A Study on the antioxidation effects and radiation damage tolerance by Lettuce cultivated with EMX

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Introduction

EM-X, an extract from beneficial plants fermented with EM, has been observed of its strong power of antioxidation on numerous occasion. The power, however, is not traced back to any specific antioxidant, while a variety of antioxidants and functional minerals are found existing, though in a very small quantity.

The present paper examines for its power:
Experiment 1: to eliminate existing free radicals in the living organism with anti-oxidant enzymes and antioxidants enhanced in its growth with EM-X.
Experiment 2: to prevent DNA damage induced with the acute stress of ?-irradiation at the presence of lettuce extract grown with EM-X.
Experiment 3: to reduce the generation of hydroxyl radical to be induced with the acute stress at the presence of lettuce extract grown with EM-X.

Details of EM

EM was developed by Professor Dr. Teruo Higa, at the University of the Ryukyus, Okinawa, Japan. The fundamental principle of this technology is the introduction of a group of beneficial microorganisms to improve soil conditions, suppress putrefying (disease-inducing) microbes and enhance crop utilization of organic matters.
Researches has shown that the introduction of EM cultures to the soil/plant ecosystem can improve the quality and health of soil, and the growth, yield, and quality of crops. Als, when used in animal husbandry, similar benefits have been observed.

EM can significantly enhance the beneficial effects of good soil and crop management practices such as crop rotations, use of organic amendments, conservation tillage, crop residue recycling, and biocontrol of pests. EM can help the decomposition process of organic materials, and during fermentation will produce normally unavailable organic acids, amino acids, and malic acid, bioactive substances and vitamins. A key factor in this process in this process is is the existence of organic matter, which can be ensured by recycling crop residues, green manures and animals manure. In addition, this process leads to increased humus in the soil. Lactic acid bacteria that are major microorganisms in EM will suppress pathogenic microbes, both directly and indirectly through the production of actinomyctes. Also known is EM's antioxidation effect which improves the immune system of plants and animals.

Details of EM-X

EM-X is an epoch-making refreshment drink extracted from rice bran and seaweeds that have been fermented with Effective Microorganisms (EM) and contains potent antioxidants. Major components of EM-X are minerals, antioxidants, and bioactive substances. Based on the firm belief that nature provides the best, EM-X does not contain any artificial vitamins, Chinese medicines or chemical additives that are often added to other health drinks.

EM-X is made from only those substances provided in the natural fermentation process, and is rich with antioxidants, 40 kinds of minerals, and bioactive substances such as NAD, NMN, peptide, and such amino acids as L-alanine and L-glutamine. It has power strong enough to stop chain reactions that are initially caused by activated oxygen.

Materials & Method

Lettuce (Lactuca sativa) was cultivated in hydroponic with only a chemical fertilizer, Otsukahouse, and chemical fertilizer with 1% EM-X. We sued Otsukahouse brand chemical fertilizer and dissolved 150mg type 1 and 100mg type 2 in 100L water and irrigated them.

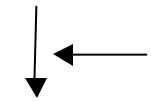
Chemical alone and chemical with 1% EM-X were adjusted for the fertilizer effect by the EC value, respectively.

Table 1 parameter of Analyses

Parameter	**Method**
SOD activity	TBZ method
Ascorbic acid	Hydrazine method
TBARS (MDA)	TBA method
Aldehydes	TBA method

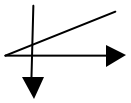
Experiments 2 and 3 Leaf Extraction

Leaf 1.0g



Add of 10 ml DW

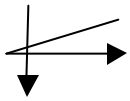
Homogenized



Spain down

Filtration (double gauze)

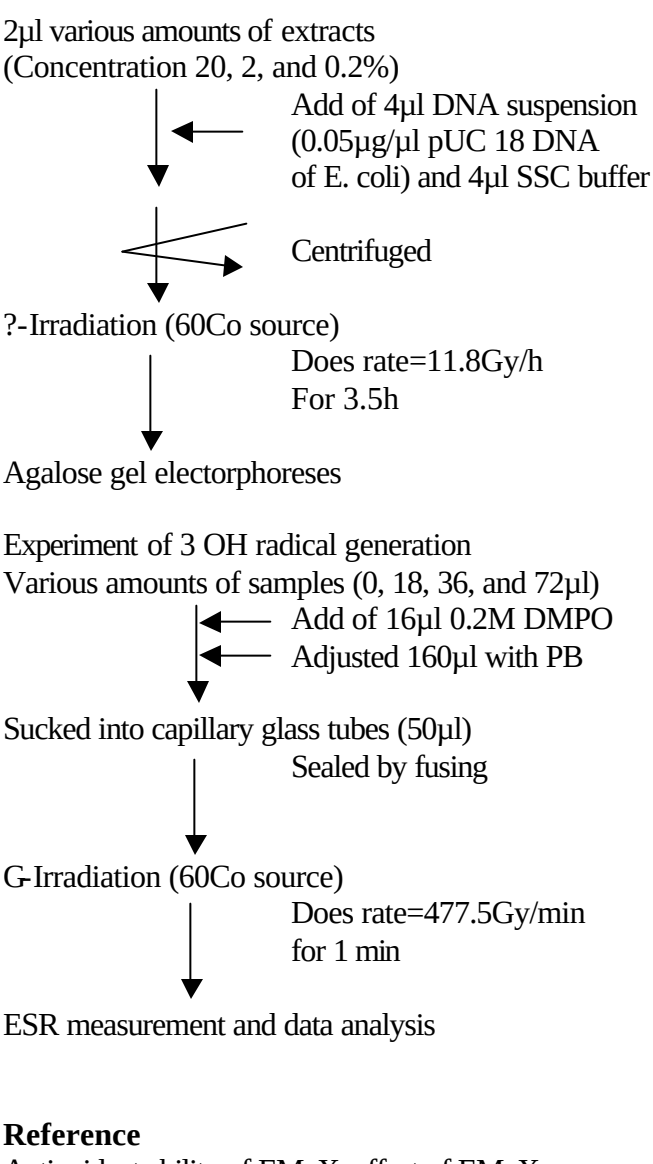
Centrifuged
15,000 x g for 15 min



Suspernatant

Stored -20^{o}C until analysis

Experiment 2 DNA oxidative damage



Experiment of 3 OH radical generation

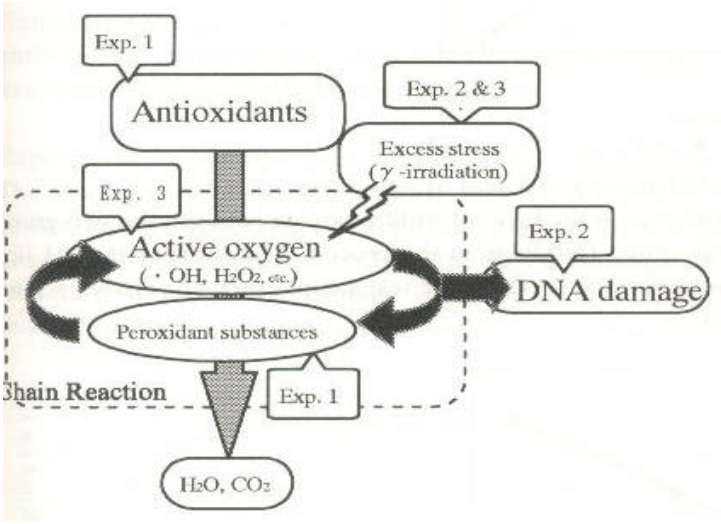
Reference

Antioxidant ability of EM-X: effect of EM-X on prevention of DNA oxidative damage

Sample name	IC_{50}	
	DNA	ESR
Epigallocatechin gallate	0.0015	
EM-X 1	0.007	0.26
EM-X 2	0.01	0.37

The number of following “EM-X” indicates the sample number.

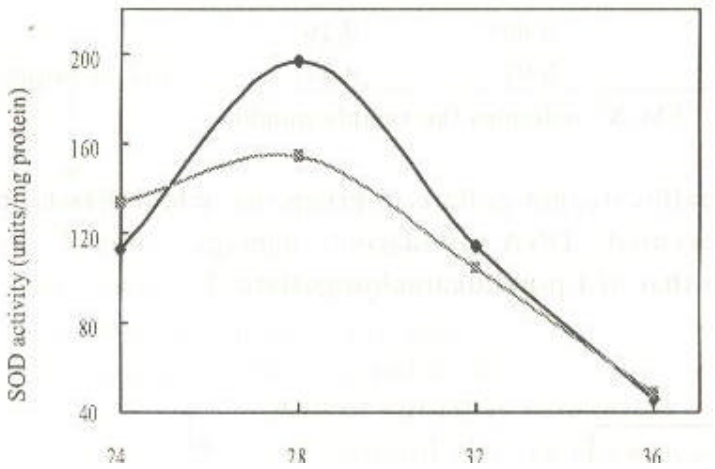
Result from comparison with Epigallocatechin gallate in green tea, which has high scavenging ability, EM-X prevented DNA oxidative damage caused by Flow char of Experiments:



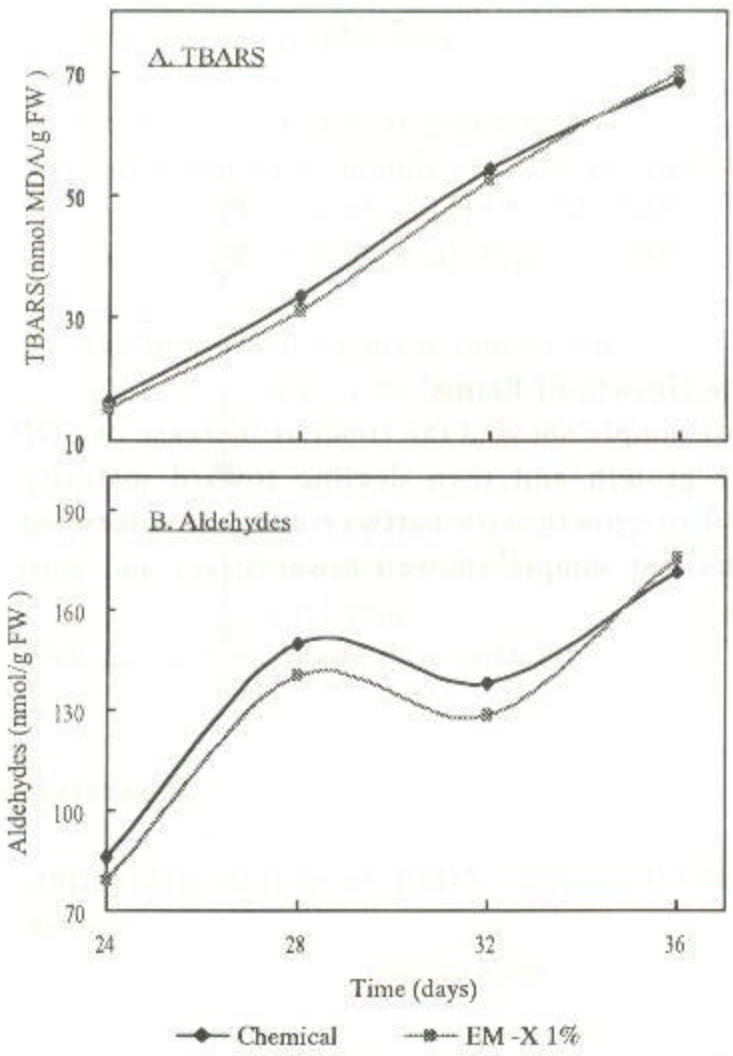
Results and Discussion

Experiment 1: "Effect of EM-X on the Growth of Plants"

1. Both the test sample and the control sample showed the trend of increase of SOD activity till the midway point of growth and then decline toward maturity, indicating it received stress related to growth with halfway and less afterward. Comparing the trends for both, the test sample showed lower stress and more stability than the control. (Fig. 1).



2. There was no statistical difference in the level of lipid peroxide between the two group nor in the level of Aldehyde group (products in the process of decomposition of lipid peroxide). On the whole, however, the level of r the test sample showed relatively lower than for the control. (Fig. 2)



3. Lettuce grown with EM-X, the test sample, showed the same level of Total ascorbic acid as one grown with chemical fertilizers. The former had a tendency to have a lower rate of oxidized ascorbic acid, hence a higher level of reduced ascorbic acid. (fig. 3)

EM-X effects on the growth of plant is summarized in the following:
Controlling the generation of oxides internally, allowing the plant to produce less antioxidant enzymes and antioxidants, and subsequently conserving nutrients and hence using more nutrients for its growth.

Experiment 2: "Comparison with the level of DNA damage from Acute Stress"
The availability of the test sample, that is, the effect of EM-X is expected to have controlled the generation of free radicals against ?-irradiation or eliminated it once generated and hence controlled the DNA damage. The test sample showed the tendency of less DNA damage with higher concentration level, while the control showed no such a tendency with higher concentration level. (Fig. 4)

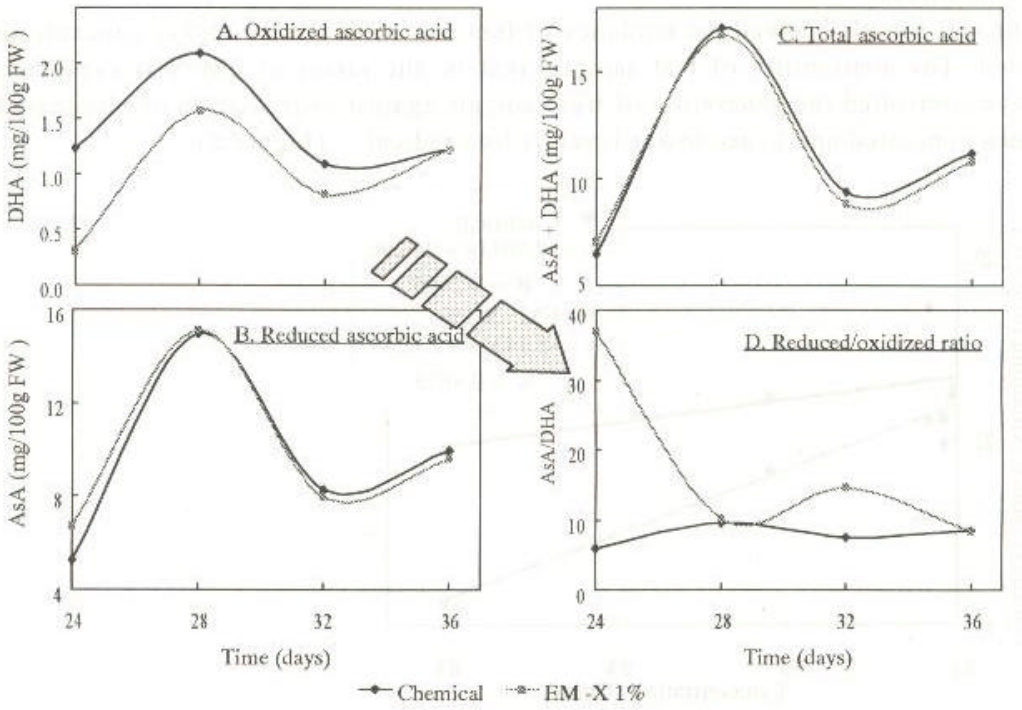
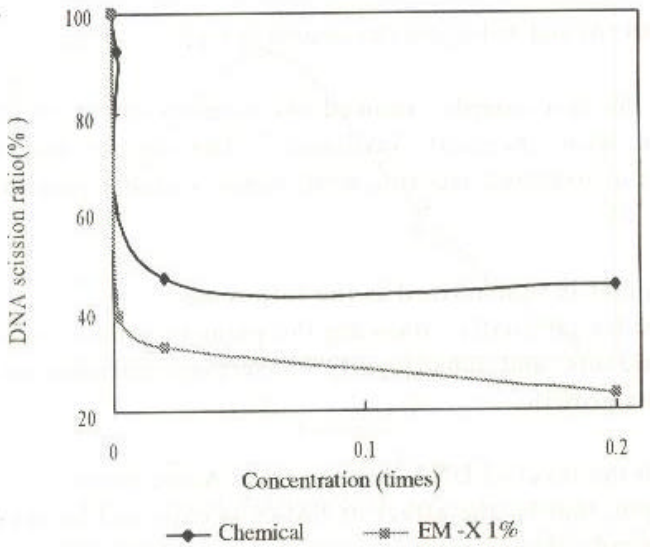


Fig. 3 Effect of EM-x 1% on oxidized ascorbic acid (A), reduced ascorbic acid (B), total ascorbic acid (C) and reduced/oxidized ratio (D) in leaves.



Experiment 3: "Comparison with the level of Hydroxyl Radical Generated from the Acute Stress"
The test sample showed the tendency of less free radical with higher concentration level. The availability of test sample, that is, the effect of EM-X is expected to have controlled the generation of free radicals against ?-irradiation or eliminated it once generated and hence lower level of free radical. (Figure 5)

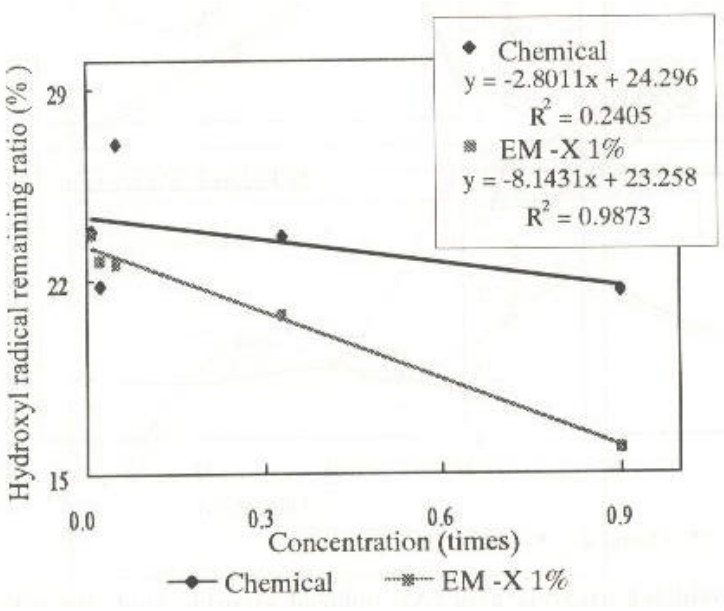


Fig. 5 Effect of EM-X on hydroxyl radical generation in leaf extract.

Conclusion:
Recapitulating the studies, the following are concluded and also identified as necessary to further confirm the mechanisms how EM-X functions as a highly powerful antioxidant:

1. EM-X, as it becomes available when grown with itself, controls the generation of free radicals in the plant through some kind of mechanism and subsequently the generation of oxides internally.
2. EM-X is highly capable to help the plant to grow stable under the controlled condition of nutrients availability of hydroponics, and to control the overall mechanism of maintaining free radicals low.
3. EM-x is considered to have a high level of antioxidant on the whole.