

## Quantitative Aspect of Constitutive Regulation Intestinal Condition by Fermented Herbal Decoction through Permeability

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### Abstract

**Background:** This report concerns the CAM menu that regulates intestinal conditions. In our several reports, fermented herbal decoction (FHD) could activate the complement surrogate pathway through the intestinal membrane with an appropriate molecular weight to hit the complement initiation molecules. The purpose of this report is to confirm the change of the physiological state by the uptake of the bystander molecule through FHD.

**Methods and Participants:** We tried many menus of CAM in healthy subjects who could control leukocyte subsets. This paper mentions menus such as hot spring water therapy, light exercise, floor heating, and famous decoction of TCM including fragrance. Leukocyte subsets were tracked before and after the CAM menu, and the time schedule was adjusted to match the circadian rhythm.

**Results:** FHD could activate the complement alternative pathway through the intestinal membrane with an appropriate molecular weight to hit complement. Under the same conditions, minerals, Ca, Mg, K, and vitamin, Vc, and Vd molecules showed high molecular absorption. In such conditions, minerals tended to be transported faster than vitamins.

### Conclusion:

- 1) Quantitative regulation of the total leukocyte count could be assessed by complement activation.
- 2) FHD increased the intestinal absorption of minerals Ca, Mg, and K.
- 3) FHD was found to increase intestinal absorption of vitamin C and vitamin D.
- 4) Absorption proceeded to minerals and then vitamins.

**Keywords:** QOL; Finger Movements; Constitution; Complement; Decoction of Fermented Herbs; Intestinal Flora; Intestinal Absorption; Bowl Movement

### Abbreviations

CAM: In addition to complementary and alternative medicine and Western medicine, there are other traditional medicine and health promotion menus around the world; DM: Diabetes Mellitus; FHD: Fermented Herb Decoction; G-rich type: Peripheral blood granulocytes account for more than 60% and are more common among young people

### Introduction

The allergic reaction is fundamentally the normal physiological reaction for non-self substance and immune function. Asthma, however, is characterized by chronic airway inflammation and is associated with bronchial hyper responsiveness, causing various airflow

limitations and respiratory symptoms [1]. Airway epithelial cells play an important role in innate immune function in the lung. Airway epithelial cells are also characteristic of mucociliary cells and physically remove pathogens through a process known as mucociliary movement. This process involves trapping pathogens in mucus produced by the inflamed airways and removing the mucus through the movement of cilia present on epithelial cells [2]. Preventive attention is very important in planning treatment. For that purpose, it is important to consider the constitution from the prevention side of allergic reaction. The purpose of this report is to introduce the CAM menu which can change the composition by some menus of CAM with moderate menu, not boldly. Abo reported that white blood cell subsets and lymphocyte/granulocyte ratios varied among individuals [3-5]. Then, the purpose of this report is to show the importance of writing the digital word in order to clarify the condition of each physiological condition in allergic disease. The selection of the health menu associated with each constitution is difficult due to the lack of information about these interactions among the general public and among health professionals resulting in potentially important health scales. In other words, there is still no interim measure to assess the degree of concentration of each trial. Therefore, we report the best candidates for digitally expressing the lymphocyte/granulocyte ratio and the results of the regulatory vector expressed as a linear function through [6-9] such as CAM therapy. With these reports we reported that the immune system were closely related to the QOL that crosstalk with leukocyte in quantitatively and qualitatively [10-17].

## Target and Method

### Subject

15 healthy subjects (Mean age  $45.5 \pm 10.2$  years, 29 ~ 65 years for both sexes) were recruited and concurred according to the Problem Ethics Committee of Kanazawa Medical University. Gender was 46.6% for women and 53.4% for men. His entourage was those of medical schools and their descendants. The participants were divided into two groups, each group being expelled from the menu, and blood samples were taken at the same time as the initial sampling. One to several more weeks after cooling, the attendant was changed for testing.

### Create FHD

Commercial fermented herbal contemplation labeled MANNYOH (FHD) was purchased with (Echigo Yakusa Co., Ltd. Joetsu City, Niigata Prefecture). The conventionally available purified water prepared by reverse dialysis in the laboratory of Kanazawa Medical University was set as the control [18].

### FHD preparation, fermentation, and GABA production

80 commercially available wild herbs were prepared and roasted materials were extracted up to 10 gr for 3 min in 100 ml hot water (98°C). Fermentation was carried out by *Lactobacillus leutia* at 40°C for 5 days. The ratio of powder, lactobacilli and water produced by ECHIGO YAMUSOU was 100: 50: 850 (Joetsu City, Niigata Prefecture). After centrifugation at 2000 xg for 10 minutes at room temperature, the supernatant was subjected to FHD and GABA: gamma-amino acid butyric acid was evaluated in the test system [19]. Followings were the method for quantifying  $\gamma$ -aminobutyric acid, which comprises the steps of producing reduced nicotinamide adenine di-nucleotide phosphate by using a specific aminotransferase and a dehydrogenase that needs to use oxidized nicotinamide adenine dinucleotide phosphate as a coenzyme and deactivating the enzymes, thereby removing any amino acid having a similar structure to that of GABA and acting an electron carrier on NADPH produced in the aforementioned step in the presence of a tetrazolium salt that can produce a water-soluble formazan dye and measuring the water-soluble formazan dye described and reported in references [20].

### Statistical analysis

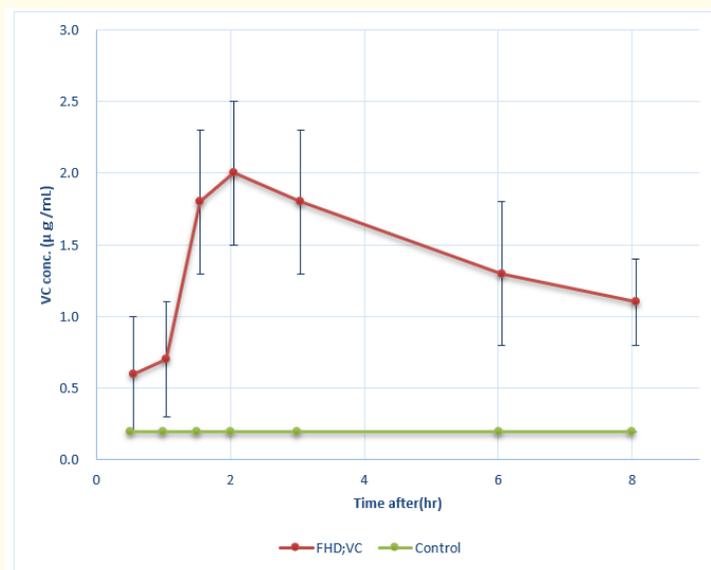
The statistical analysis along with the groups (before and after trial) for the test of significant difference were calculated by paired t-test and wilcoxon signed-ranks test. As for the examination of the correlation was calculated a spearman's correlation coefficient by rank test. Data are expressed as means  $\pm$  standard error of mean (SE). A P value  $< 0.05$  was regard to be statistically significant.

## Result

### Absorbance evaluation of minerals and vitamin molecules

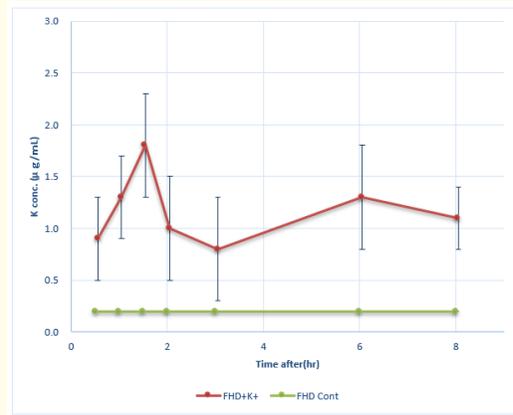
Selected 15 healthy attendants (Mean age  $46.5 \pm 10.2$  years, 29 ~ 65 years for both sexes) were notified to and agreed with the Kanazawa Medical University Ethics Committee. Total leukocyte count and differential leukocyte count in peripheral blood of 15 patients tended to increase after FHD administration (Figures 1 through 5). Reproducible results were obtained in Figure, indicating that the vector turning point was 40 years of age (Figure 3).

According to the Kanazawa Medical University Ethics Committee, 15 recruited healthy attendants (Mean age  $46.5 \pm 10.2$  years, 29 ~ 65 years for both sexes) were informed and agreed to participate. The temporal disappearance of endogenous antioxidants in human plasma associated with the appearance of various classes of lipid hydroperoxides, determined by HPLC post-column chemiluminescence detection, was investigated under two oxidation conditions. Exposure of plasma to aqueous peroxy radicals generated at a constant rate results in immediate oxidation of endogenous ascorbic acid and sulfhydryl groups, followed by sequential depletion of bilirubin, uric acid, and  $\alpha$ -tocopherol. Plasma FHD stimulation initiates very rapid oxidation of ascorbic acid, followed by a partial increase in uric acid. When ascorbate is completely consumed, micromolar concentrations of plasma phospholipids, triglycerides, and cholesterol ester hydroperoxides appear simultaneously, even though sulfhydryl groups, bilirubin, urate, and  $\alpha$ -tocopherol are still present in high concentrations. Non-esterified fatty acids, the only lipid class in plasma that is bound to albumin but not transported into lipoproteins, are conserved after complete oxidation of ascorbic acid with each oxidative damage, possibly due to site-specific antioxidant protection by albumin-bound bilirubin (Figure 1-3).



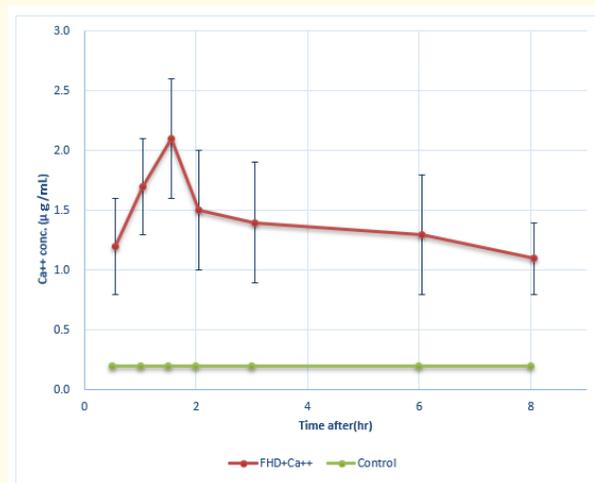
**Figure 1:** Assessment of K Ion.

About the same trial was set up after the cooled down to the same volunteers. Instead of mineral ion from vitamins, another member subjected in this study were after giving informed and consented. We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm [21] of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).



**Figure 2:** Assessment of Mg Ion.

About the same trial was set up after the cooled down to the same volunteers. Instead of mineral ion from vitamins, another member subjected in this study were after giving informed and consented. We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm [21] of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).

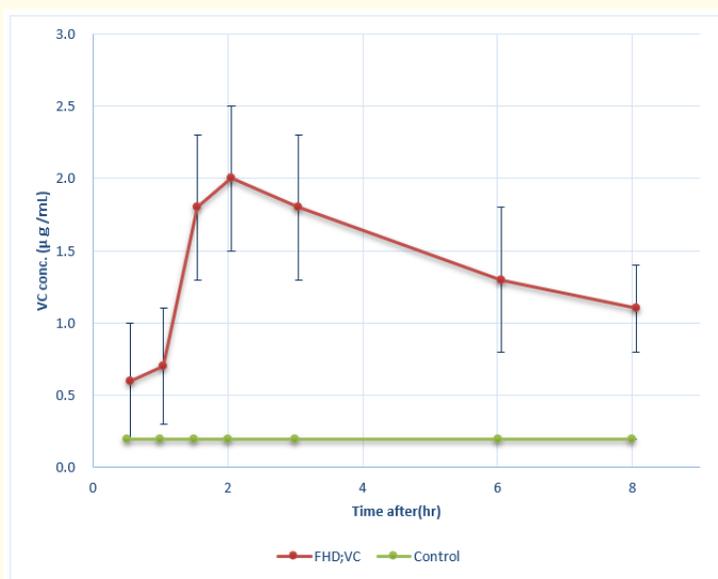


**Figure 3:** Assessment of Ca Ion.

About the same trial was set up after the cooled down to the same volunteers. Instead of mineral ion from vitamins, another member subjected in this study were after giving informed and consented. We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm [21] of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).

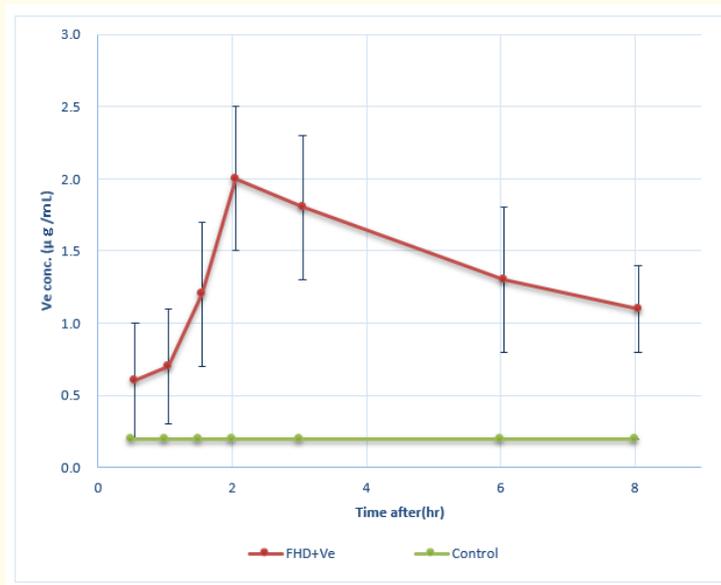
### Evaluation of vitamin molecules

According to the Kanazawa Medical University Ethics Committee, 15 recruited healthy attendants (Mean age  $46.5 \pm 10.2$  years, 29 ~ 65 years for both sexes) were informed and agreed to participate. The temporal disappearance of endogenous antioxidants in human plasma associated with the appearance of various classes of lipid hydroperoxides, determined by HPLC post-column chemiluminescence detection, was investigated under two oxidation conditions. Other members of the study were informed and agreed to. All participants resumed material exchange for the same test after a cooling period of 20 days. The vitamin molecules were vitamin C and vitamin D. Peripheral blood was collected from the same 15 volunteers before and after the study at the same time on the day of the study to adjust the circadian rhythm of white blood cell counts [21]. Exposure of plasma to an aqueous peroxy radical solution generated at a constant rate rapidly oxidized endogenous ascorbic acid and sulfhydryl groups, followed by sequential depletion of bilirubin, uric acid, and  $\alpha$ -tocopherol. Plasma FHD stimulation initiates very rapid oxidation of ascorbic acid, followed by a partial increase in uric acid. When ascorbate is completely consumed, micromolar concentrations of plasma phospholipids, triglycerides, and cholesterol ester hydroperoxides appear simultaneously, even though sulfhydryl groups, bilirubin, urate, and  $\alpha$ -tocopherol are still present in high concentrations. Non-esterified fatty acids, the only lipid class in plasma that is bound to albumin but not transported by lipoproteins, are conserved per oxidative damage even after complete oxidation of ascorbic acid, probably due to site-specific antioxidant protection by albumin-bound bilirubin and possibly albumin itself. Thus, in plasma ascorbic acid and site-specific methods, bilirubin appears to be much more effective in protecting lipids from per oxidation (Figure 4 and 5).



**Figure 4:** Assessment of Vc Molecule.

About the same trial was set up after the cooled down to the same volunteers. Instead of mineral ion from vitamins, another member subjected in this study were after giving informed and consented. We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm [21] of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).



**Figure 5:** Assessment of Vd Molecule.

About the same trial was set up after the cooled down to the same volunteers. Instead of mineral ion from vitamins, another member subjected in this study were after giving informed and consented. We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm [21] of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).

**Addition of floras classified according to the object**

Volunteers aged  $10.2 \pm 46.5$  years with a range of 29 ~ 65 years were nominated for this study. Before the final stage, all the applicants were self-surveyed. At the end of the study, volunteers were selected based on their medical history. To confirm the patient’s condition, each participant provided written information and agreed to the study. As a representative of the absorption enhancement by FHD, the substances effective for the absorption enhancement were compared with the health promotion standard which is famous for the nutrient of intestinal flora such as yogurt, chitosan, oligosaccharide. The results are shown in tables 1a-1d, respectively. From the results of this study, FHD was the best material for upregulating goody bacillus. A similar trend revealed that FHD was most suitable for down-regulation of ciliated rod counts in the intestine.

<b>Table 1a</b>							
	<b>0.5 hr</b>	<b>1 hr</b>	<b>1.5 hr</b>	<b>2 hr</b>	<b>3 hr</b>	<b>6 hr</b>	<b>8 hr</b>
FHD+Mg++	1.0 ± 0.4	1.2 ± 0.4	1.5 ± 0.5	2.2 ± 0.5	1.6 ± 0.5	15 ± 0.5	10 ± 0.3
FHDx2+Mg++	12 ± 0.5	1.4 ± 0.4	1.6 ± 0.4	1.5 ± 0.6	1.4 ± 0.4	0.7 ± 0.6	0.5 ± 0.3
Cont	<1						

<b>Table 1b</b>							
	<b>0.5 hr</b>	<b>1 hr</b>	<b>1.5 hr</b>	<b>2 hr</b>	<b>3 hr</b>	<b>6 hr</b>	<b>8 hr</b>
FHD+K+	1.2 ± 0.4	1.7 ± 0.4	1.8 ± 0.5	2.0 ± 0.5	1.8 ± 0.5	1.3 ± 0.5	1.1 ± 0.3
FHDx2+K+	1.6 ± 0.5	1.8 ± 0.4	1.9 ± 0.4	2.2 ± 0.6	1.9 ± 0.4	0.8 ± 0.6	0.9 ± 0.3
FHD Cont	<1						

<b>Table 1c</b>							
	<b>0.5 hr</b>	<b>1 hr</b>	<b>1.5 hr</b>	<b>2 hr</b>	<b>3 hr</b>	<b>6 hr</b>	<b>8 hr</b>
FHD+VC	0.2 ± 0.4	0.7 ± 0.4	0.8 ± 0.5	0.5 ± 0.5	0.2 ± 0.5	0.3 ± 0.5	0.1 ± 0.3
FHDx2+VC	0.6 ± 0.5	0.8 ± 0.4	0.9 ± 0.4	0.17 ± 0.6	0.6 ± 0.4	0.4 ± 0.6	0.6 ± 0.3
Cont	<1						

<b>Table1d</b>							
	<b>0.5 hr</b>	<b>1 hr</b>	<b>1.5 hr</b>	<b>2 hr</b>	<b>3 hr</b>	<b>6 hr</b>	<b>8 hr</b>
FHD+Vd	1.2 ± 0.4	1.7 ± 0.4	1.8 ± 0.5	2.0 ± 0.5	1.8 ± 0.5	1.3 ± 0.5	1.1 ± 0.3
FHDx2+Vd	1.6 ± 0.5	1.8 ± 0.4	1.9 ± 0.4	1.7 ± 0.6	1.6 ± 0.4	0.8 ± 0.6	0.9 ± 0.3
Control	< 1						

**Table 1a-1d:** We figured out the purpose of this study by setting the control group at the same interval and same building. After 20 days of cooling down for all attendants, we started again for the same trial exchanging a mineral for trial. We collected peripheral blood from the same 15 volunteers before and after the trial, at the same time zone on the day, adjusting a circadian rhythm of leukocyte by gas chromatography (assayed and reported by MILS International Co. Ltd., Yokohama, Japan).

### Discussion

The data in this report showed that within 24 h after each CAM, leukocytes in the peripheral blood could be significantly controlled not only by the total number of leukocytes but also by the lymphocyte and granulocyte subset ratios. The results showed that these subsets could reflect immunocompetent cells in quantity and quality [21-26]. For example, in individuals with low numbers of granulocytes, such numbers are upregulated after the menu, but in other individuals with high cell numbers, they are downregulated. Our results also suggested that the leukocyte subset could be an interesting indicator for the efficacy of alternative drug therapy. Many ethnic medicine practices have been used to evaluate Western therapies aimed at curing the symptoms of this disease. The constitutional studies reported that there were two major types of leukocyte subsets, the lymphocyte-rich type and the granulocyte-rich type, depending on the content of the lymphocyte subsets. The granulocyte rich type showed over 60% of granulocyte in the leucocyte, on the other, lymphocyte rich type exhibit over 40% of lymphocyte [7,22].

This classification was evident in emotional hormones, adrenaline, and dopamine. Men of the same age and sex can be classified into G-rich type (Granulocyte 60%) and L-rich type (Lymphocytes 40%). We intend to regulate the mature immune response by the fragile daily condition of COPD by environmental stress, etc. In this report, acupuncture treatment, hot spring hydrotherapy, light exercise, floor heating, etc. are made to be main menus, and the control mechanism of light motion walking is introduced as a tailored scale. Except for contraindications, it has been medically recognized to be effective in improving many stress disorders, dysfunction of biological rhythms, and chronic diseases. It is important to be conscious of the circadian rhythm in order to evaluate the change after the menu correctly in the digital word except for VAS. The reports also proposed that the selection of peripheral leukocytes [2-4] and composition, granulocyte-rich individuals, and lymphocyte-rich individuals is possible. In this report, a simple comparison of the grouped values showed that the

simple summation and the created average did not show a valuable change in each individual. It is reported that leukocyte subset, granulocyte, lymphocyte + emotion hormone are regulated by various factors. One important point is that they are controlled by the autonomic nervous system and control the circadian rhythm [2-4]. Therefore, in order to obtain the effect in a short time, it is important to adjust the sampling time considering this factor. For example, the effects and effects of walking light exercise are widely accepted as important. However, the majority of pedestrians did not indicate the extent of the effect that is appropriate for each constitution. Then, in this report, it was made that the importance of writing the digital word was shown in order to clarify the condition of each physiological condition in the allergy to be a purpose.

One important point is that they are controlled by the autonomic nervous system and control the circadian rhythm [2-4]. Therefore, in order to obtain the effect in a short time, it is important to adjust the sampling time considering this factor. For example, the effects and effects of walking light exercise are widely accepted as important. However, the majority of pedestrians did not indicate the extent of the effect that is appropriate for each constitution. Then, the purpose of this report is to show the importance of writing the digital word in order to clarify the condition of each physiological condition in allergic disease.

### Conclusion

When leukocyte subset counts, granulocyte counts, and lymphocyte ratios were measured in different CAM modes, the following conclusions were obtained.

- 1) Quantitative regulation of total leukocyte count could be assessed by down-regulation, i.e., tailored scale for each configuration.
- 2) FHD was found to increase the total levels of minerals, Ca, Mg, and K.
- 3) FHD increased the total amount of vitamin Vc and Vd.
- 4) Each CAM was demonstrated to be down-regulated for granulocytes in number but up-regulated for lymphocytes in adults and elderly individuals.

### Conflict of Interest

No conflict of interest hit in this trial.

### Acknowledgment

We all appreciated to the relevant, especially for the student members who attended for each trial.

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