purification was purchased from Waters (Milpord, MA, USA). The polytetrafluoroethylene (PTFE) membrane filter (0.45 μm) were obtained from Merck Millipore (Darmstadt, Germany). All other chemicals were of analytical reagent grade.

## 2.2. Sesame oil production machinery

Machinery for production of BaP reduced sesame oil was designed in Figure 3. It consists of automatic washing machine, roaster, dust remover and oil press. Washing machine automatically supplies water, rotates bidirectional (clockwise and counter-clockwise), drains water and spin-dries, which allows to obtain constant water content of seeds. Roaster has ventilating system with air-inlet to help air circulation and a blower which is connected by zinc-coated spiral duct. Dust remover is composed of two types of filter; one is a mesh filter which filters foreign material bigger than sesame seed, and the other is a cyclone with a vacuuming machine to sort fine dust in seeds. All operation was controlled by control panel. Comparing the process between conventional and remodeled method is shown at Figure 4.

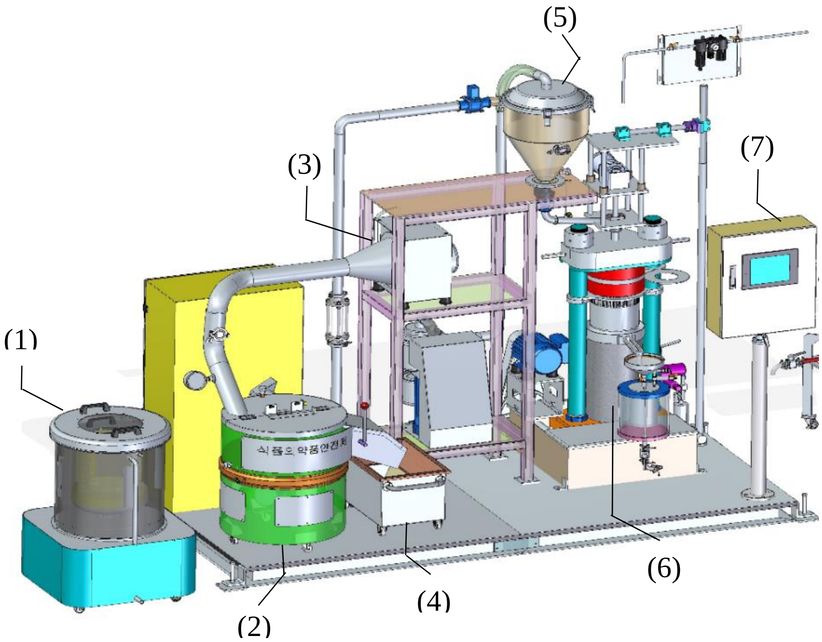



Figure 2. Outside view of sesame oil production machinery. (1) Washing machine, (2) roaster, (3) ventilation system, (4) filter, (5) cyclone, (6) oil press, and (7) control panel.

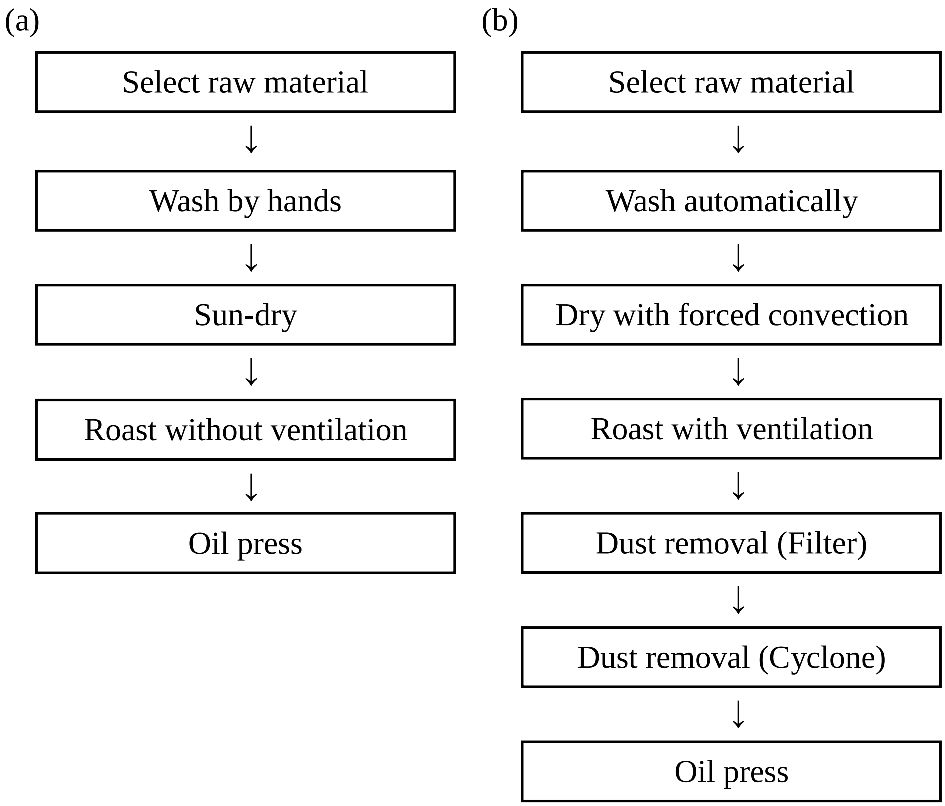



Figure 3. Manufacturing process of sesame oil using (a) conventional machinery and (b) remodeled machinery.

## 2.3. Spiking sesame seeds with BaP

To spike 10 μg/kg of BaP on sesame seeds, standard solution was diluted to 100 μg/kg by acetonitrile, and 600 mL of solution was sprayed to 6 kg of sesame seed. Then, mixed thoroughly with wood spatula.

## 2.4. Preparation of sesame oil

Six kg of sesame seeds was weighed and 10 μg/kg of BaP was spiked on them. Spiked seeds were washed with tap water for 2 min by changing rotating direction every 5 s. Washed seeds were dehydrated by spinning 1500 rpm for 2 min and dried by forced convection oven. The sesame seeds with various moisture contents were then roasted with electric roaster until the seed temperature reaches the setup temperature. Roasted seeds were filtered by shaking sorter and automatically transported to cyclone by vacuum. Then, seeds were stayed in the cyclone to be cooled and be dust off. After cooled, seeds were transported to oil presser and pressed one time with 600 $kg_f$.

### 2.4.1. Water content control

Washed seeds were dried with forced convection oven at 80℃ for 2 hours, and moisture content was checked with 10 min intervals to identify drying time. Sesame seeds were controlled to moisture content 25, 15 and 10%. Control group was not washed having about 5% of moisture content which the raw sesame seed normally contains.

### 2.4.2. Roasting temperature control

In order to evaluate the temperature effects, heater temperature was controlled to 220, 250℃ and roasted until the seed temperature reaches 150, 180, 210℃.

### 2.4.3. Ventilation speed control

Ventilator, blower (DB-200, DongKun, InCheon, Korea), was operated to suck out the smoke while roasting. Ventilating speed was set to 0, 15, 30, 45 and 60 Hz (i.e. 0.07, 0.14, 0.21, 0.28 and 0.35 $m^3/s$) by controller.

### 2.4.4. Dust removal control

Dust remover is composed of a filter and a cyclone. Foreign materials bigger than sesame seed was removed by an automatically shaking sieve. Fine dusts were eliminated by cyclone and vacuum system that suck out the dusts.

## 2.5. Fluorescence spectrophotometry of benzo(a)pyrene

SpectraMax m2e multi-mode microplate reader (Molecular Devices, Orleans, CA, USA) was used for fluorescence spectrophotometry. Scanning through the absorption spectrum of a fluorophore while recording the emission intensity at a single wavelength generated the excitation spectrum. Likewise, exciting the fluorophore at a single wavelength (again, preferably the wavelength of maximum absorption) while scanning through the emission wavelengths revealed the emission spectral profile. The excitation and emission wavelength of maximum absorption was 282, 406 nm, respectively (Figure 5).

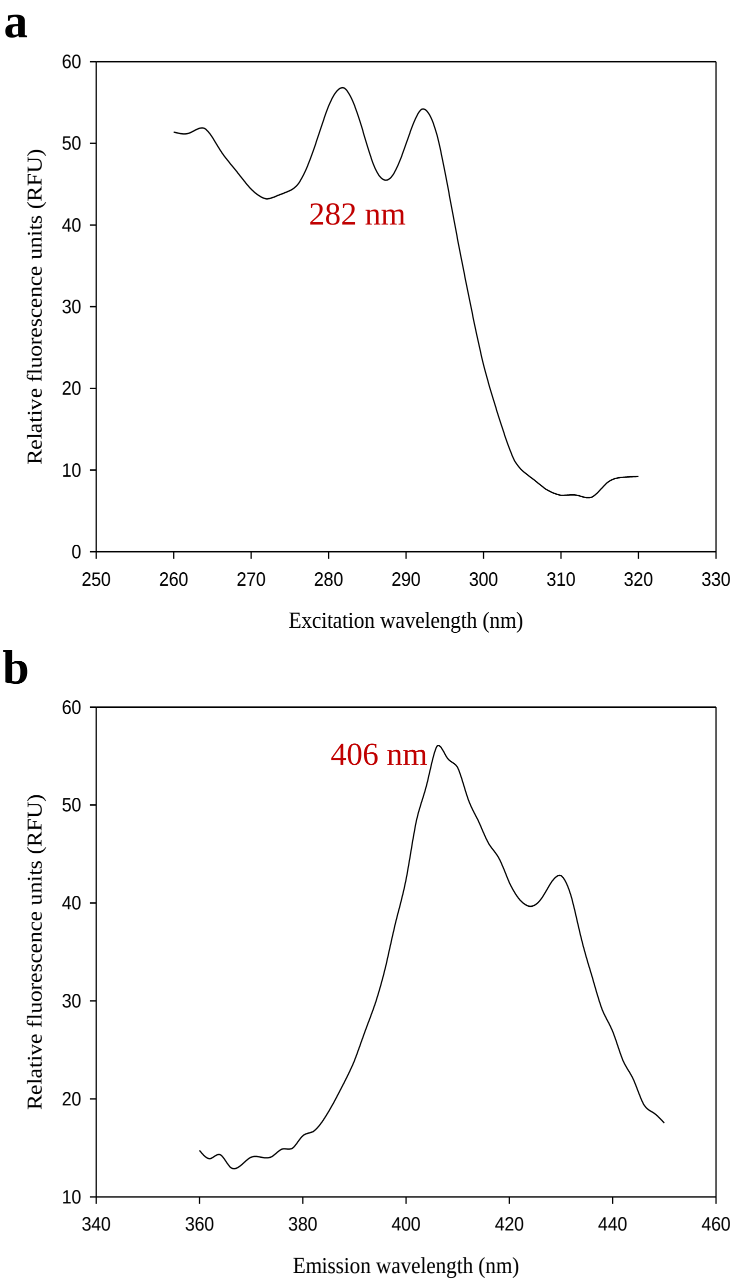



Figure 4. Fluorescence (a) excitation peak between 260 and 320 nm and (b) emission peak between 360 and 450 nm of BaP standard.

## 2.6. Extraction and clean-up

Extraction and purification procedures were based on the method suggested by the Food Code of KFDA, BaP analysis method from meats and edible oil.

### 2.6.1 Seed extraction and clean-up

Five g of sesame seed sample was ground and put into a flask, saponified in 1 M KOH ethanol under reflux at 80°C to isolate BaP bound to the sample or to eliminate the matrix that would hinder BaP analysis. After cooling, 50 mL of *n*-hexane was added through the reflux condenser, and the material was filtered and transferred into a 500-mL separating funnel with 50 mL of an ethanol-*n*-hexane (v/v, 1:1) mixture and *n*-hexane. The resultant solution was extracted twice with 50 mL of *n*-hexane. After combining the extract, it was washed 3 times with 50 mL of distilled water, dried with $Na_2SO_4$ and concentrated under reduced pressure on a rotary evaporator to 2 mL at 40°C.

The cleaned up samples were first eluted with 5 mL of *n*-hexane and 15 mL of *n*-hexane-DCM (3:1) mixture through a florisil cartridge activated in advance. The effluent was blown down by the rotary evaporator at 40°C. The dry residue was re-dissolved in 2 mL of ACN and passed through a PTFE 0.45-μm membrane filter. An aliquot of 1 mL of this solution was injected into the HPLC system.

### 2.6.2 Oil extraction and clean-up

Ten g of homogenized sample was put into a 500-mL separating funnel with 50 mL of an *n*-hexane and 100 mL of N,N-DMF-water (v/v, 9:1) mixture. The resultant solution was extracted twice with 50 mL of *n*-hexane. After combining the extract, it was washed 3 times with 50 mL of distilled water, dried with $Na_2SO_4$ and concentrated under reduced pressure on a rotary evaporator to 2 mL at 40°C. The clean-up process is identical to seed extraction.

## 2.7. Analysis of BaP by HPLC/FLD

HPLC (2695 Separations module, WATERS, Milford, MA, USA) coupled to a FLD (474 FLD, WATERS). Separation of the BaP was achieved by a C-18 column (Hibar 250-4, LiChrospher 100 RP-18 endcapped (5 μm), Merck). Mobile phase A consisted of water and mobile phase B consisted of ACN. The flow rate of 1.0 mL/min was used over 50 min with the following gradient: 0 min, 85% B; 0-30 min, 100% B; 30-50 min, 85% B. The sample injection volume was 40 μL, detection was by fluorescence using excitation at 282 nm and emission at 406 nm. Table 1 shows HPLC/FLD condition using Hibar 250-4, LiChrospher 100 RP-18 column to analysis BaP.

BaP was identified by both retention time and its fluorescence spectra. BaP was quantified on the base of the relative peak ratio of its peak areas to that of the standard curve.

The calibration curve was constructed by BaP standards in ACN. The linearity of the response was tested on 5 calibration levels over the range of 1-100 μg/kg. The limit of detection (LOD) and limit of quantification (LOQ) for BaP in sesame seed and oil were obtained using the signal-to-noise ratio as signal/noise=3 and signal/noise = 10, respectively. Accuracy and precision were evaluated for BaP at the level of 1, 5, 10, 20, and 100 μg/kg.

Table 1. HPLC/FLD conditions for BaP analysis

| | | | |
|---|---|---|---|
| **HPLC/FLD system** | 2695 Separations module, 474 FLD, WATERS | | |
| **Column** | C-18 Hibar 250-4, LiChrospher 100 RP-18 endcapped (150 mm length x 4 mm I.D., 5 μm) | | |
| **Injection volume** | 40 μL | | |
| **Mobile phase** | Gradient method | | |
| | Time (min) | Water (%) | ACN (%) |
| | 0 | 15 | 85 |
| | 0-30 | 0 | 100 |
| | 30-50 | 15 | 85 |
| **Flow rate** | 1 mL/min | | |
| **Detection wavelength** | $\lambda_{ex}$ = 282 nm, $\lambda_{em}$ = 406 nm | | |

## 2.8. Yield of pressed oil

To calculate oil yield, oil cake and pressed oil was used for calculation. Oil cake and pressed oil was weighed after pressing, and the oil ratio was calculated between these two.

$$\text{Yield (\%)} = \frac{Weight\ of\ pressed\ oil\ (kg)}{Weight\ of\ oil\ cake + Weight\ of\ pressed\ oil\ (kg)} \times 100$$

## 2.9. Statistical analysis

The difference between data was represented by means of triplicate determinations and standard deviation. All results were analyzed using Duncan’s test with IBM SPSS Statistics 22 (IBM Co., Armonk, NY, USA).

# III. RESULTS AND DISCUSSION

## 3.1. Validation study

The analytical method used to extract and quantify the sesame oil BaP concentrations was shown to be adequate. In fact, the regression analysis of the data showed that BaP responses were linear over the range of the examined concentrations. The correlation coefficient obtained exceeded 0.9996, demonstrating a good linearity (Figure 6). The LOD and the LOQ were 0.0058 and 0.021 μg/kg, respectively. The peak of BaP standard and Bap extracted from sesame oil is shown at Figure 7.

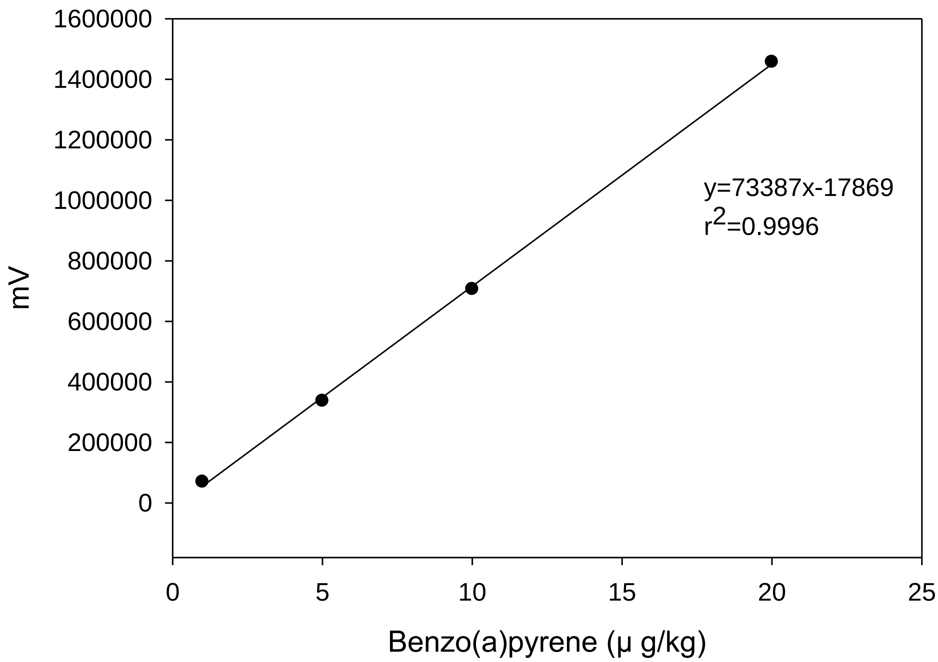



Figure 5. Standard curve of BaP.

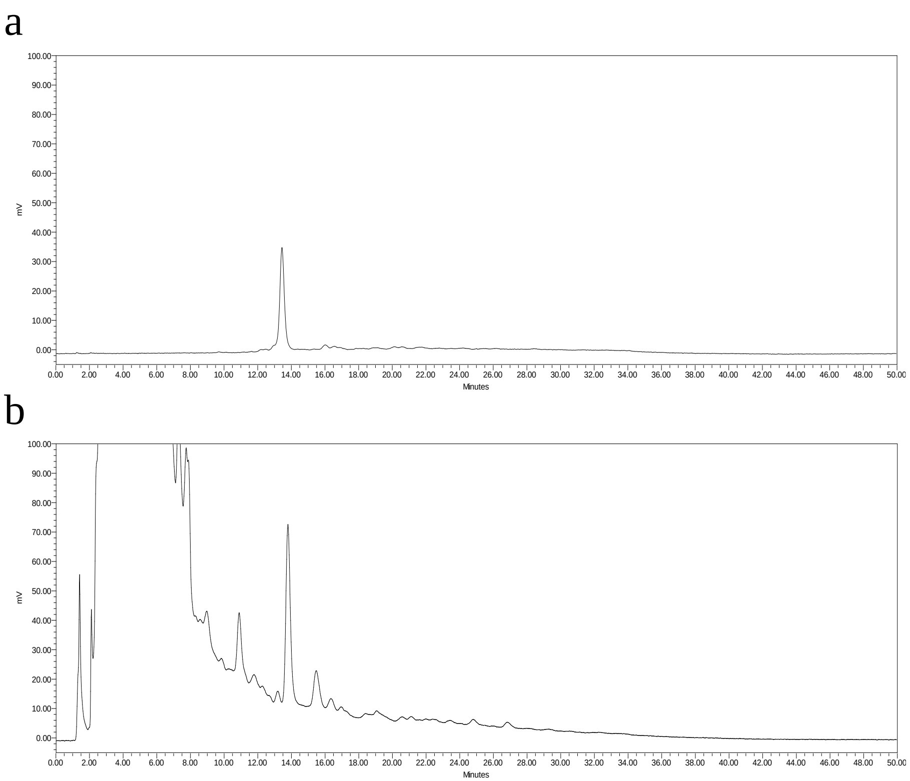



Figure 6. HPLC chromatogram of (a) BaP standard and (b) BaP in sesame oil.

## 3.2. BaP level in sesame oil

Throughout the manufacturing process, BaP content of samlples was checked after each process. According to the result (Table 2), the highest and the second highest BaP production results from the oil bucket and sticked matter of the oil press, respectively. However, it was thought to be solved by cleaning the bucket or the oil press. After that, the absorbed material from smoke and duct fouling had high BaP content, which means the smoke generated during roasting contains BaP. The BaP from the smoke have chance to re-adhere to sesame seeds when not being removed. From this assumption, ventilation could prevent further contamination of BaP during roasting process. Also, dust from roasted seeds contained BaP which is from the burnt hull or foreign materials included in seeds. High BaP content in oil cake might be caused by high temperature and high pressure of the oil press. Thus, double or three times of pressing must represent higher BaP level. In addition, repeated wash produced more BaP in sesame oil, and water content was thought to be the cause. Because sesame seeds were spiked with BaP, all data was represented by the variation of BaP content between spiked seeds and oil.

Table 2. Sample and BaP content according to sesame oil manufacturing process

| Process | Sample | BaP content (μg/kg) |
|---|---|---|
| Raw material | Sesame seed | 0.2 |
| Washing | Washed seed | 0.3 |
| | Washed water | 0.2 |
| Roasting | Roasted seed | 0.4 |
| | Smoke absorption | 0.7 |
| | Duct fouling | 0.8 |
| Dust removal | Dust, Foreign materirals | 0.6 |
| Pressing | Oil press sticked matter | 0.9 |
| | Oil cake | 0.6 |
| | Bucket sticked matter | 1.1 |
| Sesame oil | Sesame oil | 0.5 |

### 3.2.1. Water content and BaP

Raw material, sesame seed, contains about 5% of water content. After washing 2 min, water content increases to 35% and decreases to 25% by drying at 80°C for 30 min. with forced convection dryer. In addition, drying for 90 min. lost moisture of sesame seeds to 15%. As shown at Figure 7, BaP content in sesame oil decreased comparing to spiked seeds after washing and drying to 15% of water content of seeds. Unlike 5% of non-washed seeds, 15% of washed and dried seeds shown lower BaP, meaning that washing might have revealed washing effect which wash off dust from seeds and prevent foreign material get burnt at roasting process. These burnt foreign materials may incline BaP content by being pressed with seeds and getting dissolved to the oil.

However, high moisture content after washing shown less decrease of BaP in sesame oil. When wet seeds go through the roasting process, the water might interfere complete combustion by lowering heating temperature or interrupting oxygen to access. Also, wet seed can be swollen and be separated with its hull which can be burnt during roasting. For this reason, high water content of seeds may have chance to meet incomplete combustion and bring up high BaP concentration in sesame oil. Therefore, drying process should be added in the manufacturing process of sesame oil after washing sesame seeds to prevent additional BaP production during roasting.

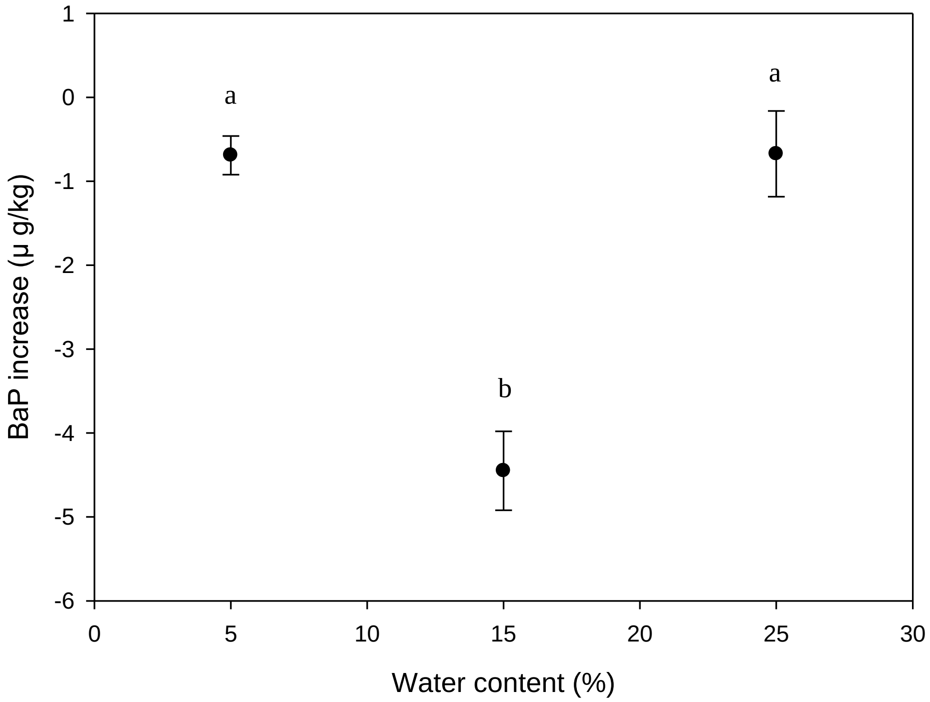



Figure 7. BaP increase in sesame oil from water content of sesame seeds.

### 3.2.2. Roasting temperature and BaP

Sesame oil production contains seed roasting with high temperature, and this process is necessary to bring unique flavor of sesame oil. However, roasting mostly cause high BaP in the oil. Roasting temperature is a widely known factor that cause BaP production. Incomplete combustion of hydrocarbon at high temperature generates BaP especially in foods high in fat. Heater temperature was controlled and roasted until seed temperature reaches the setup temperature. In contrast with earlier studies (Jang GH, 2011; Seo, Nam, & Shin, 2009), as heater temperature was higher, BaP content was lower (Figure 10). Lower heater temperature (220℃) caused seeds to be exposed to high temperature longer time. Therefore, BaP might be increased in this case. Also, Figure 8 represents that BaP content had no statistically significant difference as seed temperature varies. It is also unlike studies (Jang GH, 2011; Kim & Song, 2008; Seo, Nam, & Shin, 2009) that BaP content increased proportional to roasting time.

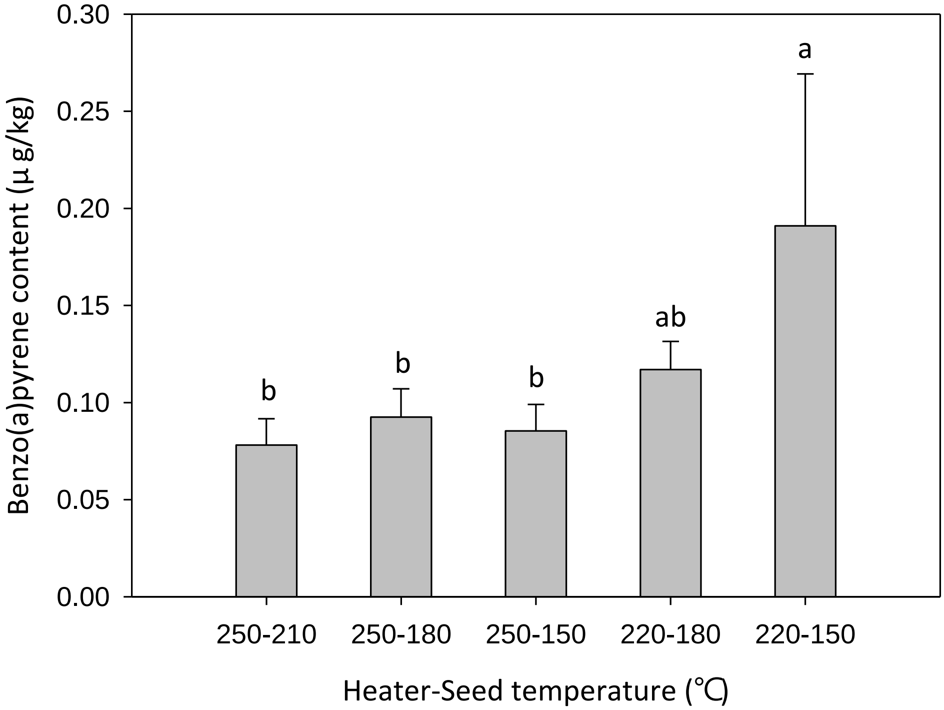



Figure 8. Change of BaP content between spiked sesame seeds and sesame oil which is roasted with different heater temperature (220 and 250℃) and seed temperature (150, 180, and 210°C).

### 3.2.3. Ventilation speed and BaP

During roasting, smoke generated and contained the most BaP in all process (Table 2). Smoke is able to be re-adhered to seeds causing higher BaP in oil product. Removing the smoke by ventilator is essential to reduce BaP. Likewise, Kim, H. and Song, D. (2008) researched that open type roasting contained less BaP than closed type roasting. Ventilating speed influenced BaP content in sesame oil remarking that high speed of ventilation lowers BaP (Figure 11). At low speed, BaP content slightly increased in sesame oil than non-ventilation roasting. It could be thought that low ventilation help hot air circulation in the roaster, and the temperature inside the roaster get higher than before. It might cause more burnt seeds and increase BaP. Until 30 Hz of ventilating speed, BaP content was not significantly decreased but declined at 45 Hz. However, it increased again at 60 Hz of high speed. Too high speed of air circulation brought too much intake of cool air of outside by the inlet. The air temperature inside the roaster had been lowered cooling the seeds and the heater too. Then, the heater indicated heater temperature to be low and raised much higher. The part of the seeds which is near to the heater burnt incompletely and inhomogeneously resulting in higher content of BaP.

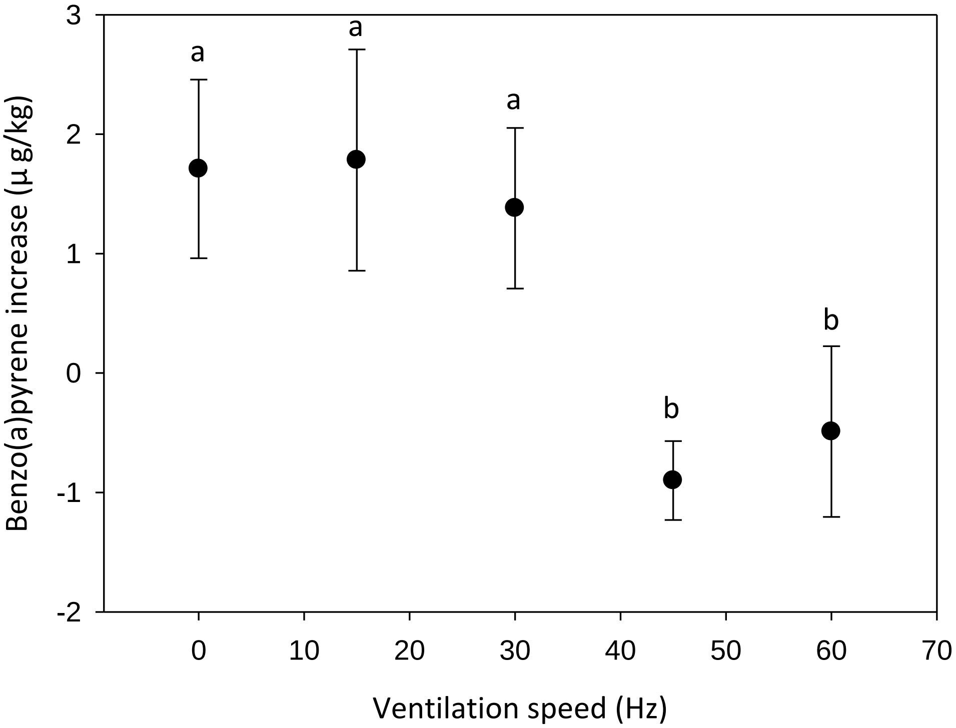



Figure 9. Change of BaP between spiked sesame seeds and sesame oil which is roasted with 0, 15, 30, 45, and 60 Hz of ventilation speed.

### 3.2.4. Dust removal and BaP

Dust removal process was operated automatically after roasting by going through a filter and a cyclone. The filter sorted bigger material than sesame seeds, and the cyclone removed dusts by centrifugal force and vacuuming. Sorting bigger material, for example, rocks or sticks can prevent burnt materials being pressed together and increasing BaP concentration in oil product. The cyclone rotated the seeds and made centrifugal force that separates lighter materials to go upper part of the cyclone, and vacuum system suck out the materials. From removing other things from sesame seeds, BaP content in oil got lower than before (Figure 12a). According Figure 12a, statistical indifference resulted from high deviation, however, all non-operated and all operated condition made a difference between two. Especially dust removing ability declined BaP more than the filter, which indicates that fine dusts from burnt soil or hull contains more BaP. This result correspond to pre-experimented data (Table 2). Also, regardless of the seed temperature or staying time in the cyclone before pressing, BaP content decreased because of the cyclone itself (Figure 12b).

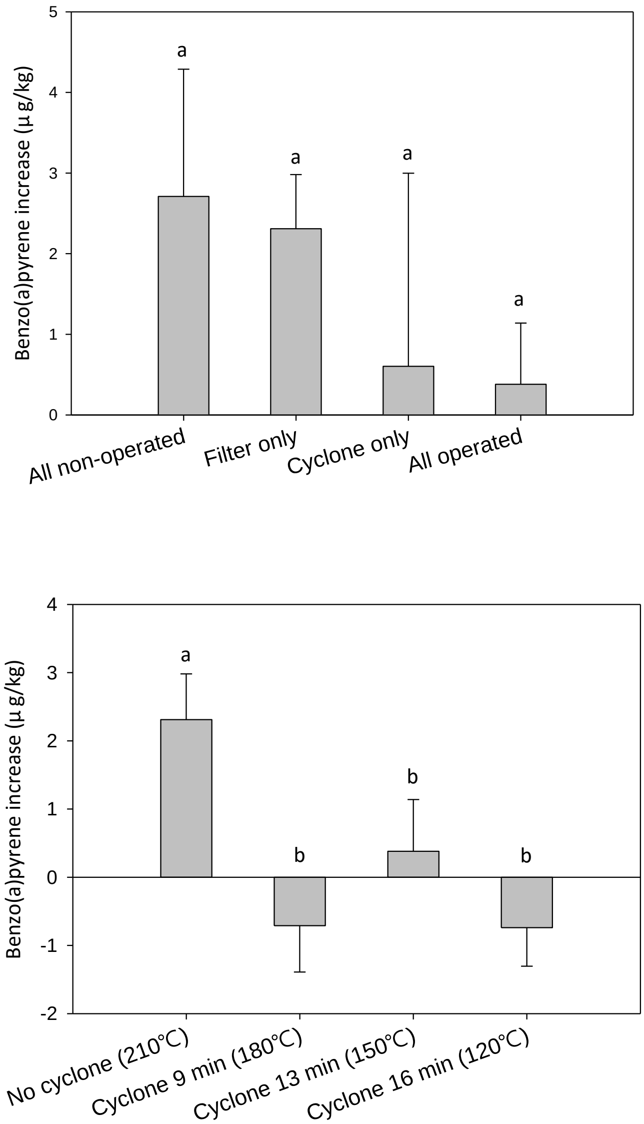



Figure 10. Change of BaP between spiked sesame seeds and sesame oil which is (a) filtered and dust removed and (b) which varied cyclone staying time until seed temperature reaches 180, 150, and 120°C.

## 3.3. Interaction effect of water content and ventilation on BaP concentration in sesame oil

To find out interacting effect of factors that influenced BaP concentration in sesame oil, water content and ventilation speed was controlled for each other. Sesame seeds were non-washed (5%) and washed and dried to water content 25, 15%, then, spiked with 10 ppb of BaP. These seeds sere roasted at 250°C with 0, 20, 40, and 60 Hz of ventilating speed. The BaP content increase between spiked seeds and final oil product are shown in Table 3 and Figure 13. The lowest BaP increase was appeared to be at 15% of water content and 40 Hz of ventilation speed. It represents that appropriate water content and ventilation speed lowers BaP generation during sesame oil manufacturing process. Drying after washing and 40~45 Hz (i.e. 0.19~0.21 $m^3/s$) of ventilation speed is required to produce BaP reduced sesame oil.

Table 3. BaP increase between spiked sesame seeds and sesame oil product which is produced with 5, 15, and 25% of water content and roasted with 0, 20, 40, and 60 Hz of ventilation speed

| Water content (%) \ Ventilation (Hz) | 0 | 20 | 40 | 60 |
|---|---|---|---|---|
| 5 | 0.699 | 0.696 | -0.691 | -0.147 |
| 15 | 3.780 | 0.410 | -4.450 | -3.273 |
| 25 | 1.710 | -0.288 | -1.162 | -0.471 |

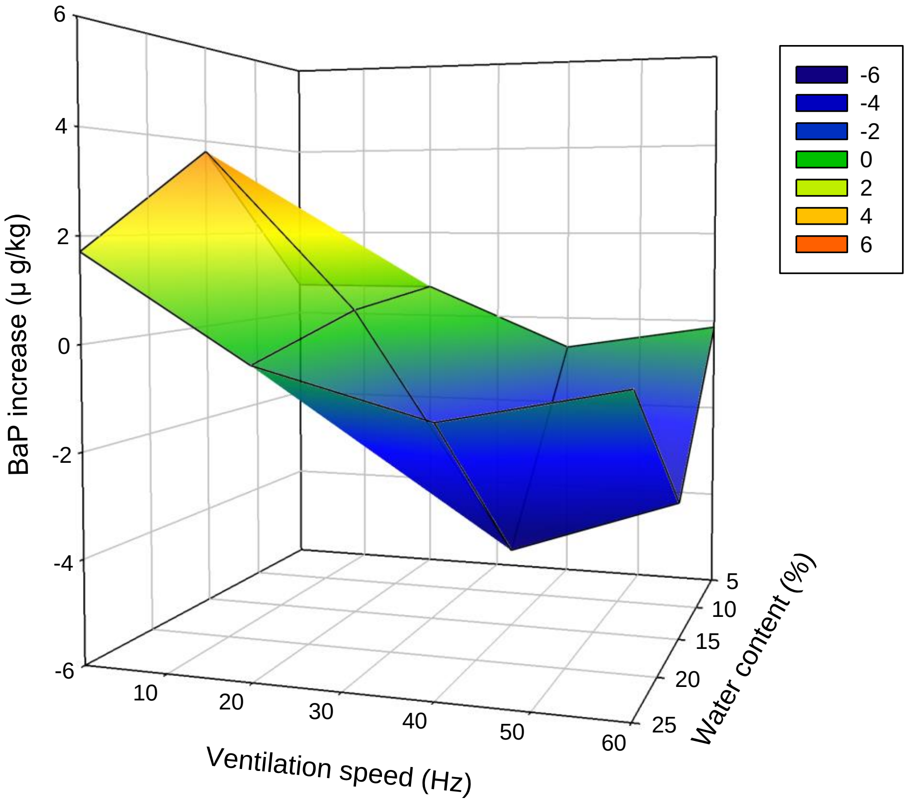



Figure 11. 3d mesh of BaP increase between spiked sesame seeds and sesame oil product which is produced with 5, 15, and 25% of water content and roasted with 0, 20, 40, and 60 Hz of ventilation speed

## 3.4. Yield of pressed oil

Yield of oil from certain amount of sesame seeds is important to sellers or companies. Weight of oil cake and pressed oil was checked every sample and compared to themselves. Regardless of water content and ventilating speed, oil yield was not significantly different from each other (Figure 13a, c). However, roasting temperature affected the oil yield that high heater temperature and high seed temperature caused high yield of oil (Figure 13b). In other words, roasting sesame seeds at high temperature for longer time can make relatively more amount of oil. In addition, seed temperature before pressing, which matches time stayed in the cyclone, affected the oil yield (Figure 13d). Yield increased with seed temperature, but decreased at high temperature which was not using cyclone (210°C). Roasting at high temperature over 200°C might have denaturated protein or structure of outside of the seed and allowed inner oil get out easier. However, too high temperature before pressing without any cooling process caused lower oil yield that might result from blocked structure by excess denaturation. It could make hard outside to be placed inside the seed preventing oil pressed out, and the oil cake gets heavier (Table 3). Considering yield, process of sesame oil production had better be performed with low water content, high roasting temperature for long time, high ventilation speed, and high seed temperature before roasting, but regarding BaP content in the oil, it is recommended to be performed with a certain time of drying after washed, roasting for short time, ventilating adequately, and using cyclone for dust removal.

Among the process, water content, roasting temperature, ventilation speed, and dust removal reduced BaP 35%, 59%, 46%, and 38%, respectively. Roasting temperature and ventilation during roasting were most effective factor on reduction of BaP generation. As a result, considering the effect of BaP reduction and oil yield, 10% of water content after washing, roasting at 250°C until reaching 210°C, ventilating for 0.21 $m^3/s$, and using filter and cyclone was the optimal condition to reduce BaP production in sesame oil manufacturing process. Following the optimal process, sesame oil that have 67% of BaP reduction effect was obtained.

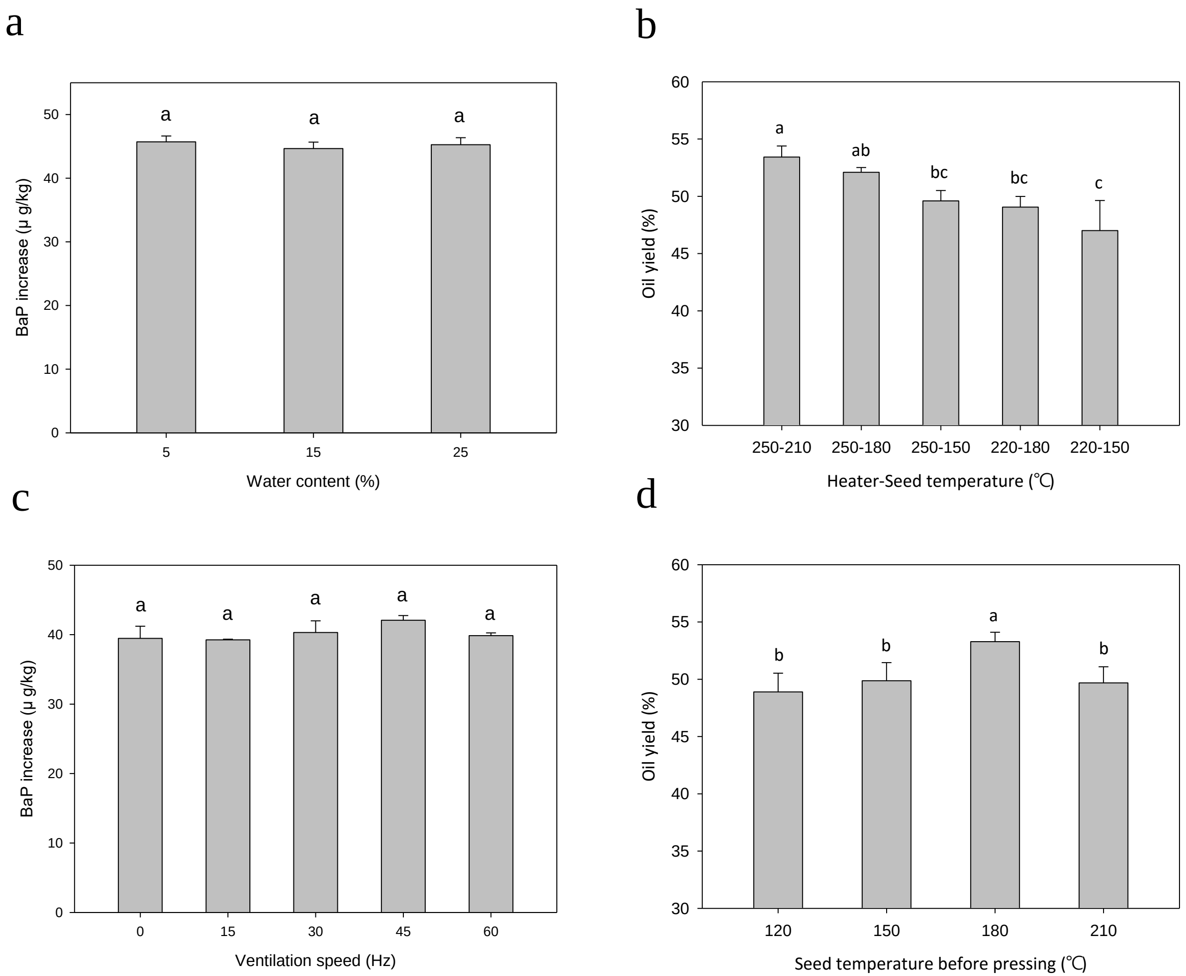



Figure 12. Yield of pressed oil with diverse (a) water content, (b) roasting temperature, (c) ventilation speed, (d) dust removing process.

Table 4.Weight of oil cake and pressed oil that produced by containing diverse water content, roasting temperature, ventilating speed, and seed temperature before pressing

| | Oil cake (kg) | Pressed oil (kg) |
|---|---|---|
| **Water content (%)** | | |
| **5** | 2.13 ± 0.00 | 1.80 ± 0.07 |
| **15** | 2.02 ± 0.14 | 1.63 ± 0.14 |
| **25** | 2.11 ± 0.05 | 1.74 ± 0.04 |
| **Roasting temp. (°C)** | | |
| **250-210** | 2.50 ± 0.07 | 3.14 ± 0.05 |
| **250-180** | 2.66 ± 0.02 | 3.14 ± 0.03 |
| **250-150** | 2.82 ± 0.04 | 3.02 ± 0.07 |
| **220-180** | 2.69 ± 0.18 | 2.81 ± 0.10 |
| **220-150** | 2.86 ± 0.27 | 2.65 ± 0.11 |
| **Ventilation speed (Hz)** | | |
| **0** | 3.21 ± 0.10 | 2.09 ± 0.09 |
| **15** | 3.15 ± 0.12 | 2.03 ± 0.07 |
| **30** | 3.20 ± 0.06 | 2.17 ± 0.13 |
| **45** | 3.08 ± 0.07 | 2.24 ± 0.01 |
| **60** | 3.16 ± 0.01 | 2.09 ± 0.03 |
| **Seed temp. before pressing (°C)** | | |
| **120** | 2.73 ± 0.07 | 2.84 ± 0.12 |
| **150** | 2.72 ± 0.04 | 2.94 ± 0.19 |
| **180** | 2.53 ± 0.08 | 3.14 ± 0.05 |
| **210** | 2.71 ± 0.10 | 2.90 ± 0.06 |

# IV. CONCLUSIONS

In sesame oil, BaP generates during production process from sesame seeds. By repeating washing cycle and extending drying time, water content was increased or decreased, respectively. Both result indicated that roasting seeds high in water content cause more BaP generation. Also, during roasting, heating temperature, completing seed temperature, and ventilating speed was controlled to observe BaP content changing. Heating temperature and time shown significant increase in BaP, which is instructed by exposing more BaP generation at low heater temperature with longer roasting time. Ventilation mostly help to reduce BaP by adequate speed. It seemed high ventilating speed lowers BaP production, but too high speed leaded to high BaP content in oil. In the environment of this study, 45 Hz (i.e. 0.21 $m^3/s$) of ventilation was the optimum speed. In addition, dust removing process was also an important factor for reduction. Especially, the cyclone which gets rid of fine dusts from roasted seeds was effective, but using both of the filter and cyclone brought synergetic effect on BaP reduction. According to each step, water content, roasting temperature, ventilating speed, and dust removing contributed 35%, 59%, 46%, and 38%, respectively, to reduction of BaP content in sesame oil. Combination of these made 67% reduction, in total, showing valid effect of the process.

# V. REFERENCES


1. Jiang, H., et al. (2007). "Metabolism of benzo [a] pyrene in human bronchoalveolar H358 cells using liquid chromatography–mass spectrometry." Chemical research in toxicology **20**(9): 1331-1341.

2. Luch, A. (ed.). The carcinogenic effects of polycyclic aromatic hydrocarbons. World Scientific, 2005.

3. Volk, D. E., et al. Solution Structure of a Cis-Opened (10 R)-N 6-Deoxyadenosine Adduct of (9 S, 10 R)-9, 10-Epoxy-7, 8, 9, 10-tetrahydrobenzo [a] pyrene in a DNA Duplex. Biochemistry, 2003, 42.6: 1410-1420.

4. Eaton, D. L.; Gallagher, E. P. Mechanisms of aflatoxin carcinogenesis. Annual review of pharmacology and toxicology, 1994, 34.1: 135-172.

5. Lijinsky, W. The formation and occurrence of polynuclear aromatic hydrocarbons associated with food. Mutation Research/Genetic Toxicology, 1991, 259(3), 251-261.

6. Shuguang, L., et al. Analysis of polycyclic aromatic hydrocarbons in cooking oil fumes. Archives of Environmental Health: An International Journal, 1994, 49(2), 119-122.

7. Qu, Y. H., et al. Genotoxicity of heated cooking oil vapors. Mutation Research/Genetic Toxicology, 1992, 298(2), 105-111.

8. Chiang, T. A., et al. Mutagenicity and polycyclic aromatic hydrocarbon content of fumes from heated cooking oils produced in Taiwan. Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis, 1997, 381(2), 157-161.

9. Shields, P. G., et al. Mutagens from heated Chinese and US cooking oils. Journal of the National Cancer Institute, 1995, 87(11), 836-841.

10. Shou, M.; Gonzalez F.J.; Gelboin H.V. Stereoselective epoxidation and hydration at the K-region of polycyclic aromatic hydrocarbons by cDNA-expressed cytochromes P450 1A1, 1A2, and epoxide hydrolase. Biochemistry, 1996, 35.49: 15807-15813.

11. Chen, S.H.; Evans, C.E.; McGuffin V.L. Selective fluorescence quenching of polynuclear aromatic hydrocarbons in microcolumn liquid chromatography. Analytica chimica acta, 1991, 246.1: 65-74.

12. Gündel, J.; Angerer, J. High-performance liquid chromatographic method with fluorescence detection for the determination of 3-hydroxybenzo [a] pyrene and 3-hydroxybenz [a] anthracene in the urine of polycyclic aromatic hydrocarbon-exposed workers. Journal of Chromatography B: Biomedical Sciences and Applications, 2000, 738.1: 47-55.

13. Ramesh, A. et al. Metabolism, bioavailability, and toxicokinetics of Benzo (α) pyrenein F-344 rats following oral administration. Experimental and Toxicologic Pathology, 2001, 53.4: 275-290.

14. Strickland, P.T. et al. Identification of 1-hydroxypyrene glucuronide as a major pyrene metabolite in human urine by synchronous fluorescence spectroscopy and gas chromatography-mass spectrometry. Carcinogenesis, 1994, 15.3: 483-487.

15. Sung, T.K. et al. Benzo (a) pyrene contents in commercial vegetable oils and changes during processing of vegetable oils. 2012.

16. Jang, G.H. The Changes of Benzo(a)pyrene in Sesame Oil Affected by Processing Conditions. J. of the Korean Oil Chemists' Soc, 2011, 28.4: 464-471.

17. Pupin, A.M.; Toledo M.C.F. Benzo (a) pyrene in olive oils on the Brazilian market. Food Chemistry, 1996, 55.2: 185-188.

18. Teixeira, V.H. PAHs content in sunflower, soybean and virgin olive oils: Evaluation in commercial samples and during refining process. Food chemistry, 2007, 104.1: 106-112.

19. RODRÍGUEZ-ACUÑA, R. et al. Polycyclic aromatic hydrocarbons in Spanish olive oils: Relationship between benzo (a) pyrene and total polycyclic aromatic hydrocarbon content. Journal of agricultural and food chemistry, 2008, 56.21: 10428-10432.

20. Jira, W. A GC/MS method for the determination of carcinogenic polycyclic aromatic hydrocarbons (PAH) in smoked meat products and liquid smokes. European Food Research and Technology, 2004, 218.2: 208-212.

21. Orecchio, S. et al. Polycyclic aromatic hydrocarbons (PAHs) in coffee brew samples: analytical method by GC–MS, profile, levels and sources. Food and Chemical Toxicology, 2009, 47.4: 819-826.

22. Kazerouni, N., et al. Analysis of 200 food items for benzo [a] pyrene and estimation of its intake in an epidemiologic study. Food and chemical toxicology, 2001, 39.5: 423-436.

23. FALCÓ N, M.S.G., et al. Enrichment of benzo [a] pyrene in smoked food products and determination by high-performance liquid chromatography-fluorescence detection. Journal of Chromatography A, 1996, 753.2: 207-215.

24. Simko, P. Changes of benzo (a) pyrene contents in smoked fish during storage. Food chemistry, 1991, 40.3: 293-300.

25. Anastasio, A. et al. Levels of benzo [a] pyrene (BaP) in "Mozzarella di Bufala Campana" cheese smoked according to different procedures. Journal of agricultural and food chemistry, 2004, 52.14: 4452-4455.

26. Simko, P., et al. Influence of cooking on benzo (a) pyrene content in smoked sausages. Meat science, 1993, 34.3: 301-309.

27. Kim, H. W. et al. Studies on the physicochemical characteristics of sesame with roasting temperature. KOREAN JOURNAL OF FOOD SCIENCE AND TECHNOLOGY, 1999, 31.5: 1137-1143.

28. Ha, J. H. et al. Changes in the physico-chemical properties of the meals from the defatted sesame seeds at various roasting temperature and time. Korean Journal of Food Science and Technology, 1996, 28.2: 246-252.

29. Kahyaoglu, T.; Kaya, S. Modeling of moisture, color and texture changes in sesame seeds during the conventional roasting. Journal of Food Engineering, 2006, 75.2: 167-177.

30. Cejpek, K., et al. Changes in PAH levels during production of rapeseed oil. Food Additives & Contaminants, 1998, 15.5: 563-574.

31. Moss, J. R.; Otten, L. A relationship between colour development and moisture content during roasting of peanuts. Canadian Institute of food science and technology journal, 1989, 22.1: 34-39.

32. Sebastian, P., et al. Drying and smoking of meat: heat and mass transfer modeling and experimental analysis. Journal of food engineering, 2005, 70.2: 227-243.

33. Hwang, K.S, et al. Quality evaluation of sesame oil by high performance liquid chromatography. Korean Journal of Food Science and Technology, 1984, 16.3: 348-352.

34. Lee, J.Y.; Kim, M.J; Choe, E.O. Study on the changes of tocopherols and lignans and the oxidative properties of roasted sesame oil during manufacturing and storage. Korean Journal of Food Science and Technology, 2008, 40.1: 15-20.

35. Park, C.H. et al. Studies on the improvement of roasting condition of sesame seeds for producing seed season and oil. Korean Journal of Crop Science/Hanguk Jakmul Hakhoe Chi, 2011, 56.3: 205-211.

36. Shin, B. R.; Yang, S. O.; Kim, Y. S. Trends in the reduction of benzo [a] pyrene in sesame oils Food Ind. Nutr, 2014, 19: 5-12.

37. Kim, H.Y.; Song, D.S. Minimizing benzo (a) pyrene content in the manufacturing of sesame oil and perilla oil. Korean Journal of Food Preservation, 2008, 15.4: 556-561.

38. Seo, I. W.; Nam, H. J.; Shin, H. S. Influence of polycyclic aromatic hydrocarbons formation in sesame oils with different roasting conditions. Korean Journal of Food Science and Technology, 2009.

39. Oh, S.C. Benzo(a)pyrene Reduction in Sesame Oil Using Microwaving Method. J. of Korean Oil Chemists' Soc., 2012, 29.2: 323-329.

40. Ledesma, E.; Rendueles, M.; DÍAZ, M. Spanish smoked meat products: Benzo (a) pyrene (BaP) contamination and moisture. Journal of Food Composition and Analysis, 2015, 37: 87-94.

41. Lijinsky, W.; Ross, A. E. Production of carcinogenic polynuclear hydrocarbons in the cooking of food. Food and cosmetics toxicology, 1967, 5: 343-347.

42. Cho, H.K.; Shin, H.S. Analysis of benzo [a] pyrene content from smoked food products in Korea. Food Science and Biotechnology, 2012, 21.4: 1095-1100.

43. Nam, H.J.; Seo, I.W; Shin, H.S. Analysis of Polycyclic Aromatic Hydrocarbon Content in Coffee Beans with Different Preparation Method. Korean Journal of Food Science and Technology, 2009, 41.2: 157-161.

44. Peterson, G. S., et al. Evaluation of a fluorometric screening method for predicting total PAH concentrations in contaminated sediments. Environmental monitoring and assessment, 2002, 78.2: 111-129.

45. Pinckney, J.; Papa, R.; Zingmark, R. Comparison of high-performance liquid chromatographic, spectrophotometric, and fluorometric methods for determining chlorophyll a concentrations in estaurine sediments. Journal of Microbiological Methods, 1994, 19.1: 59-66.

46. Hu, S.J, et al. Determination of polycyclic aromatic hydrocarbons in processed foods. Analytical Science and Technology, 2010, 23.2: 196-204.

47. Chiu, C. P.; Lin, Y. S.; Chen, B. H. Comparison of GC-MS and HPLC for overcoming matrix interferences in the analysis of PAHs in smoked food. Chromatographia, 1997, 44.9-10: 497-504.

48. Purcaro, G.; Moret, S.; Conte, L. S. Rapid validated method for the analysis of benzo [a] pyrene in vegetable oils by using solid-phase microextraction–gas chromatography–mass spectrometry. Journal of chromatography A, 2007, 1176.1: 231-235.

49. Lee, W., et al. Studies on the Analysis of Benzo (a) pyrene and Its Metabolites on Biological Samples by Using High Performance Liquid Chromatography/Fluorescence Detection and Gas Chromatography/Mass Spectrometry. Bulletin of the Korean Chemical Society, 2003, 24.5: 559-565.

50. Schwarz, F. P.; Wasik, S. P. Fluorescence measurements of benzene, naphthalene, anthracene, pyrene, fluoranthene, and benzo [e] pyrene in water. Analytical chemistry, 1976, 48.3: 524-528.

51. Prough, R. A.; Patrizi, V. W.; Estabrook, R.W. The direct spectrophotometric observation of benzo (a) pyrene phenol formation by liver microsomes. Cancer research, 1976, 36.12: 4439-4443.

# VI. 국문초록

참기름은 국내에서 가장 많이 소비되는 향미유로, 최근 1 급 발암물질인 벤조피렌이 기준치(2 μg/kg)를 초과한 참기름이 적발되면서 그 안전성이 우려되고 있다. 참기름은 높은 열로 볶는 과정을 거치고 높은 압력으로 착유를 함으로써 생성공정 중에 비의도적인 벤조피렌이 생성된다. 따라서, 본 연구에서 자동 세척기와 볶음솥, 배기장치, 이물제거기, 착유기로 이루어진 설비를 고안하고, 설비를 통하여 벤조피렌 저감화를 위한 참기름 제조 공정을 최적화 하였다. 공정 최적화를 위해 세척 이후의 수분함량, 볶음 온도, 배기 속도, 이물제거 여부를 통한 참기름 제조 후 벤조피렌 함량 변화를 확인하였다. 원재료인 참깨의 수분함량은 약 5%로 세척 이후 25%로 증가하였다. 세척 횟수와 강제 대류 건조를 통해 수분함량을 30, 25, 15, 10%로 조절하였고, 수분함량이 높을수록 벤조피렌 함량이 증가되는 것을 확인하였다. 하지만 세척을 하지 않은 참깨를 이용하여 제조된 참기름에서 벤조피렌 함량이 조금 높게 나타났고, 이는 참깨 중

이물질의 탄화로 인해 나타난 것을 알 수 있었다. 또한 볶음 과정 중 볶음 온도와 배기는 참기름의 벤조피렌 함량에 매우 큰 영향을 끼치는 요인이었다. 볶음 온도는 히터 온도를 220, 250°C 로 조절하였고, 완료 곡물 온도가 150, 180, 210°C 가 될 때까지 가열하였다. 히터 온도 250°C 로 볶음 시 40 분 이내로 210°C 에 도달하였고, 220°C 로 볶음 시 150, 180°C 에 도달하는 데에 1 시간 이상의 시간이 소요되었으며 벤조피렌 함량이 높게 나타났다. 이를 통해 볶음 온도보다 볶음 시간이 벤조피렌 함량에 더 영향을 끼치는 것을 알 수 있었다. 볶음 중 배기의 영향 또한 벤조피렌 함량을 저감 시키는 요인 중 하나였고, 이를 확인하기 위하여 배기 속도를 0, 15, 30, 45, 60 Hz 로 조절하였다. 45 Hz 까지 배기 속도를 증가시킴에 따라 벤조피렌 함량이 감소하는 경향을 보였으나, 60 Hz 의 빠른 배기 속도에서는 오히려 벤조피렌이 증가하는 것을 알 수 있었다. 이는 배기 속도가 빨라지면서 외부의 차가운 공기의 유입이 많아지고 볶음솥 내부의 온도를 떨어뜨리는 역할을 하여, 볶음 시간이 길어지며 히터에 직접적으로 맞닿은 부분은 더 타는 효과가 나타났다. 그러므로 적절한 배기 속도에 대한 계산이

필요할 것으로 생각된다. 이물제거 단계는 크기가 큰 이물을 거르는 filter 와 미세 이물을 제거하는 cyclone 으로 이루어져 있으며, cyclone 의 벤조피렌 저감 효과가 더 큰 것으로 나타났다. 하지만 filter 와 cyclone 을 동시에 사용할 시, 시너지 효과를 내어 더 많이 저감되는 것을 확인할 수 있었다. 결론적으로, 낮은 수분함량, 짧은 볶음 시간, 적절한 배기 속도, 이물제거 공정을 통해 각각 35, 59, 46, 38%의 저감효과를 확인하였고, 전 공정을 통해 기존 제조 공정에 비하여 벤조피렌이 67% 저감된 참기름을 생성하였다. 이 연구를 통하여 벤조피렌이 저감된 참기름을 제조 할 수 있었으며, 다른 압착유 기준 공정 제조에 바탕이 될 수 있을 것이다.

주요어: 참기름, 벤조피렌, 제조 공정, 수분 함량, 볶음 온도, 배기 속도, 이물제거

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