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Effects of Zinc Supplementation on Immunity and
Taste Acuity in the Elderly

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**Effects of Zinc Supplementation on Immunity and
Taste Acuity in the Elderly**

by

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Krithakorn Pratumvong

บทคัดย่อ

ผลของการเสริมสังกะสีต่อภูมิคุ้มกันโรคและการรับรสอาหารในผู้สูงอายุ

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บทคัดย่อ

การขาดเกลือแร่สังกะสีในผู้สูงอายุถูกนำไปสู่การเกิดการสูญเสียการรับรู้รสอาหาร การอยากอาหาร และทำให้ความต้านทานลดลงจึงติดเชื้อง่าย โดยเฉพาะช่วงการเปลี่ยนจากวัยผู้ใหญ่เป็นผู้สูงอายุ ในศึกษาวิจัยเชิงทดลองครั้งนี้มีวัตถุประสงค์เพื่อศึกษาผลของการเสริมสังกะสีต่อภูมิคุ้มกันโรคและการรับรู้รสอาหารในผู้สูงอายุ ได้ทำการศึกษาในประชากรผู้สูงอายุระหว่าง 60 – 79 ปี ทั้งชายและหญิง ที่เข้ารับการตรวจร่างกายประจำปีอย่างสม่ำเสมอที่ศูนย์วิจัย คณะแพทยศาสตร์ โรงพยาบาลรามาธิบดี ซึ่งจะทำการคัดเลือกผู้สูงอายุที่มีสุขภาพปกติแต่มีน้ำหนักตัวต่ำกว่าปกติ มีอาการเบื่ออาหาร จำนวน 20 คน จากประชากรดังกล่าว โดยทำการตรวจคัดกรองจากประวัติทางการแพทย์ และประวัติการบริโภคอาหารและสภาวะโภชนาการได้ทำการแบ่งผู้สูงอายุ ออกเป็น 2 กลุ่ม ตามกลุ่มอายุ คือ กลุ่มทดลอง 10 คน ให้ได้รับสังกะสีซัลเฟต 15 มิลลิกรัมต่อวัน และเปรียบเทียบกับกลุ่มควบคุม 10 คน (ไม่รับสังกะสีซัลเฟต) ระยะเวลา ก่อนและหลัง ให้สังกะสีซัลเฟตจะมีการควบคุมสภาวะโภชนาการทั้ง 2 กลุ่ม โดยให้สูตรอาหารปกติ (normal diet) ตลอดการทดลอง และได้ทำการตรวจผลทางชีวเคมีของเลือด (สภาวะสังกะสีและภูมิคุ้มกัน) ชั่งน้ำหนัก และทดสอบการรับรส ทุกเดือนเป็นเวลา 3 เดือน ผลการทดลอง พบว่า ผู้สูงอายุในกลุ่มทดลองมีน้ำหนัก จำนวนที-ลิมโฟไซท์ (lymphocyte) และสังกะสีในพลาสมาเพิ่มขึ้นอย่างมีนัยสำคัญ มีการรับรู้รสชาติของเกลือและน้ำตาลได้มากกว่ากลุ่มควบคุม แต่ไม่มีความแตกต่างระหว่างการรับรู้รสชาติของเกลือและน้ำตาลในกลุ่มทดลอง จะเห็นว่าการศึกษานี้บ่งชี้ว่า นอกจากการเสริมสังกะสีจะช่วยเพิ่มความต้านทานโรค รักษาสุขภาพ การรับรส และเพิ่มน้ำหนักตัวแล้ว ยังอาจช่วยสนับสนุน และ/หรือส่งเสริมสภาวะโภชนาการของสารอาหารชนิดอื่น ๆ ได้ด้วยเพราะสังกะสีช่วยปรับการรับรสให้ดีขึ้น จึงเพิ่มความอยากอาหารของผู้สูงอายุ ทำให้สามารถรับประทานอาหารได้เพิ่มขึ้นด้วย อย่างไรก็ตาม ผู้สูงอายุยังต้องการความรู้เพิ่มเติมในเรื่องการบริโภคอาหารที่มีธาตุสังกะสีที่เหมาะสมสำหรับผู้สูงอายุ จึงควรจัดการให้ความรู้ด้านโภชนาการและการดูแลสุขภาพผู้สูงอายุให้แก่ผู้สูงอายุและครอบครัวรวมทั้งจัดทำคู่มือการส่งเสริมการบริโภคธาตุสังกะสีจากอาหารที่มีอยู่ในท้องถิ่นเพื่อสามารถนำไปใช้ปฏิบัติได้ด้วยตนเอง

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Abstract

Effects of Zinc Supplementation on Immunity and Taste Acuity in the Elderly

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Deficiencies of Zn could lead to taste acuity, poor appetite and decreased resistance to infection. Inadequate zinc intakes are prevalent in the elderly especially during the transition period from adult to aging. The objective of this experimental study was to examine the efficacy of daily zinc on immunity and taste acuity of the elderly. A total of 20 apparently healthy elderly subjects age 60 - 79 years but underweight and loss of appetite were screened to provide standardization with normal diet for 3 days assessed in three months. Ten subjects received 15 mg of Zinc (Zn) and compared to ten controls matched for age and Zn status. Zinc concentration of plasma and taste detection for sucrose and sodium chloride, and immunity were assessed. Zinc treatment had influence on weight gain in both sex, the number of total circulating lymphocytes and could significantly improve immune function. The detection thresholds for sodium chloride and sucrose improved but not significantly among subjects receiving Zn supplementation. Our findings indicate that zinc supplementation did not improve only zinc status and some immune response but also their nutritional status from improved taste acuity. It was recommended from the study that the public health and relevant agencies should take into consideration on health and nutrition promotion agenda for the elderly and their families for better quality of life of elderly.

KEY WORD : Zinc supplement, Immunity, Taste acuity, Elderly.

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Contents

	Page
ACKNOWLEDGMENT	iii
ABSTRACT	iv
List of Tables	viii
List of Figures	ix
CHAPTER	
I. Introduction	1
1.1 Rationale and Justification	1
1.2 Objective	1
1.3 Hypothesis	2
1.4 Operational Definition	2
1.5 Applications	3
II. Literature Review	4
2.1 Zinc (Zn) nutriture in the elderly	4
2.2 The role of Zn : handing the body	5
2.3 Zn deficiency problems	7
2.4 Zn Homeostasis	9
2.5 RDA for Zn in elderly	10
2.6 Zn supplement, Usage and toxicity	13
III. Materials and Methods	17
3.1 Criteria of subjects	17
3.2 Experimental subjects	17
3.3 Dietary assessment	17
3.4 Biochemical and Immunological assessments	21
3.5 Taste acuity testing	22
3.6 Study area	23
3.7 Statistical analysis	23
IV. Result and Discussion	25
V. Conclusion	30

Contents (cont.)

	Page
Reference	31
Appendix	35

List of Tables

	Page
Table	
1 Recommended daily intakes for energy and Zn	11
2 Mean Daily energy and Zn intakes of elderly participants in national survey	11
3 Zn content of selected foods	12
4 Dose and Effects of Zn supplementation of elderly	15
5 Study design at pretreatment period (Baseline)	18
6 Study design at treatment period (Experiment)	19
7 Weight and blood sample analysis	25
8 Taste detection thresholds of sucrose and sodium chloride	27

List of Figures

	Page
Figure	
1 Taste detection thresholds of sucrose compares in both groups	28
2 Taste detection thresholds of NaCl compares in both groups	28

CHAPTER I

INTRODUCTION

1.1 Rational and Justification

The etiology of sub-optimal Zinc (Zn) nutriture in elderly persons has been related to changes in eating habits that may reduce both the amount and bio-availability of dietary Zn (Sandstead, et al. 1982). Age-associated changes in physiological function further altered by disease and concomitant use of medication may also place elderly individuals at greater risk of poor Zn status. The prevalence of poor Zn nutriture in older adults is unknown Zn status is difficult to assess (Solomons, N. W. 1979), no single index has adequate sensitivity and specificity. The concentration of Zn in serum is reported to decrease slightly with age (The Second National Health and Nutrition Survey. 1984), but circulating levels of Zn do not necessarily reflect tissue levels. The concentration of Zn in blood cells may provide an accurate index of whole - body Zn nutriture (Jone, et al. 1981). Leukocyte zinc concentration, for example decreased in adult men fed Zn restricted diets (Prasad, et al. 1978). The detrimental effects of long-term marginal intakes of zinc (Grejer, J. L. & Sciscoe, B. S. 1977), coupled with a decreased absorptive efficiency could severely compromise zinc status in older individuals. Although some studies have shown a moderate decline in plasma zinc with age (Lindeman, et al. 1971 ; Pilch, S. M., & Senti, F. R. 1984), many have detected little change (Flint, et al. 1981 ; Davies, et al. 1986 ; Vir, S. C. & Love, A. H. G. 1979) Yet it is commonly found that clinical symptoms associated with zinc deficiency such as poor wound healing, impaired immune response, and decreased taste acuity occur more frequently in elderly than in young subjects.

The purpose of this study was to broadly evaluate Zn status of healthy elderly persons, to determine relationships among Zn supplementation, immunity and taste acuity.

1.2 Objective

General Objective

To study effect of Zn supplements on immunity and taste acuity in the aged.

Specific Objective

1. To determine plasma Zn in elderly.
2. To determine immune parameters. : number of circulating T-lymphocytes.
3. To determine taste acuity in elderly.

1.3 Hypothesis

Elderly people who supplement Zn can increase plasma zinc level, increase number of T-lymphocyte, and improve taste acuity.

1.4 Operational Definition

Zinc (Zn) refers to capsule of zinc sulfate (Hausman Laboratories Inc., Smitzestand)

Supplementation refers to zinc sulfate supplemented with oral 15 mg/day for 3 months.

The elderly refers to healthy elderly which routine check up at Ramathibodi Hospital from 60 to 79 years in both sex but under weight and loss of appetite were screened to standardization with normal diet for 3 days in 3 months.

Nutritional status refers to Zn status (plasma Zn concentration) and weight.

Immunity refers to the number of T-cells.

Taste acuity refers to the taste detection for only 2 flavor modalities of sodium chloride and sucrose (salty and sweet)

Zn effects is the influence of Zn on improving nutritional status and immunity which were measured by

1. Changes in plasma Zn concentration.
2. Changes in the number of T-lymphocyte.
3. Weight gain.

1.5 Applications

1. To increase immunity in elderly.
2. To promote nutritional status from improve taste acuity.
3. To support psychological status.
4. To decrease mortality rate of the aged.

CHAPTER II

LITERATURES REVIEW

2.1 Zn nutriture in the elderly

One minor of potential interest in nutrition surveys of the aged is Zn. The typical diet is believed to contain 10 to 15 mg of Zn daily, which is 66% to 100% of the RDA (sub committee on the tenth edition of FDA, 1989). at risk of Zn deficiency in the elderly (Baler it alo, 1986). Elderly people very more in health status among themselves than do any other age group. Physiological age is much more important than chronological age in the elderly years. The common problem is taste smell immunity and slow wound healing (Wardeaw G.M. + Insel P.M., 1990 ; Calkins et al, 1986 ; Beisal W.R, 1992).

The number of taste buds present at age 30 declines by about one-third by age 70. In addition, the ability to discriminate flavors at low concentrations is lost. There is a loss in the number of taste buds, particularly those that detect sweet and salt (Luke . B, 1984 ; O' Hara Deyereaux et . al, 1981). The reduction in olfactory sensitivity may compound this reduced ability to enjoy food. When combined, impaired vision, hearing, taste, and smell may lead to a general apathy of older persons toward meals. (Milliken M.E. Campbell G, 1985) and may cause malnutrition in the aged.

It is likely that depressed immune function in elderly people is a causal factor contributing to the development of cancer and other diseases associated with aging (Weksler M.E, 1980). Malnutrition may increase the body's susceptibility and decrease its resistance to infection. Once an infection is established, the individual's previous state of nutrition is of great importance in determining the eventual outcome. Malnourished individuals are less able to withstand the toxic effects of infection, particularly on the heart, liver, and kidneys. The increased metabolism and break down of tissues cancer by infections diseases can have a more devastating effect in the malnourished individual , particular in elderly people (Hawrysh et al, 1987 ; Morly J.E., 1986).

In North America, Zn deficiency were first observed in the early 1970s in hospitalized patients on total parenteral nutrition. Zn was not added to solution prior to this time, but the protein source in the solutions was based on milk protein or blood fibrin, which are naturally rich in Zn. When the solutions were changed to include mostly synthetic amino acids as the protein source in the 1970s, deficiency symptoms quickly developed. This source of protein is very low in Zn content. (Baler et al., 1986)

2.2 The role of Zn : handing by the body

Zn absorption is subject to many different influences, several of which are currently under investigation. Absorption is facilitated by an as yet unidentified substance called zinc-binding ligand that appears to be secreted by the pancreas. Zn absorption is assumed to average about 40 percent but varies over a wide range. Pancreatic secretions release a considerable quantity of zinc into the intestine. This mixes with ingested Zn. If a person has a problem with mal-absorption, there is a potential losing more Zn in the stool than was ingested. Thus deficiency could develop relatively quickly. Absorbed zinc is carried in the blood bound to protein. It can be stored in the liver or distributed throughout the body. If a person's blood level of Zn is high, he excretes more Zn in the urine. (Dyer, et al. 1981) When Zn is absorbed into intestinal cells, it induces the synthesis of the protein metallothionein, if Zn is not transferred to the blood stream from the intestinal cells with in 2 to 5 days, it is sloughed along with the cell and excreted. Thus a "mucosal block" works against over absorption of Zn. However, if large doses of Zn are taken, it overrides the mucosal block. Luckily for over-consumers, Zn is also excreted via the pancreas to the intestinal tract. (Foster, et al. 1979 ; Lee, et al. 1989 ; Cousin & Hempe. 1990)

Over 200 enzymes require Zn as a cofactor for optimum activity. Adequate Zn intake is necessary to support many bodily functions, such as (Cousin & Hempe. 1990 ; Wegner. 1985 ; Prasad and Oberleas. 1976)

1. Zn and the immune response : Zn is essential for integrity of host defense mechanisms. This is supported by a large body of evidence from animal

studies and clinical observations. The cell-mediated or T-cell immunity system appears particularly vulnerable to Zn deficiency, but Zn has a role in the humoral aspects of immunity as well. The potential for Zn related immuno-deficiencies in the elderly should be recognized. It should not be assumed that immune dysfunction is simply a consequence of normal aging. Discrepancies among reports of immune function in the aged may be attributed to failure to consider underlying nutritional and health status.

2. Zn and wound healing : Since Zn is essential for normal tissue regeneration processes, poor Zn nutrition contributes to delay in wound healing. Where as studies focused on the elderly are limited, the possibility of retarded wound healing due to inadequate Zn status should be appreciated. For example, there is evidence that decubitus ulcers respond to Zn treatment in some patients. If at all possible, steps to ensure adequate Zn status should be taken prior to surgery or hospitalization, as well as a normal balanced diet emphasizing foods of high Zn density. Substantial Zn losses occur as the result of injury and increased catabolism. However, some of the Zn released from soft tissues during catabolism is also apparently available to support the healing process.

3. Role of Zn in taste, smell, and vision : The role of Zn in maintenance of the special senses (taste, smell and vision) is also of interest. Because these functions (like immune response and tissue regeneration) are complex, multiple factors may contribute to their impairment. Nevertheless, the essentiality of Zn to these processes should be appreciated. Despite early speculation, based on animal studies, that normal appetite regulation was Zn - dependent, numerous clinical studies of gustatory and olfactory functions have thus far suggested that impairment of these two functions in the elderly is unusually unrelated to Zn nutrition. The role of Zn in vision is perhaps less well - known. Strong evidence supports the development of Zn - responsive night blindness in clinical Zn deficiency states. Limited data also suggest a role for Zn in color vision, central macular vision, and optic nerve integrity. Visual impairments are observed in patients with untreated acrodermatitis enteropathica. The Zn - responsive dendrite disorder.

4. As a component of Zn - containing metalloenzymes : The most important function of Zn is as a constituent of metalloenzymes. Zn metalloenzymes are found virtually in every metabolic pathway. This disproportionate influence in relation to its absolute quantitative importance is probably related to the chemical properties of the element, particularly its capability of forming coordinate bonds with nitrogen and sulfur atoms of amino acids. (Wapir. 1990)

5. In the conformation of polysomes : The importance of Zn - binding fingerloop domains in DNA - binding proteins as regulators of gene expression has been recognized. The presence of Zn in these proteins is essential for site - specific binding to DNA and gene expression. The Zn ion apparently serves as a strut that stabilizes folding of the domain into a finger loop, which is then capable of site - specific binding loop proteins provide one of the fundamental mechanisms for regulating gene expression of many proteins. (FDA. 1989 ; Luke. 1984 ; Prasad. 1991)

2.3 Zn deficiency problems

The reported physical consequences of marginal zinc deficiencies include the following (Hambidge. 1977; Alic. 1984 ; Greer and Geissler. 1978)

1. Abnormal Sense of Taste and Poor Appetite.

Nobody understands exactly why, but zinc is somehow necessary for the normal operation of one's sense of taste and smell. People with too little zinc in their bodies frequently find that all their food tastes bad - like garbage, or tinfoil, or dirt. In other cases the food may simply seem to have no taste at all. This miserably distressing symptom can easily set up a vicious circle, for the person with such unpleasant taste perceptions will understandably eat less, and therefore become even more zinc-deficient, and soon have even worse symptoms, and so on. The zinc - deficient children in the Denver Study just mentioned were described as "picky eaters," with long catalogs of foods they refused to eat. It is hard to guess whether their poor eating habits caused their zinc deficiency or vice-versa, but both factors certainly tend to reinforce each other once the syndrome is established.

Along with dysgeusia, zinc - deficient individuals often have "dysomnia," or unpleasant distortions of their sense of smell. They may for no reason suddenly experience the smell of burning rubber, or body odor, or rotten fish, and be unable to get away from it. People with dysosmia are often categorized as "cranks" by the doctors they consult, because smelling imaginary odors, like hearing imaginary voices, is usually considered a symptom of mental illness. Sometimes a person with a normal sense of taste or smell will suddenly develop dysgeusia or dysosmia, or even lose the ability to taste or smell all together. This happens occasionally after a case of flu or a blow to the head, and sometimes for no obvious reason at all. In such cases, careful medical examination must be done to rule out such ominous causes as brain tumors. But when no explanation can be found, zinc therapy restores normal taste and smell in a surprisingly large number of cases.

2. Improper Wound Healing.

Zn is necessary for cell division because it is required by the enzymes that control the manufacture of new protein and new DNA. The body sends high concentrations of zinc to the healing edges of any kind of wound. When laboratory rats are made zinc - deficient and then given surgical incisions, their wounds heal more slowly and break open more easily than is the case in their adequately nourished littermates. In some studies on human patients, the evidence suggests that oral zinc therapy hastens the healing of some kinds of wounds. Along these same lines, it is interesting that people who have just undergone surgery or some other kind of serious injury often complain that the hospital food is tasteless or bad - tasting. Some experts in zinc nutrition suggest that these patients' zinc supply is being concentrated in their healing wounds, leaving too little for their taste buds.

3. Sexual Problems.

One common claim of nutritional pitchmen is that Zn tablets will improve one's sexual performance, and cure all sexual problems. This is almost entirely false but not quite. First, let us emphasize that the most common and troublesome sexual problems-impotence, premature ejaculation, and failure of

female orgasm are usually due to psychological rather than physical causes. If Zn tablets have any effect in these cases, it is mere or another good example of placebo action, whose power must not be underestimated. But having said that, we should also note that Zn deficiency does carry with it certain sexual consequences. That is, severe Zn deficiency not only dwarfs children, it prevents their normal sexual maturation. And mild zinc deficiency, as has been experimentally shown in American college students, lowers sperm counts. Moreover, Zn supplementation has been shown to improve sexual function in a group of men who were impotent as a complication of severe kidney disease. Of course, Zn-responsive impotence in kidney failure patients does not necessarily imply that anyone else's impotence would also respond to Zn therapy. Whether or not marginal dietary deficiencies of Zn have any effect on sexual function remains to be seen.

4. Miscellaneous Problems.

Zn deficiency has been implicated by some experimental evidence in several other human health problems. At this writing, the scientists involved in this work have not reached agreement about whether or not zinc deficiency is really an important cause of these ailments, but the unfolding debate will bear watching. Cancer, for instance, is believed by some researchers to be more likely in people whose bodies are Zn deficient. This may relate to the impairment of wound healing discussed earlier, for slow – healing tissues seem to have more chance of becoming cancerous. At the opposite extreme of the importance spectrum, acne has been found in some studies to improve when Zn is taken orally. Finally, there is considerable evidence from animal and human studies that Zn deficiency during pregnancy may increase the risk of deformities in the young. This finding is particularly troubling in light of the fact that the diets of pregnant American women are often disturbingly low in their Zn content.

2.4 Zn homeostasis

Individuals tend to be protected from Zn depletion through normal homeostatic Mechanisms. Human populations are apparently able to adapt to a

wide range of Zn intake from Plant and/or animal sources. Homeostatic adjustments occur in intestinal absorption, tissue distribution, and excretion (Luke B., 1984 ; Greger J.L. & Geissler, 1978).

The amount of Zn that needs to be absorbed daily in the healthy well-nourished adult is about 2.5 to 4.0 mg. Zn is excreted almost entirely through the gut. For example, on Zn intakes of 14.0 to 15.1 mg/day, average fecal Zn excretion is 11.7 to 13.7 mg and urinary output ranges from 0.6 to 0.7 mg. fecal Zn includes unabsorbed Zn, as well as Zn that is selected from the vascular space into the intestine.

Available evidence indicated that the intestinal cell is a major site for regulation of Zn homeostasis. Although dietary components such as phytate and fiber tend to reduce Zn bio-availability, overall Zn status appears to have a greater influence :

The first phase of Zn absorption - transport across the brush border membrane surface - is influenced primarily by composition of diet and by other factors in the intestinal lumen. Zn status is the major regulatory factor in subsequent phases of absorption. These include :

1. Distribution of Zn within the intestinal cell ;
2. Efflux of Zn to the plasma carrier, albumin ; and
3. The secretion of endogenous Zn back through the intestinal cell to the lumen.

In the process of redistribution among tissues, plasma Zn levels may be normal despite an overall deficient status. Normal mechanisms for Zn conservation may therefore complicate the diagnosis of Zn deficiency. (Cousins Hempe. 1990 ; Lee, et al., 1989)

2.5 RDA for Zn in Elderly

The adult RDA for Zn in elderly is shown in **table 1 – 2**

Table 1 Recommended daily intakes for energy and Zn*

Group	Age	Weight (kg)	Height (cm)	Energy needs (kcal)	Range	Zn (mg)
Men	51 – 75	70	178	2,400	(2,000 – 2,800)	15
	76 ⁺	70	178	2,050	(1,650 – 2,450)	15
Women	51 – 75	55	163	1,800	(1,400 – 2,200)	15
	76 ⁺	55	120	1,600	(1,200 – 2,000)	15

*Base on data from the food and Nutrition Board, National Research Council.
(Freedmen & Ahronhem. 1985)

Table 2 Mean Daily energy and Zn intakes of elderly participants in national survey*

Group	Age	Energy (kcal)	Zn (mg)	Zn density (mg/1,000 kcal)
Men	65 – 74	1,970	10.5	5.3
	75 ⁺	1,808	9.3	5.1
Women	65 – 74	1,444	7.6	5.3
	75 ⁺	1,367	7.0	5.1

*Based on data from Gupta et al. 1988.

From the investigation of Food and Nutrition Board, 1980 to determine normal serum Zn value in healthy Thai adults up to 60 years old found that serum Zn for age 51 - 60 years are 110.4, 18.9 (86.8 - 172.4) g/dL both male and female. (no statistical difference)

If a daily Zn supplement is taken, it should be at or near the current RDA of 15 mg. However, for the long term prevention of deficiencies of Zn (as well as other micronutrients), Attention to quality of chief is a better approach. (Sandstead et al., 1982 ; Panida, 1989 ; Kutti S. & Kietti J., 1986)

Table 3 Zn content of selected foods*

Food	Serving	size	Zn (mg)	Zn density (mg/1,000 kcal)
Oysters	3	oz	8.0	186
Crabmeat	3	oz	3.8	44.7
Beef liver	3	oz	4.3	21.5
Shrimp	3	oz	1.8	18.0
Chicken, dark meat	3	oz	2.4	16.4
Ground beef	3	oz	3.8	15.8
Ham	3	oz	3.4	13.6
Pork loin	3	oz	2.6	8.4
Tuna	3	oz	0.8	7.2
Milk, low-fat	1	cup	0.9	6.4
Eggs	2		1.0	6.3
Black – eyed peas	½	cup	1.5	15.8
Cooked oatmeal	1	cup	1.2	8.3
Cooked dried beans	½	cup	0.9	7.5

*Based on data from Sandstead et al., 1982.

Dietary Requirements and Sources

In general, protein – rich diets are also rich in Zn. The most nutrient – dense sources of Zn are oysters, shrimp, crab, beef, and mushrooms. (Table 3) As with iron, nutrient density is not the only issue : bio-availability is probably more important. Animal foods are the best Zn sources because Zn from animal sources

is neither bound by phytate, nor so affected by local soil conditions. Animal foods supply almost half Zn intake, but their cost makes Zn a very expensive nutrient. We shouldn't discount good plant sources of Zn, such as whole grains, peanuts, and beans, however, because studies show they can deliver substantial amounts of Zn to body cells. Zn is not part of the enrichment process, so refined flours are not a good source. Sea-foods are good, readily bio-available sources of Zn.

The same dietary factors that interfere with iron absorption seem to interfere with zinc absorption as well. Fiber and phytates pose a potentially serious problem. In addition, high intake of non-heme iron (as from iron-fortified foods or iron supplements taken with meals) can seriously reduce absorption of inorganic forms of zinc. Absorption of "organic" zinc from oyster is not reduced by excess iron. Human milk contains a zinc-binding ligand that greatly enhances absorption of the zinc contained in the milk. Wine apparently contains a nonalcoholic substance that promotes zinc absorption. Other dietary factors that significantly enhance zinc absorption are unknown.

2.6 Zn supplement, usages and toxicity

Self-prescribed Zn supplementation is common, due to recent widespread promotion in the popular media. In some people such supplementation can be expected to produce adverse side effects, such as copper (cu) deficiency anemia. Large doses of Zn may also irritate the gastrointestinal tract. Also, if a reduction in hemoglobin is observed, Zn supplementation should be considered as a possible cause.

When Zn deficiency is a clear risk, Zn supplementation is of course warranted. The compound most commonly used for supplementation is Zn sulfate. Daily supplements in the range of 15 to 40 mg will be safe and adequate for most patients. Although larges daily doses of 150 mg Zn (as 220 mg Zn sulfate) have been used without apparent harm, a Zn deficiency is easily corrected by much lower levels. (Wagner, 1985)

Traditionally, treatment of Zn deficiency has involved daily oral dosages of 150 mg of Zn in three divided doses. Given the risk of inducing Cu deficiency

of this regimen. Sandstead 1979 has recommended that therapeutic doses of oral Zn to treat Zn deficiency be limited to 40 mg/day. In some conditions, such as acrodermatitis enteropathica, lesser amounts of medicinal Zn will produce the desired therapeutic effect. (Sandstead et al. 1982)

A new class of conditions is being described in which oral Zn in "Therapeutic levels" is claimed to be effective even in the absence of preexisting Zn deficiency. These are so – called "Zn – responsive" conditions, with claims for daily doses in excess of 40 mg. There are claims for beneficial effect on immune responses by supplementing elderly subjects (> 60 years) with 150 mg of Zn/day. The long term benefits of this immuno reconstitution have not been evaluated in terms of its ultimate influence on carcinogenesis or infection susceptibility. Another "Zn – responsive" condition is to maintain "decuppered" state in Wilson's disease, after initial treatment with D-penicillamine, by high level for oral Zn. The amount of Zn required to suppress absorption of dietary and endogenous Cu is 150 mg daily.

When individuals are unable to consume nutrients orally, total parenteral nutrition (TPN) may be instituted. If TPN continued for more than a few days, Zn must be added to the regimen unless evidence for prior depletion indicates immediate parenteral therapy. (Panida. 1989 ; Kutti. 1986 ; Duchateau et al. 1981)

Another popular notion is that Zn can remedy the common cold, and people take Zn lozenges in hopes this is true. Although Zn does affect cold viruses in the laboratory, there is no convincing evidence that Zn is useful in treating colds. Some investigators have shown that taking large doses of Zn may even interfere with the body's natural immune response. There is currently no indication that Zn supplements have any value for people with AIDS and related syndromes. (Luke. 1984)

The experiments about Zn supplement in elderly are followed as sequences in **table 4**

Table 4 Dose and Effects of Zn supplementation of elderly

Investigators	Oral Dosage / time	Effect
Greger, J. L. and Geissler, A. H. 1978	15 mg/d for 95 days	Improved taste slightly
Duchateau J. et al. 1981	220 mg twice per day for 1 month	Increase antibody response clearly
Sandstead H. H. et al. 1982	10 mg/d for 56 days	Increase T – cell number, Improve wound healing, but no change in taste acuity
Bogden J. D. et al. 1987	15 up to 100 mg/d for 3 months	Increase only Immunocompetence in the 100 mg Zn while Zn 15 mg had no response
Swanson C. A. et al. 1988	30 mg/d for 28 days	Increase serum Zn concentration, but immunoglobulin and leukocyte did not change

Zn toxicity can result from or parenteral exposure, and can be either acute and chronic. The different forms produce different manifestation. (Panida. 1989 ; Hambidge. 1987 ; Grzegorezyk et al. 1979)

Acute toxicity : With oral Zn, doses above 25 mg can produce nausea, metallic taste, abdominal cramps, headache, dizziness, vomiting and chills. Doses in excess of 255 to 450 mg are frankly emetic. Several outbreaks of food poisoning have resulted from preparing fruit drinks in galvanized containers. Treatment of a premature infant with a daily dose of 3 mg of Zn per kg produced irritability, tremor, seizures, and tachycardia, and serum Zn rose to 224 mg/dl. Overly rapid infusion of Zn in TPN solutions can produce symptoms. Bos et al. (1977), observed transient flushing, blurred vision, and sweating when 10 mg of

Zn was perfused with in a brief period. A fatality resulted from a massive inadvertent infusion of 1.6 gm of Zn over a three - day period in a woman on heme dialysis.

Chronic toxicity : The major consequence of the chronic ingestion of Zn for medicinal purposes is Cu deficiency and anemia as a result of the intestinal interaction of Zn and Cu. These conditions developed rapidly in a patient given 5 gm of Zn daily. Long - term administration of a 150 mg daily dose produced the same result. Gastric erosion is another complication of daily dosage of Zn in the 150 mg range. A 10 - fold error in formulating of a TPN solution, providing 23 mg of Zn daily instead of the prescribed 2.3 mg, resulted in asymptomatic hyperamylasemia in 7 patients.

CHAPTER III

MATERIALS AND METHODS

3.1 Criteria of subjects

The study will be performed in 20 healthy elderly that loss of appetite and underweight from age 60 - 79 years old. The subjects will be chosen by screening from medical and nutritional histories and willingness to admit in the hospital to provide standardization with normal diet, donation of blood samples, and test for taste acuity. Each subject will be considered to be healthy on the basis of a clinical and biologic routine check-up.

3.2 Experimental design

The experiment is done in metabolic ward for 2 periods : pretreatment period (3 days) or baseline (**Table 5**) and treatment period (3 months) or experiment (**Table 6**). At the pretreatment period, blood sample for Zn status and immune function, taste acuity, and weight of all subjects will be measured at the first and third day of admission.

During 3 days period of admission, normal diet items will be prepared to all subjects.

At the treatment period the subjects will be divided into 2 groups (control and treatment), match for age and biochemical status of Zn. The treatment group will be supplemented with oral Zn 15 mg/d for 3 months. Both groups will received normal diet take home everyday in continuing through 3 months. The follow up of biochemical assessments for Zn status, immune function and taste acuity will performed every month.

3.3 Dietary assessment

Dietary assessment was carried out in each individual one day prior to the venipuncture by twenty-four hour dietary recall method. The data were then calculated for total daily caloric intake per capita and It percentage distribution

Table 6 Study design at treatment period (Experiment)

Experiment period																			
Date	Subject no.	Weight (kg)						Plasma Zn (μ mol/L)						T-lymphocyte (%)					
		Control			treatment			Control			treatment			Control			treatment		
		m.1	m.2	m.3	m.1	m.2	m.3	m.1	m.2	m.3	m.1	m.2	m.3	m.1	m.2	m.3	m.1	m.2	m.3

derived from fat, carbohydrate and protein. The computation was made by using Thai Food Consumption Table (Thai food composition table. 2000) and Food Consumption Table (Freedman & Ahronhem. 1985)

The menus served during the days before and 3 months during Zn supplementation period. They will be analyzed by food composition tables (Freedman & Ahronhem. 1985). They consist of 1,800 – 2,000 kcal (Protein 12 – 15%, Fat 30 – 35%, Carbohydrate 50 – 55% (Smith J. C. et. al. 1979).

3.4 Biochemical and Immunological assessments

Sample collection and analysis for :-

3.4.1 Direct measurement of plasma Zn concentration by Atomic Absorption Spectroscopy. The method is performed by Smith J. C., and Butrimovitz G. P. (Swanson. et. al. 1988).

3.4.2 Lymphocyte study :

Lymphocyte will be separated from 25 ml. of heparinized blood by density gradient centrifugation on Ficoll-Hpaque ; the cells will be washed three times in Hanks' solution without calcium and magnesium (Gibco-Biocult). The number of T-cells was determined by their ability to form Σ - rosettes (Σ - RFC) according to the method of Jondal, Holme and Wisell (Shils & Yiong. 1988)

(a) Rosette-forming T-lymphocytes :

Lymphocytes were washed twice with Hank's balance salt solution, then centrifuged at 200 g for 5 minutes and discarded the supernatant. 0.1 ml of heat-inactivated sheep red blood cell-absorbed AB serum was added to the pellet of lymphocytes and then added 0.1 ml of the 2% sheep red blood cell suspension in Hank's balance salt solution. The cells were gently mixed, incubated at 37°C for 5 minutes, sediment at 200 g for 5 minutes and then maintained at 4-5°C for 1 hour. The pellet was gently re-suspended and transferred to observed under oil immersion. Two hundred lymphocytes were counted and all cells binding more than two sheep red blood cells were tallied. The counting was done in three slides. The results were expressed as percent rosette-forming T-lymphocytes.

(b) PHA-induced lymphocyte transformation :

Lymphocytes were separated aseptically from other blood elements by a density gradient of Ficoll-Hypaque. Lymphocytes were washed twice with cold tissue culture media 199 (Difco). The concentration of lymphocytes in the final suspension was determined by counting in duplicate in a haemocytometer.

Lymphocytes were culture in tissue culture media 199, with L-glutamine (Grand Island Biological Co., New York) plus 20% autologous serum, penicillin (100 U per ml), and streptomycin (100 µg/ml) to a final concentration of 1×10^6 lymphocytes per ml. 1.5 ml of suspension were cultured in screw capped test tube at 37°C for 72 hours.

For stimulation, 0.05 ml PHA-P (Difco) was added to 2 tubes, with 0.05 ml 0.9% saline as control in another 2 tubes. The pH was checked daily and corrected if necessary with NaHCO₃ solution or with 0.1 N-HCl. After 72 hours, 0.1 µCi of tritiated thymidine was added to each tube. After incubation for an additional 18 hours, the cells were remove from the incubator, centrifuged, washed twice in ice-cold saline, then in ice-cold 2% acetic acid. The sediment was left to dry at 37°C. 0.5 ml hydroxide of hyamine 10x (Packard) was added to re-suspend the sediment. The suspension was left overnight in a dark cupboard. 0.5 ml absolute ethanol was then added, the tubes were shaken and kept in a 45°C water-bath for 3 hours. 5 ml scintillating liquid (Popop 0.3 g and Ppo 3 g per litre of toluene) were added and radioactive counted in a Nuclear Chicago liquid scintillation counter.

Results were expressed as (cpm = counts per minute) :

$$\text{Proliferative index} = \frac{\text{PHA cells cpm} - \text{background cpm}}{\text{Un-stimulated cells cpm} - \text{background cpm}}$$

3.5 Taste acuity testing

Taste detection thresholds will be determined using a staircase technique for increasing and decreasing stimulus concentration. A forced-choice design will be used to help minimize possible bias due to the age of the subjects. Water

rinses between the stimuli will be utilized to reduce the effects of possible age difference in saliva composition (Smith J. C. et al. 1979 ; Moore, et al. 1982)

To avoid taste fatigue, taste acuity for only two flavor modalities (salty and sweet) will be tasted. Taste acuity for salt is of special interest because McConnell and Henkin (Alic A. 1984) have noted an increase in sodium chloride preference in Zn deficient rats. Also Schechter et al have found that hypertension patients had an increased preference for sodium chloride. Subjects will be asked to differentiate between deionized water and three concentrations (12, 24, 48 m μ) of sodium chloride and sucrose in forced choice triangle tests (Prasad A. S. 1991 ; Hambidge R. M. 1977). The solutions within a test and the tests themselves will be presented in a random order to subjects. Deionized water was available for subjects to rinse their mouths between solutions.

No subject had eaten in the hour before the test. Previously, no time differences in taste acuity were noted when subjects participated in these tests in the mid-morning and mid-afternoon. Hence subjects will be tested between 9.30 to 11.30 and 2.00 to 4.00

The lowest level of sodium chloride and sucrose correctly identified as different from deionized water will be considered the subject's detection threshold for the compound. The lowest concentration of the compounds that the subject correctly identified as sweet or salty will be considered the subject's recognition threshold for the compound.

3.6 Study area :

Research center in Faculty of Medical, Ramathibodi Hospital, Mahidol University.

3.7 Statistical analysis

The results of plasma Zn, T-lymphocyte and weight gain will be presented as mean \pm SD. in both control and treatment groups, and the parameters will be compared by paired t - test.

The results of taste acuity test will be presented as percentages for treatment group.

The overall improvement in plasma Zn concentration, T-lymphocyte, weight gain, and taste acuity by Zn supplementation will be presented by repeated measurement.

CHAPTER IV

RESULT AND DISCUSSION

The number of subjects completing 3 months, treatment group were administered orally with 15 milligram per day of Zn supplementation. Plasma Zn concentration were measured at 1, 2 and 3 months after treatment, were increased to 16.5, 21 and 25 μ mol/L respectively. T lymphocytes significantly increased in the treated group ($p < 0.05$) as the same time. The weight was gain in both sex as shown in **table 7**

Table 7 Weight and blood sample analysis

Parameter	First month		Second month		Third month	
	Control	Treatment	Control	Treatment	Control	Treatment
	$\bar{X} \pm SD$	$\bar{X} \pm SD$	$\bar{X} \pm SD$	$\bar{X} \pm SD$	$\bar{X} \pm SD$	$\bar{X} \pm SD$
Plasma Zn (μ mol/L)	13.3 \pm 0.3	16.5 \pm 6	13.1 \pm 0.4	21 \pm 0.4	14.2 \pm 0.2	25 \pm 0.1
T-lymphocyte (Σ -RFC) (%)	53 \pm 2.9	61 \pm 1.2	53 \pm 0.1	76 \pm 0.5	54 \pm 0.4	88 \pm 0.4*
Weight gain (kg)						
Male	60 \pm 13	62 \pm 10	62 \pm 11	70 \pm 12	63 \pm 12	78 \pm 13
Female	53 \pm 11	58 \pm 12	54 \pm 13	59 \pm 18	60 \pm 8	67 \pm 11

*Significant difference $p < 0.05$

It is not known if elderly individuals are at increased risk of poor Zn status as a consequence of consuming diets that do not provide Zn intakes at RDA (Committee on Dictary Allswances. 1980). Our population of healthy elderly individual had plasma Zn concentration of 13 mol/1. In the Second National Health and Nutrition Examination Survey (NHANES II), more than 2000 blood samples were collected from elderly individual aged 65 – 74 years (The Second

National Health and Nutrition Examination Survey, 1984). In summary, elder adults consuming selected diets were not found to be in poor Zn nutriture.

The present results clearly show that in elderly the oral administration of Zn significantly increase the antibody response. Increasing in the number of circulating T – lymphocyte after Zn treatment might be explained either by a direct effect of Zn ion on the lymphocyte membrane or by a stimulation of thymus endocrine function. It is clear that most of the measures of immune function assessed after 3 month supplementation were strongly related to supplementation. Another study of the interaction between zinc and immune response did not include an assessment of zinc nutrition. Fifteen institutionalized, apparently healthy persons, ages 81 ± 5 yr were given 100 mg zinc daily for 1 month, while 15 control subjects, ages 79.6 ± 4.2 yr were given placebo. The group given zinc displayed an increased percentage of circulating T – lymphocytes : an increased frequency and magnitude of delayed hypersensitivity skin reactions to purified proteins ; and a greater IgG antibody response to tetanus toxoid. No difference in phytohemagglutinin or Concanavalin-A stimulated in vitro lymphocyte transformation was observed in contrast to another study in younger subjects (Duchateau, et al. 1981). Because the zinc nutriture of the elderly subjects was not evaluated, the relation of the changes observed to their prestudy zinc untriture is unclear. On the other hand, it seems evident that certain aspects of immune status may be beneficially influenced in elderly persons by pharmacological supplements of zinc.

Table 8 Taste detection thresholds of sucrose and sodium chloride

Level of chemical Detected (mM)	First month		Second month		Third month	
	Control (%)	Treatment (%)	Control (%)	Treatment (%)	Control (%)	Treatment (%)
Sucrose						
12	4	20	13	38	13	56
24	12	40	20	56	21	72
48	39	60	40	71	43	89
Unable to indentify any of 3 levels of sucrose	58	47	56	38	40	7
NaCl						
12	6	21	10	38	13	48
24	10	39	15	62	18	70
48	41	69	40	75	41	86
Unable to indentify any of 3 levels of NaCl	50	40	48	13	32	4

Figure 1 Taste detection thresholds of sucrose compares in both groups

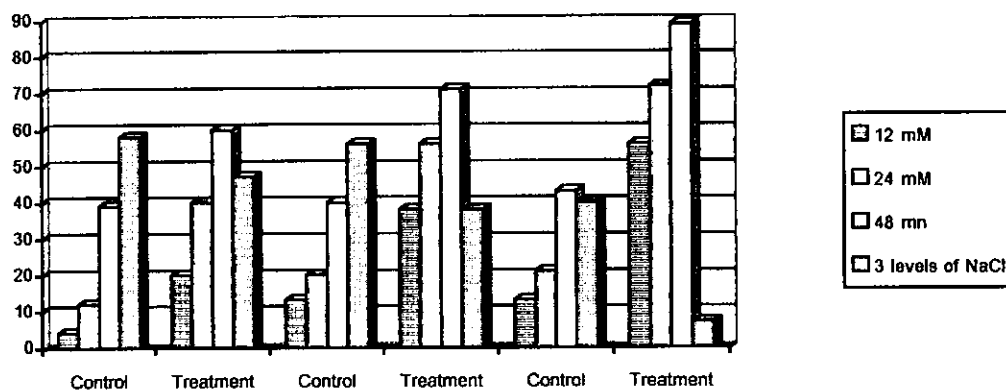
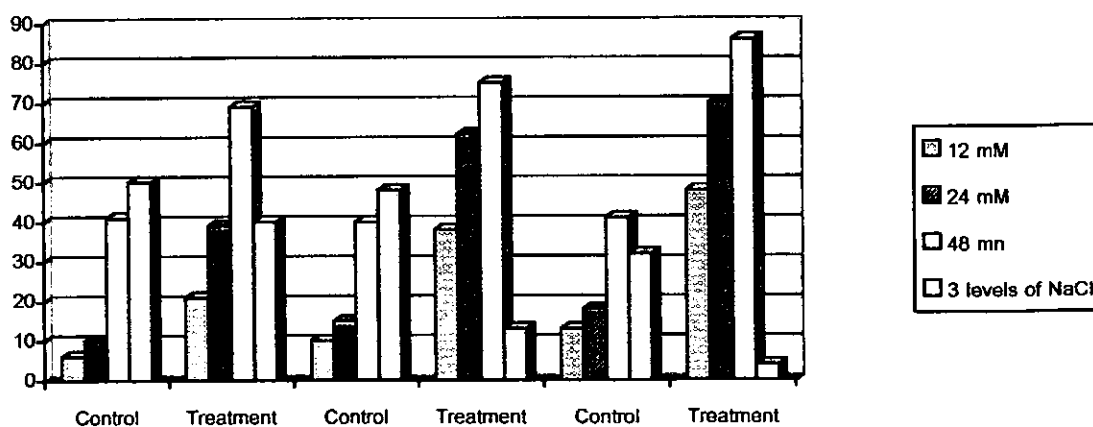


Figure 2 Taste detection thresholds of NaCl compares in both groups



Taste detection thresholds of sucrose and salt increase from first to Third month (**table 8, Figure 1, 2**). There was no significant difference in taste acuity between the treatment and control group. In the present study, significant relationships are not observed between fasting plasma levels of zinc or plasma

zinc response and detection thresholds for sodium chloride or sucrose in elderly subjects. Thus, it appears that the diminished taste acuity in our elderly subjects was in general not due to poor zinc status.

It must be recognized that the relatively small numbers of subjects in our study may contribute to the lack of statistical differences between groups. Four of the five subjects with fasting plasma zinc levels in the deficient range exhibit elevated detection thresholds. In fact, the plasma response curves are consistently lower in the high threshold groups as compared to those with low thresholds, and this trend is particularly evident in elderly subjects. It is unknown if this observation relates directly to the aging process. The possibility that significant relationships between plasma zinc response and taste acuity might have been observed in a larger group of similar subjects cannot be ruled out.

CHAPTER V

CONCLUSION

The adequacy of zinc (Zn) nutriture of older persons in the United States is the subject of recent and current research. (Kenneth, et al. 2001). In this we evaluate the current knowledge as it relates to the hypothesis that Zn deficiency is a significant health problem among the elderly. Particular attention is given to the content and bio-availability of Zn in diet of the elderly, laboratory evidence consistent with Zn deficiency, and the relationship of poor taste acuity and immune dysfunction in elderly persons to Zn nutriture. The typical American is believed to consume 10 to 15 mg of Zn daily. Among the aged, Zn intake is often less than two-thirds of the Recommended Dietary Allowance for Zn. However, Zn is not universally abundant in all foods. Generally animal proteins contain more bio-available Zn than cereal or vegetable products. Hence, among aged with low incomes and those residing in extended care facilities, which substitute vegetable products for animal proteins, the potential of low Zn intake is great. Thus, nutritional intervention to enhance immune function or prevent its decline could result a decrease in incidence and mortality of some diseases in elderly people. It was recently noted that Zn is the nutrient best characterized with respect to its ability to influence immune functions taste acuity and increase wound healing. From these properties of Zn it is interesting to study the effect of Zn Supplement in improving taste acuity and correct malnutrition in the aged which is leading to improve immune function and decrease mortality rate. It was recommended from the study that the public health and relevant agencies should take into consideration on health and nutrition promotion agenda for the elderly and their families for better quality of life of elderly.

REFERENCES

1. Sandstead H H, Henriksen L K, Greger J L. et al. (1982). ***Zinc nutriture in the elderly in relation to taste acuity, immune response, and wound healing.*** Am J Clin Nutr ; 36 : 1046-59.
2. Solomons N W. (1979). ***On the assessment of zinc and copper nutriture in man.*** Am J Clin Nutr ; 32 : 856-71.
3. Expert Scientific Working Group, Federation of American Societies for Experimental Biology. (1984). ***Assessment of the zinc nutritional status of the US population based on data collected in the Second National Health and Nutrition Examination Survey, 1976-1980.*** Bethesda, MD : LSRO.
4. Jones R B., Keeling P W N, Hiltion P J., Thompson R P H. (1981). ***The relationship between leukocyte and muscle zinc in health and disease.*** Clin Sci ; 60-237-9.
5. Prasad A S., Rabbani P., Abbasii A., Bowersox E., Fox M R S. (1978). ***Experimental zinc deficiency in humans.*** Ann Intern Med ; 89 : 483-90.
6. Greger J L., Sciscoe B S. (1977). ***Zinc nutriture of elderly participants in an urban feeding program.*** J Am diet Assoc ; 70 : 37-41.
7. Lindeman R D., Clark M L., Colmore J P. (1971). ***Influence of age and sex on plasma and red – cell zinc concentrations.*** J Gerontol ; 26 : 358-63.
8. Pilch S M., Senti F R. (1984). ***Assessment of the zinc nutritional status of the US population based on data collected in the Second National Health and Nutrition Examination Survey, 1976 – 1980.*** Rockville, M D : Fed Am Soc Exp Biol (Life Sciences Research Office).
9. Flint D M., Wahlqvist M L., Smith T J., Parish A E. (1981). ***Zinc and protein status in the elderly.*** J Hum Nutr ; 35 : 287-95.
10. Davies I J T., Musa M., Dormandy T L. (1968). ***Measurements of plasma zinc.*** J. Clin Pathol ; 21 : 359-65.
11. Vir S C. Love A H G. (1979). ***Zinc and copper status of the elderly.*** Am J Clin Nutr ; 32 : 1472 - 6.

12. Subcommittee on the tenth edition of FDA, National Research Council. (1989). ***Zin : Recommend dietary allowances.*** 10th revised edition, Washington DC : National Academic Press, 205-11.
13. Baler C W. Steinman L C., Freeland-Graves J H., et al. (1986). ***The effect of age on plasma Zinc uptake and taste acuity.*** Am J Clin Nutr, 44 : 664-9.
14. Wardlaw G M., and Insel P M. (1990). ***Perspective in nutrition.*** Boston : Mosby College Publishing, 439-45 ; 517-43.
15. Calkins E., Davis P J., and Ford A B. (1986). ***The practice of geriatrics.*** Hong Kong : W. B. Saunders Company.
16. Beisel W R. (1992). ***Single nutrients and immunity.*** Am J Clin Nutr ; 35 : 417-68.
17. Luke B. (1984). ***Principles of nutrition and diet therapy.*** Boston : Little, Brown and Company, 465-589.
18. O'Hard-Deyereaux M., Andrus L H., Scott C D., and Gray M I. (1981). ***Eldercare ; a practical guide to clinical geriatrics.*** New York : Grune & Stratton.
19. Milliken M E., Campbell G. (1985). ***Essential competencies for patient care.*** St. Louis : The C. V. Mosby Company.
20. Weksler M E. (1980). ***The immune system and the aging process in man.*** Proc. Soc Exp Biol Med, 165 : 200-5.
21. Hawrysh Z J., Donald E A., Basu K., et al. (1987). ***Age-related olfactory and taste changes and interrelationships between taste and nutrition.*** J Am diet Ass. 87 : 1543-50.
22. Morly J E. (1986). ***Nutritional status of the elderly.*** Am J Med, 81 : 679-95.
23. Swarson C A. Mansourian R., Dirren H., Rapin C H. (1988). ***Zinc Status of healthy elderly adults : response to supplementation.*** Am J Clin Nutr, 48 : 343-9.
24. Dyer A R., Stamler J. Oglesby P. et al. (1981). ***Serum cholesterol and risk of death from cancer and other cancers in three Chicago epidemiological studies.*** J Chronic Dis. 34 ; 249.

25. Foster Dm, Aamodt R T., Henkin R. I., Berman M. (1979). ***Zinc metabolism in human : a kinetic model.*** Am J Physio, 237 : R340-9.
26. Cousins R J. Hempe J M. (1990). ***Zinc.*** In present knowledge in nutrition. Brown M L editor. 6th ed., Washington DC : International Life Science Institute Nutritional Foundation, 251-60.
27. Lee H H., rasad A S., Brewer G J., Owyong C. (1989). ***Zinc absorption in human small intestine.*** Am J Physiol, 256 : G87-91.
28. Wagner P A. (1985). ***Zn nutriture in the elderly.*** Geriatrics, 40 : 111-125.
29. Prasad A S., Oberleas D. (1976). ***Trace elements in human health and disease.*** Vol I, Zinc and Copper. New York : Academic Press.
30. Wapnir R A. (1990). ***Protein nutrition and mineral absorption.*** Boston : CRC Press.
31. Prasad A S. (1991). ***Discovery of human Zinc deficiency and studies in an experimental human model.*** Am J Clin Nutr, 53 : 403-12.
32. R. M. Hambidge. (1977). ***The role of Zinc and other trace metals in pediatric nutrition.*** Pediat. Clin. N. Amer. 95.
33. Greger J L. Geissler A H. (1978). ***Effect of Zn supplementation on taste acuity of the aged.*** Am J Clin Nutr, 31 : 633-7.
34. Division of Nutrition Department. (2000). ***Thai food composition table.*** Public Health Ministry.
35. Freedman M L., Ahronhem J. C. (1985). ***Nutritional needs of the elderly : Dabate and recommendations.*** Geriatric, 40 : 45-62.
36. Gupta K L., Dworkin B., and Gambert S R. (1988). ***Common nutritional disorders in the elderly : Atypical manifestations.*** Geriatrics, 43 : 87-97.
37. Food and Nutrition Board. National Research Council. (1980). ***Recommended dietary allowances.*** 9th ed. Washington, DC : National academy of Sciences.
38. Panida Kittivaarawut. (1989). ***Zinc, Copper, and related enzyme levels in blood of Thai adults up to 60 years old.*** A thesis. Bangkok : Mahidol University.

39. Kutti S S., Kutti J. (1986). ***Zinc supplementation in anorexia nervosa.***
Am J Clin Nutr, 44 : 582-2. (a letter).
40. Duchateau J., Delespesse Guy, Vereecke P. (1981). ***Influence of oral zinc supplementation on the lymphocyte response to mitogens of normal subjects.*** Am J Clin Nutr, 34 : 88-93.
41. Shils M E., Young V R. (1988). ***Modern nutrition in health and disease.***
7th ed., Philadelphia : Len & Febiger.
42. Duchateau J., Delepesse G., Vrijens R., Collet H. (1981). ***Beneficial effects of oral zinc supplementation on the immune response of old people.***
Am J Med, 70 : 1001-4.
43. Bogden J D., et al. (1988). ***Zinc and immunocompetence in elderly people : effects of zinc supplementation for 3 months.*** Am J Clin Nutr, 48 : 655-63.
44. Swanson C A., Mansourian R., Dirren H., Rapin C H. (1988). ***Zinc status of healthy elderly adults : response to supplementation.*** Am J Clin Nut, 48 : 343-9.
45. Smith J C., et al. (1979). ***Direct measurement of Zn in plasma by Atomic Absorption Spectroscopy Clinical Chemistry.*** 25 : 1487-91.
46. Moor L M., Nielsen C R., Mistretta C M. (1982). ***Sucrose taste thresholds : age-related differences.*** J Gerontol, 64-9.
47. Grzegorexyk P B., Jones S W., Mistretta C M. (1979). ***Age-related differences in salt taste acuity.*** J Gerontol, 1134 : 834-40.
48. Committee on Dietary Allowances, Food and Nutrition Board, National Research Council. (1980). ***Recommended dietary allowances.*** 9th ed. Washington, DC : National Academy Press.
49. Kenneth, H. B., Sara, E. W. & Jan, M. P. (2001). ***The importance of zinc in human nutrition and estimation of the global prevalence of zinc deficiency.*** Food and Nutrition Bulletin, 22 ; 113-123.

Appendix

Medical history :

Sex Female male

Weight kg.

1. Education primary school
 high school
 Bach. degree

2. Occupation housework
 employes
 others

3. Smoking Yes No

4. Alchohol Yes No

5. Recent major personal problems

.....

6. Prior disease

.....

7. Current illness and medications

.....

Menu planning for 1 week

Date	Breakfast	Lunch	Dinner
1			
2			
3			
4			
5			
6			
7			

