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# Effect of Sodium Hydroxyde (NaOH) and Sodium Hypochlorite (NaHClO) on Morphology and Mineral Concentration of *Zea mays* Hairs (cornsilk)

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## ABSTRACT

Prehistoric and ancient civilizations have long relied on dietary minerals in the prevention of diseases. Major minerals serve as structural components of tissues, function in cellular and basal metabolism, and in water and acid-base balance. This study is discussing on the effect of NaOH and NaHClO on morphology and mineral concentration of cornsilk. A smear layer like soft gelatinous adherent covering the uppermost part of the NaOH treated cornsilk while the cornsilk treated by NaHClO was filled with various features of grape-like globules. The amounts of Ca, K, Mg, Mn and Zn in fresh and oven dried cornsilk were comparatively higher than in samples treated with NaOH and NaHClO. The concentration of Ca in fresh and oven dried cornsilk was accounted for 546 and 323 mg/L, respectively and higher than the concentration of cornsilks macerated with NaOH (108 mg/L) and NaHClO (7.6 mg/L). Both cornsilk treated with NaOH and NaHClO recorded the highest concentration of Na (30533 and 60200 mg/L, respectively). In conclusion, NaOH and NaHClO were detrimentally and distorted natural cytoskeleton of the tissues by removing essential organic and inorganic components. The highest mineral contents presented in fresh and oven dried cornsilk were Ca, K, Mg, Mn and Zn.

**Keywords**: *Maydis stigma* or cornsilk, NaOH, NaHClO, mineral

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## INTRODUCTION

Herbs, also known as botanicals, are one of humanity's oldest known health care therapeutic essentials for a sustainable health, where essentially it forms the basis platform of modern medicines. Globally, multi-diverse primitive and ancient civilizations relied on herbs for healing, as do contemporary cultures throughout the world. In fact, the World Health Organization (WHO) has expressed an estimated 80% of the world's population will continues to use traditional therapies, a major part of which are derived from locally available plants.

Into the new millennium, throughout the world, there is now an inclination and search for stable diet with added value thus an increasing interest in the importance of dietary minerals in the prevention of several diseases. There are different types of dietary or nutritional supplements. Vitamin and mineral supplements are types of dietary supplements containing micronutrients meant to help a healthy body function smoothly. Minerals are of critical importance in the diet, even though they comprise only 4–6% of the human body. Major minerals are those required in amounts greater than 100 mg per day and they represent 1% or less of bodyweight (Lukaski, 2004). These include calcium, phosphorus, magnesium, sulfur, potassium, chloride and sodium. Trace minerals are essential in much smaller amounts, less than 100 mg per day, and make up less than 0.01% of bodyweight. Essential trace elements are zinc, iron, silicon, manganese, copper, fluoride, iodine and chromium (Ozcan, 2004). Human, as well as animal, studies originally showed that optimal intakes of elements such as sodium, potassium, magnesium, calcium, manganese, copper, zinc and iodine could reduce individual risk factors, including those related to cardiovascular disease (Sanchez-Castillo et al., 1998).

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Cornsilk is a collection of the stigmas (fine, soft, yellowish threads) from the female flowers of the maize plant. This yellowish thread-like strands or tassels called stigmas are found inside the husks of corn. They are relatively 4 – 8 inches long with a mild sweetish taste. Cornsilks are scientifically referred as *Maydis stigma* or *Zea mays* which reflect the soft, fiber-like growth which accompanies the ear of the corn. The stigmas are found on the female flower of corn and are collected for traditional herbal medicine remedy before the plant is pollinated (Maksimovic et al., 2005). Cornsilk is highly valued in herbology as a support to the urinary system (Maksimovic and Kovacevic, 2003). It contains silicon, B vitamins, para aminobenzoic acid (PABA) and small amounts of iron, zinc, potassium, calcium, magnesium and phosphorus (Fleming, 2000). Consilk was used to soothe the urinary tract and can give relief to the bladder, kidneys and small intestine (Yeşilada et al., 1995). Although not scientifically proven, rhetorically, cornsilks have long been reported in ancient literatures to be able to assists with prostate problems, bed-wetting, carpel tunnel syndrome, edema and obesity (Velazquez et al., 2005). It has also been used to lessen the effects of premenstrual syndrome, and it promotes relaxation (Maksimovic et al., 2005).

Because of the believed that these soft silk or 'hairs' are postulated to be comfortably eliminated during gastrointestinal processing, they are thus considered as an anecdote for several ailments related to soothing and healing from urinary tract infections, as antispasmodic, as a mild diuretic, urinary demulcent, to pass stones and gravel from the kidneys and urinary bladder, depuratives and vermifuges (Ozcan, 2004). It is also thought to act against benign prostatic hyperplasia, cystitis, gout (Maksimovic et al., 2005), chronic nephritis and anti-hyperglycemic effect (Li et al., 2004). Similar findings have

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been published in several ethno-pharmacological studies dedicated to surveys of popularly used medicinal plants in various regions, or the plants used specifically for medical treatment of urological disorders (Cáceres et al., 1987; Yeşilada et al., 1995). Also, some local species are used as tea, powdered as a food additive and flavorings agents in several regions of the world (Koedam, 1986; Yesilada and Ezer, 1989). Cornsilk have also been reported to have polyphenol compounds, an entity considered as an important parcel of a possible commercial viable herbal drug (Maksimovic and Kovacevic, 2003).

There are indications that utilization and acceptance tendency towards medicinal herbs to give relief and treat human ailments are globally very positive although there are side effects. The interest toward elucidating the chemical composition of medicinal herb products (Basgel and Erdemoglu, 2006), volatile and non-volatile components of cornsilk (El-Ghorab et al., 2007) and antioxidant activities of cornsilks (Ebrahimzadeh et al., 2008) are also growing as commercializing exploitation increases. The objective of this study is to investigate the effect of sodium hydroxyde (NaOH) and sodium hypochlorite (NaHClO) on morphology and mineral concentration of *zea mays* hairs (cornsilk).

## MATERIALS AND METHODS

### Sample preparation

Fresh *Zea mays* hair or cornsilk (dried cut stigmata of *Zea mays* L, Poaceae female flowers) was collected from Pantai Cahaya Bulan, Kota Bharu, Kelantan, Malaysia. Harvested samples were divided into 2 groups. The samples were either macerated with (a) NaOH or (b) NaHClO. To prepare macerated samples, the cornsilk threads were

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soaked and agitated (Rotamax 120, Heidolph, German) for 2 hours in 2.5M NaOH and 5.25% sodium hypochlorite (NaHClO). The macerated samples were then filtered with filter paper (Whatman no.1). The macerated and fresh samples were then freeze dried (Ilshin model TFD5505 Korea) for 24 hours.

**Scanning electron microscopy**

The dried samples were coated with a thin-layer of gold in a vacuum evaporator (Baltex SCD005 Sputter Coater, Hi-Tech Germany) and their structure and morphology were studied in a LEO 1455 VPSEM under 5.5 Pa, using the secondary electron mode, at a working distance of 10 mm and 5.0 kV (Wan Rosli et al., 2008).

**Determination of mineral and trace elements**

The concentration of macrominerals (Ca, Na, K and Mg), trace elements (Fe, Zn, Cu, and Mn) hypothesed were determined using Atomic Absorption Spectrophotometer (AAS) model AAnalyst 800 (Perkin Elmer) according to the adapted method by (Tee et al., 1996). The organic fraction of cornsilk was initially destroyed by dry ashing at 550$^{o}$C for 5 hrs (until a whitish ash) was obtained. The ash was dissolved in concentrated HCl and then filtered (utilising Whatman no.1), diluted into 10 ml volumetric flask and read directly on the AAS. Hollow cathode lamp of Ca and Mg element operated at 5 mA were used at 393 and 279 nm wavelengths, respectively, with optimized air-acetylene flame. For preparation of all solutions and samples, high-purity water from a Milli-Q system (Millipore, Milford, USA) was used. During the experiments, all glassware and equipment was carefully cleaned initially with 2–4% solutions of $HNO_3$ and finally rinsed

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repeated with deionized water to prevent contamination. All the standard solutions used (1, 10, 100 and 1000 µg/l) were prepared by diluting 1 mg/ml stock multi-element standard solutions for AAS (AAnalyst 800 Perkin Elmer).

**Statistical analysis**

Data obtained were tested for significance using ANOVA and Duncan Multiple Range Test with SAS version 6.12 (SAS, 1989). Significance was established at $P \leq 0.05$ unless otherwise indicated.

## RESULTS AND DISCUSSION

Figure 1a and 1b electron photomicrographs of cornsilks post macerated with 2M of sodium hydroxide (NaOH) revealed cross-sectional diameter measurements that range from 247 to 249 µm. As illustrated in Figure 1a, the protuberances are structurally collapsed. When the longitudinal infolding was further inspected, numerous fine micro cracks were observed present indenting the hairs. The NaOH treatment is seems here to have caused substantial detrimental surface and internal texture tissues changes illustrated by morphological presence of the smear layer covering the outermost surface of the threads. These smear layer was observed present at various interval of the infolding of the threads. Smear layer is like soft gelatinous adherent like structure/texture covering the uppermost part of the folded surface (1b). The smear layer is hypothetically are organic and inorganic sediments of NaOH maceration of the hairs. Although, there was efforts to explore individual microtubule of the sample preparation, it seems that the whole tubule have totally collapsed and is now occluded.

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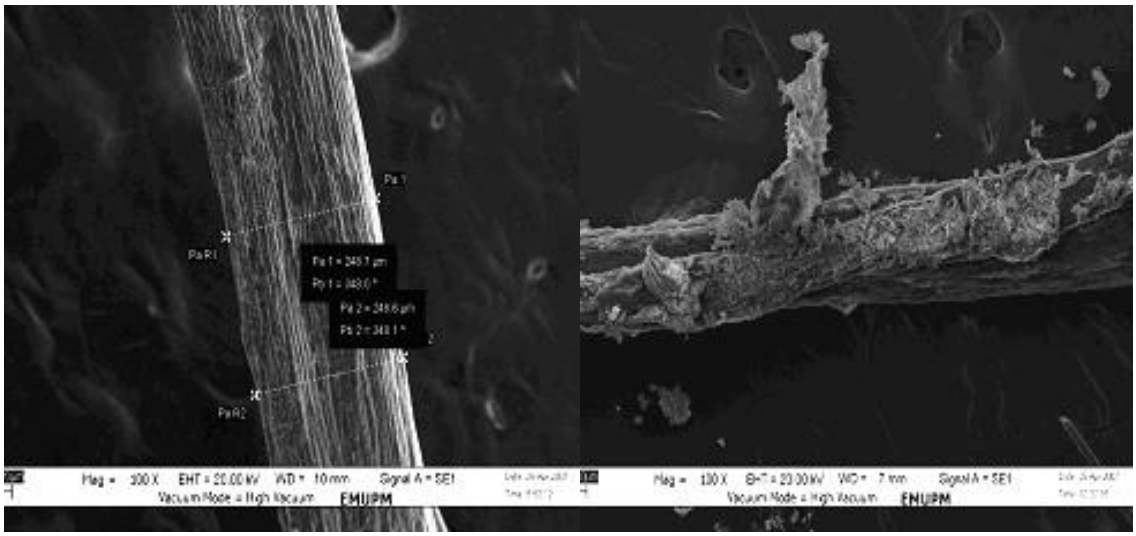

1a 1b

**Figure 1. Scanning electron photomicrographs of a cornsilk fiber post-treatment with 2M NaOH showing the absent of protuberances (1a) and the present of gelatinous adherent like texture layer (1b)**

Figure 2 shows scanning electron photomicrographs of cornsilk threads bleached with 5.25% NaHClO. At 100X magnification, the bleached cornsilk have a smear or shine layer on it surface (2a). The formations of various smear layers exhibit a form like globule-like structure (2b). There were so many globule-like structures covering the surface of the longitudinal hairs. Although the thread of *Zea mays* hair treated by NaHClO flattened but the tubules are still patented and open although the surrounding area filled with various features of grape-like globules (2a). Enlarged grape-like globules can be seen in Figure 2b. The specimen of another bleached thread fibre was almost transparent in presentation (2c). The presence of the protuberance of the other side of the body can be observed through and through observation (2c). This protuberance was staying posterior side of the thread (marked with white circle). At the

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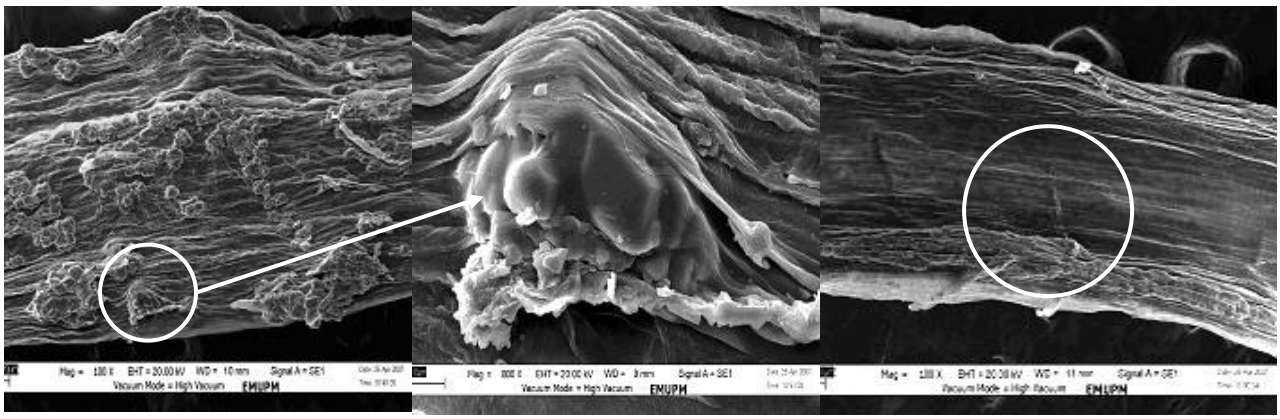
terminal of this filament, the microtubules observed are almost structurally collapse and flatten.

From this observation, it can be suggested that macerated treatments have dissolved some of the compounds like pectins and hemicellulose. Alkali treatment can partially or completely cleave the ester bonds in cell walls (Sun and Hughes, 1999).



**2a** **2b** **2c**

**Figure 2. Scanning electron photomicrographs of a cornsilk fiber post-treatment with 5.25% NaHClO at 100X magnification (2a), 800X magnification (2b) and bleached transparent thread fibre (2c)**

The concentrations of the eight elements were determined via AAS (Table 1). It was observed that all cornsilk samples contain significant values of elements and the element content in the samples presented a wide variability. The amounts of Ca, K, Mg, Mn and Zn on fresh and oven dried cornsilk were comparatively higher than mineral detected in samples treated with NaOH and NaHClO.

The concentration of Ca in fresh and oven dried cornsilk was 546.0 and 323.3 mg/L, respectively. These values were significantly higher than the values detected in cornsilks

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macerated with NaOH (108.0 mg/L) and NaHClO (7.6 mg/L), respectively. Oven dried cornsilk was recorded the highest in Mg concentration (1433 mg/L) followed by fresh cornsilk (409.3 mg/L). However, this macromolecule was detected at lower concentration in both cornsilk macerated with NaOH and NaHClO which accounted for 56.3 and 78.3 mg/L, respectively. The decline in Ca and Mg observed in both macerated cornsilk may be due to the bleaching effect done by these chemicals. They dissolve a small part of membranes and cell wall of the cornsilk. The decline in Ca and Mg concentration detected in cornsilk treated with NaOH and NaHClO also could be due the dissolution of the Ca and Mg which function as a structural role as components of membranes and cell wall (Lopez et al., 1997).

Calcium and Magnesium seems to be ubiquitous in all the samples studied. The role of these two elements in health and disease are well known. Calcium is a major macronutrient and believed to possess the ability to prevent precancerous cell morpho-differentiation into malignancy by binding to cancer promoting fats thus inhibiting their ability to initiate cancerous growth. Calcium is also an important regulator of many cellular mitotic activities (Kirschmann, 1998). Per se, magnesium has been quite a forgotten cation from the therapeutic point of view, given that, although its properties have long been elucidated for more than a century ago, its usage has been always empirical or isolated. deficit of magnesium is obvious in clinical situation associated with hypercalcemia and hypopotassemia, (del Castillo et al., 1992).

Both cornsilk treated with NaOH and NaHClO recorded the highest concentration of Na (30533 and 60200 mg/L, respectively) compared to the other treatments. The

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concentration of Na in fresh and oven dried cornsilk was 246.3 and 400.7 mg/L, respectively and significantly lower than macerated samples (NaOH and NaHClO).

In fresh and oven dried cornsilks, K was the most abundant element. This electrolyte accounted for 20950 mg/L and 6000 mg/L in both oven dried and fresh cornsilk and significantly higher than cornsilk macerated with NaOH and NaHClO (431.3 and 471.7 mg/L), respectively. This result has confirmed the finding reported in previous study which documented that potassium being the most plentiful electrolyte detected via Energy Dispersive X-ray (EDX) technique in fresh and oven dried cornsilks (Wan Rosli et al., 2007). Dietary potassium deficiency is not known to occur in man, because it is well absorbed and are abundantly present in natural foods especially in raw vegetables (Prasad et al., 2000). The sodium content of plants and vegetables is often increased during processing procedures, while the potassium content is generally depleted, which is probably due to leaching (Prasad et al., 2000). In plants or vegetables, both electrolytes (Na and K) are mainly in the phloem and play an important role in the maintenance of the membrane potential (Lopez et al., 1997).

The trace element or oligo-element zinc (Zn) was detected in high concentration (64 and 72 mg/L, respectively) in both fresh and oven dried cornsilk. These values were significantly higher than the values detected in cornsilks macerated with NaOH and NaHClO (11.7 and 19.7 mg/L, respectively). Fe detected in lower concentration ranging from 12.1 – 36.4 mg/L in all samples. Together with Ca and Mg, Fe is believed to be closely bounded to the cell wall structure (Lopez et al., 1997).

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Cu content was detected for 4.7 and 4.1 mg/L, respectively in fresh and oven dried samples and not significantly different compared to NaHClO (5.0 mg/L). The concentration of Mn detected in raw fresh and oven dried cornsilks were significantly higher (9.7 and 6.4 mg/L) than the value detected in cornsilk treated with NaOH (1.9 mg/L) and NaHClO (not detected). The documented function of Mn in the plant cell is mainly related to the photosynthesis (Barcelo et al., 1992) and no other structural function has yet to be described.

Mineral are elemental atoms or ions, in contrast to micro molecules they do not undergo changes during digestion or when the body utilizes them. Minerals play many important roles in the body and they are not destroyed by heat, light or alkalinity. The major mineral serve as structural components of tissues and function in cellular and basal metabolism and water and acid-base balance (Nielsen, 1984; Ozcan, 2004). Magnesium, iron, zinc, copper and chromium serve as structural or catalytic components of enzymes and regulate cellular energy transduction, gas transport, antioxidant defense, membrane receptors functions, second-messenger systems and integration of physiologic systems (Lukaski, 2004). The elements found in living organisms may be essential, i.e. necessary for growth and health, or they may be nonessential, accidental reminders of our geochemical origins or indicators of environmental exposure. Plant foods can be excellent sources of several minerals, but the minerals content of plants can vary dramatically depending on the minerals in the soil where the plants are found (Lopez et al., 1997).

Bioavailability of the minerals depends on the form of their bond with the constituents of the soil. Plants readily assimilate through the roots such elements, which are dissolved in water and occur in the ionic forms. Additional sources of these elements

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available to plants are rainfall, atmospheric dusts, plant protection agents and fertilizers, which may adsorbed through the leaf blades (Basgel and Erdemoglu, 2006).

## CONCLUSION

Cornsilk treated with NaOH shows a smear layer like soft gelatinous adherent covering the uppermost part of the cornsilk surface while the cornsilk treated by NaHClO was filled with various features of grape-like globules. NaOH and NaHClO were detrimentally and distorted natural cytoskeleton of the tissues by removing essential organic and inorganic components. Atomic Absorption Spectroscopy methods used have given the wide variability in the concentration of elements presented in the cornsilks. The highest mineral contents presented in fresh and oven dried cornsilk were Ca, K, Mg, Mn and Zn. To understand the medicinal benefits of cornsilk and its related extracts, further detail investigations on the other nutritional properties and pharmaceutical characteristics of cornsilk extracts will be conducted soon.

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**Table 1: Concentrations of elements in fresh, oven dried and macerated cornsilks (**mg/L)

| | Ca | Cu | Fe | K | Mg | Mn | Na | Zn |
|---|---|---|---|---|---|---|---|---|
| Fresh | 546.0 ± 2.7[a] | 4.7 ± 0.5[ab] | 12.1 ± 0.7[d] | 6000 ± 1.5[b] | 409.3 ± 2.5 [b] | 9.7 ± 0.1 [a] | 246.3 ± 0.6 [d] | 64.0 ± 2.0 [b] |
| Oven dried | 323.3 ± 0.6[b] | 4.1 ± 0.2[b] | 32.1 ± 0.4[b] | 20950 ± 7.7[a] | 1433 ± 5.7 [a] | 6.4 ± 0.1 [b] | 400.7 ± 1.5 [c] | 72.0 ± 1.0 [a] |
| NaOH | 108.0 ± 2.0[c] | 2.9 ± 0.0[c] | 36.4 ± 0.1[a] | 431.3 ± 2.1[d] | 56.3 ± 0.6 [d] | 1.9 ± 0.0 [c] | 30533 ± 57 [b] | 11.7 ± 1.5 [d] |
| NaHClO | 7.6 ± 0.1[d] | 5.0 ± 0.1[a] | 27.1 ± 0.0[c] | 471.7 ± 1.5[c] | 78.3 ± 0.6 [c] | Not-detected | 60200 ± 173 [a] | 19.7 ± 1.2 [c] |

[a-d] Mean values within the same column bearing different letters differ significantly (P<0.05)

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