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The Effects Of Consumption Of A Vitamin A Deficient Diet And Caffeine On Voluntary Ethanol Intake By Rats

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THE EFFECTS OF CONSUMPTION OF A VITAMIN A DEFICIENT DIET AND CAFFEINE ON
VOLUNTARY ETHANOL INTAKE BY RATS

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ABSTRACT

The research performed in conjunction with this thesis tests the correlation between consumption of a vitamin A deficient diet and voluntary alcohol intake by rats. It also tests the effects of caffeine intake on alcohol consumption by rats fed a vitamin A deficient diet.

Statistical evidence shows both a vitamin A deficient diet and caffeine addition to the diet increase voluntary alcohol consumption. The results are compared to a control group which received a balanced diet with available alcohol.
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INTRODUCTION

In 1953, it was estimated that 65% of the adult population in the United States consumed alcohol (Greenberg, 1953). By the 1980's, figures showed at least 65 million dollars per year were spent on medical care or consumed, due to lost production, as a result of alcoholism. In addition, 20% of all hospital beds, nationwide, were filled by people with an alcohol-related illness (Pechter, 1983).

Clearly, alcohol use and alcoholism is not a minor problem. It is a problem that is increasingly prevalent. A definite cause and cure are not known even though it is a problem of great magnitude. A current theory suggests the cause is concerned with vitamin deficiency.

Research has shown the tendency for alcohol consumption to increase with deficiencies in various vitamins and minerals. Recent studies also indicate caffeine may cause an increase in voluntary alcohol intake by rats.

The cure for alcoholism may involve vitamin and mineral supplementation along with consumption of a balanced, nutritious diet. Animal and human studies on vitamin therapy show success and great potential.

Caffeine consumption may also influence alcohol intake. Some researchers encourage the removal of caffeine from the diet in treatment of alcoholism.

This thesis is aimed at identifying the effects of both vitamin A deficiency and caffeine consumption on the voluntary ethanol intake by rats. The influence of dietary deficiency and caffeine consumption upon
body weights is also recorded and discussed. The effects and possible explanation of the results will be presented, based on the comparisons to the control group of rats.
Background

In order for the results of this experiment to be valuable and meaningful, it is important to know not only about the elements of the experiment, but also the laboratory animals used and their behaviors.

The behavior of a rat is governed largely by hunger. The quest for food occupies a significant portion of a rat's day. Rats consume food and liquids at approximately two hour intervals. The average adult drinks 15 - 30 ml of fluid a day and eats about 28 gm of dry food daily. Their vitamin requirements are like those of humans, with the exception of vitamins C and D. Rats can manufacture their own vitamin C. Rats require vitamin A. They are not able to convert carotenoids, from fruits and vegetables, to usable vitamin A.

Vitamin A

Vitamin A is needed by both humans and rats. Hopkins (1912) first discovered vitamin A. He reported a factor in milk which was necessary for proper growth in rats. McCollum and Simmonds (1977) identified fat-soluble A (the vitamin's first name) as the cause of xerophthalmia in rats.

Vitamin A aids in several physiological functions. These functions include growth, maintenance of skin and epithelia cells, gluconeogenesis, resistance to infection, mucopolysaccharide synthesis, maintenance of myelin and membranes, bone development, maintenance of color and peripheral
vision, and the maintenance of the adrenal cortex and steroid hormone synthesis. Vitamin A also aids detoxification in the liver (Kutsky, 1973).

Vitamin A deficiency causes xerophthalmia, nyctalopia (night blindness), hemeralopia (blindness in bright light), keratomalacia (softening of the cornea), and hyperketosis (excess formation of ketones). It also causes degeneration of columnar and cuboidal epithelia, abnormal bone growth because of a decrease in osteoblast and odontoblast activity, and cornification of epithelial structures (Kutsky, 1973). Vitamin A deficiency slows weight gain in rats.

**Caffeine**

Caffeine is 1, 3, 7-trimethylxanthine, a purine derivative. Caffeine is a central nervous system stimulant. It increases the heartbeat and basal metabolic rate. It promotes stomach acid secretion and urine output.

Caffeine functions by inhibiting phosphodiesterase. During emergency, the adrenal medulla secretes adrenalin into the blood. The adrenalin maintains the liver adenylate cyclase system which in turn maintains high concentrations of cyclic AMP in target cells. A high rate of glycogen breakdown occurs. After the emergency situation subsides, adrenalin is not secreted into the blood and the adenylate cyclase is inactivated. The cyclic AMP in cells is destroyed by phosphodiesterase, forming free 5'-adenylate. Protein kinases are inactivated and the glycogenolytic system is slowed to a resting state.

Phosphodiesterase is inhibited in the presence of caffeine. The cyclic AMP breakdown rate is slowed. Glycogen breakdown continues as if adrenalin was still present in the bloodstream.

Caffeine may also counter the effects in the brain produced by adenosine.
Adenosine, in the brain, depresses nerve cell firing, inhibits the release of neurotransmitters and binds to certain receptors.

Caffeine is structurally related to adenosine and competes with it for receptor sites because of its structural similarity. The caffeine attaches to receptor sites, preventing attachment by adenosine. The neurons fire more readily with caffeine in the receptor site. This explains the stimulatory effects of caffeine and its derivatives, ( Marx, 1981).

Caffeine may also affect alcohol consumption. Researchers found that feeding rats a regular diet with an addition of caffeine causes the rats to voluntarily consume more ethanol (Franklin, 1975). A similar study showed the same result. Caffeine consumption increased voluntary alcohol intake in laboratory animals (Frederick, 1982).

Dr. Guenther worked with alcoholic humans. He found alcoholics who ate a regular diet (supplemented with vitamins) and drank decaffeinated fluids were able to avoid alcohol. After six months, 81% of the alcoholics in this group had remained sober. Only 38% of the alcoholics consuming a regular diet but with no restriction on caffeine still avoided alcohol use (Pechter, 1983).

Alcoholism:

The use and abuse of alcohol has occurred for many centuries. Alcoholism has also been a favored topic of research by many scientists trying to find the cause and cure for alcoholism.

Effect:

Alcohol intoxication was first linked to a by-product of alcohol, acetaldehyde. Researchers William R. Klemm and Joseph Mikeska have recently
found that it is not the acetaldehyde but the ethanol that causes intoxication (Trotter, 1979).

Ethanol is oxidized to form acetaldehyde in the liver. It may affect neurotransmitters, such as catecholamines. Acetaldehyde may promote alcohol addiction, but it was not found to cause intoxication. Rats were infused with acetaldehyde to test the role of acetaldehyde in intoxication. These rats showed normal EEG's, after infusion, when concentrations of acetaldehyde were comparable to intoxication levels of rats infused with ethanol. The rats receiving ethanol in the same amount showed EEG's characteristic of intoxication (Trotter, 1979).

Ethanol addiction causes, in addition to intoxication, cirrhosis of the liver, asthma, diabetes, gout, neuritis, and stomach ulcers (Rosenberg, Feldzamen, 1974). Excess ethanol consumption decreases coronary flow because the heart pumps less efficiently (Flanagan, 1965). Ethanol consumption stimulates the flow of gastric juices and stomach mobility. This results in the sensation of hunger (Greenberg, 1953).

Other researchers have found alcohol consumption to cause the speed-up on intestinal transit time. Nutrients pass through the small intestine too fast to be absorbed and utilized. In the study volunteers consumed six to seven drinks a day for two weeks, while eating a nutritious diet. The digestive systems of the subjects were thrown into reverse within the two-week period. Their small intestines began secreting fluids and flushing the foods from their bodies before the nutrients could be absorbed and used (Mekhjian, 1979).

Researchers from the Permanente Foundation gave 150 cc. of weak ethanol (wine) solution, by mouth, to six men. They found that after
ethanol consumption, the men lost some ability to smell odors and taste sugar. The subjects experienced a decreased desire for food. Habitual drinkers also experienced this after consumption of more ethanol (Flanagan, 1951).

The loss of desire for food compounded with a lack of utilization of nutrients after excessive ethanol consumption makes vitamin deficiencies a prominent characteristic found with alcoholics. Either nutritious food is not consumed or the food is consumed but the vitamins and minerals are not absorbed properly. Deficiencies in zinc, calcium, iron, magnesium and vitamins B₆, B₁₂, C, A, and D are common.

Alcohol, in the form of hard liquor, does not contain any vitamins or minerals. Wines and brewed beverages retain some of the minerals and vitamins of the fruits or cereals they were produced from. Wines, brewed beverages and hard liquor all contain kilocalories. An ounce of whiskey contains approximately 75 kilocalories, the equivalent of 4½ teaspoons of sugar (Greenberg, 1953). Alcoholics may consume as many kilocalories as an individual consuming a balanced diet. However, the alcoholic consumes empty calories, calories with no nutritional value. Alcoholics are generally deficient in a multiple of vitamins and minerals for this reason.

Alcoholics may show any of the symptoms of vitamin deficiencies. They will have xerophthalmia, night blindness, hyperketosis and abnormal bone growth if deficient in vitamin A. Deficiency in the vitamin B's causes pellagra, beriberi, and pernicious anemia (B₁₂). Vitamin C deficiency results in scurvy. Vitamin D deficiency causes osteomalacia.
(deossification of bone) and osteoporosis (demineralization of bone). A deficiency in calcium causes these also. Deficiency in iron causes anemia. A zinc deficiency causes hypogeusia, the decrease in sharpness of the sense of taste (Sullivan, 1976). Zinc deficiency also affects the manufacture of a transport protein or its release in the liver. This protein transports vitamin A (Wright, 1981).

Cause:
The cause of alcoholism is unknown. However, several theories have been postulated. The theories are as follows:

1. Ethanol tolerance in rats is learned.
2. A genetic factor or defect causes alcoholism.
3. Drugs cause alcoholism, and
4. Vitamin deficiency promotes alcohol consumption.

Researchers proposed rats' consumption and tolerance for ethanol is learned. Rats learned to physiologically cope with ethanol after exposure to alcohol and learning. Rats trained to perform functions while intoxicated became adjusted to ethanol consumption (Wenger, et al, 1981).

Rats with a demonstrated alcohol preference were injected with large amounts of ethanol in a separate study. Researchers expected the voluntary decrease in oral consumption. They did not expect consumption to gradually increase. It did because of an increase in physical tolerance (Barry and Perbach, 1971).

Alcoholism has also been attributed to hereditary factors. Albino rats with a certain genetic background maintained a voluntary intake of ethanol. The intake rate was at a constant rate in terms of grams of
ethanol per kilogram of body weight. This constant rate was maintained with wide variations in the concentration of alcohol consumed (Barry III, Perbach Jr., 1971).

The effect of heredity on alcoholism was tested in humans by testing men who had an alcoholic family member. The blood alcohol levels obtained from these men before and after alcohol consumption was compared to those levels from control subjects. All people tested received .5 ml of ethanol per kilogram of body weight. Researchers found blood alcohol levels were significantly higher with men who had an alcoholic family member. A genetic factor may predispose carriers to intoxication or dependence on ethanol (Trotter, 1979).

The genetic factor is believed to cause a defect in how ethanol is metabolized. The ethanol is metabolized into a highly addictive, morphine-like substance, temahydioisoquinolone (THIQ) (Pechter, 1983).

Research based on the effect of drugs on alcoholism has generally shown which drugs decrease voluntary ethanol intake by laboratory animals. Drugs which inhibit ADH, such as n-butyl-aldozimo, metronidazol and disulfiram, decreased voluntary alcohol intake. Nialamide, which blocks the inactivation of catecholamines and serotonin, caused a decrease in alcohol consumption. A glutamine antagonist, methionine sulphoximide, caused an increase in ethanol consumption (Korsander, Erikson, 1971). Central nervous system active drugs have been found to have no effect on alcoholism. However, caffeine, a central nervous system stimulant, was believed to increase voluntary ethanol intake (Fowler, 1975).

Various experiments have been conducted testing the idea that food
deprivation and/or vitamin deficiencies promote alcoholism. Forced food deprivation caused rats to consume more ethanol. Total fasting or food available for only twelve hours a day both caused the increase. (Rats normally eat every two hours.)

No change in ethanol intake was found when a normal diet was changed to a high carbohydrate diet (81% carbohydrates). Rats drank more alcohol when their diet consisted of 46 or 63% carbohydrates. Fat rich diets (39% fat) were shown to decrease ethanol intake. In mice, a transition from a normal diet to one high in protein (60%) and low in carbohydrates (18%) resulted in an increase in voluntary alcohol intake (Israel, Mardones, 1971).

Researchers tested the importance of calorie and nutrient content on alcohol consumption by producing pellets with various nutrient compositions. The nutritive contents of pellets were reduced to 75, 50, 25 and finally 0% of the normal. The original shape of the pellet was retained. Water intake dropped as the nutritive values dropped. Voluntary ethanol consumption occurred in place of the water consumption. The rats maximized the calories by drinking alcohol with approximately seven kilocalories per gram when the food contained fewer calories. Rats consumed increasingly more ethanol to compensate for loss of calories through food over a 24 hour food deprivation period (Fester, Freed, 1971). Water consumption was nearly non-existent when non-nutritive pellets were present.

Charles River rats were reduced to 80% of their normal free-feeding weights. The rats were placed in specialized feeding chambers. Food
pellets were released at 120 second intervals, unless the rats consumed ethanol. A food pellet was released after 15 seconds if the rat in the chamber consumed alcohol. If the rats chose to drink water, they were allowed a maximum of eight times more pellets than if they consumed alcohol. It was expected that the rats would choose to consume water and receive more food. However, alcohol consumption continued. The rats maximized their caloric intake. They received 14.8 calories per session with alcohol consumption and 12.3 calories per session with water consumption (Lester, Freed, 1971).

Rats were tested using a .1% saccharine solution. This solution was chosen almost exclusively over water by the rats. The taste of saccharin and caloric content of ethanol are favored to maximize caloric consumption. Up to 40% of the calories lost from the non-nutritive food pellets could be consumed through ethanol intake.

A multitude of studies have been conducted on the effects of vitamin deficiencies on the tendency for rats to become alcoholic. Rats fed refined diets preferred to drink alcohol over water (Fowler, 1975). Many studies have focused on the effects of vitamin B complex deficiencies. Diets deficient in niacin created an increase in voluntary ethanol intake by LABw mice. LABw mice usually show a low ethanol preference. Researchers removed B vitamins (thiamine, riboflavin, pantothenic acid) from the diets of rats. The voluntary ethanol consumption increased. The researchers added thiamin and other B vitamins but alcohol consumption did not return to the lower levels recorded with a vitamin B supplemented diet (Mardones, Israel, 1971).
Researchers also performed a long-term study on vitamin deficiency and alcoholism. Rats were made deficient in thiamine, riboflavin, pantothenic acid and pyroxidine. Ethanol consumption increased 5, 10 and even 20 times the normal. The rats were supplied with the vitamins they were deficient in and alcohol consumption returned to the level of non-deficiency. An eight month test was conducted on vitamin-deficient rats. Their voluntary alcohol consumption increased 30% over consumption levels while not vitamin deficient. A single dose of vitamins was administered and consumption levels returned to normal. Researchers then allowed the rats to become gradually deficient again. Consumption of ethanol increased. A single vitamin dose was again administered. Ethanol consumption returned to normal. Researchers fed rats a marginal diet for sixty days. Ethanol consumption increased. Several vitamin doses were given instead of a single dose. Drinking levels became the lowest recorded and remained at those levels (Williams, 1978).

In a separate study, researches fed rats purified diets, lacking B vitamins. The rats voluntarily consumed more ethanol. They also lost weight. The rats gained weight but did not decrease ethanol consumption after adding thiamine to their diet. Researchers theorized that a factor, N, from yeast, affected alcohol consumption. Rats fed a supplement containing autoclaved yeast did not increase alcohol intake. These researchers believed it was the factor N that affected ethanol intake (Israel, Mardones, 1971).

**Therapy:**

Vitamin therapy for alcoholism treatment and cure is being used as a result of the evidence obtained from previous studies on the cause
of alcoholism. Many drugs have been tested for their ability to cure alcoholism, in addition to vitamin therapy.

A drug, antabuse, was used to inhibit oxidation of acetaldehyde. Antabuse was harmless as long as the alcoholic did not drink alcohol. The drinker experienced unpleasant symptoms with consumption of ethanol (Greenberg, 1953).

Some researchers have used estrogen and synthetic progesterone to treat alcoholic rats. The rats' appetites for alcohol decreased after treatment with these hormones (Yillsman, 1981).

Alcoholism is now being treated with vitamin therapy. Researchers found that people addicted to cocaine, heroine, and morphine could be cured by supplementing their nutritionally deficient diets. These subjects were given sodium ascorbate, protein, vitamins and minerals with a subsequent relief of their addictions (Frederick, 1982).

Similar treatment was used on alcoholics. Alcoholics were given specific diets with vitamin supplements. They were also given decaffeinated fluids, in some cases, to fight alcoholism (Rosenberg, Feldzamin, 1974). White flour was restricted since it lacks many vitamins. Glutamine was used to cut the ethanol craving experienced by alcoholics (Pechter, 1983).
MATERIALS AND METHODS

MATERIALS

Laboratory Animals:

I purchased six Chinese Hooded rats. The six rats were litter mates. They were six weeks old at the start of the experiment. From birth to six weeks they had been fed Hamster, Gerbil and Mouse Food, packaged by Eckholt's Feed and Grain. *1

Food:

The non-vitamin deficient group, Group 3, continued to be fed Hamster, Gerbil and Mouse Food, packaged by Eckholt's Feed and Grain. Group 3 also received a commercial vitamin supplement made by Hartz. *2

The vitamin-deficient groups, Groups 1 and 2, received commercially baked bread. *3 It is deficient in vitamins A and C, according to the manufacturers label.

Caffeine:

I purchased caffeine, 1, 3, 7-trimethylxanthine, in the form of a commercial stimulant, NoDoz. NoDoz contains 100% caffeine.

Fluids:

I provided a 3.13% ethanol solution, by volume, to all groups. Alcohol was diluted with distilled water.

Each group was also provided with a drinking bottle containing only distilled water, at room temperature.

During the Week 4-5 period, Group 2 received an ethanol-caffeine solution. The solution consisted of a 3.13% ethanol solution with 5 mg of caffeine added and dissolved in 8 oz. of solution.

*1 The composition of this food is shown in Table 1, of the Appendix.
*2 The composition of this vitamin supplement is shown in Table 2, of the Appendix.
*3 The composition of this bread is shown in Table 3, of the Appendix.
ENVIRONMENT

The environment for each group (2 animals) consisted of a wire cage, 12 inches wide, 18 inches long and 14 inches at its highest point. Each cage was equipped with one food dispenser and two drinking bottles. I placed the bottles on opposite walls. I placed the food dispenser an equal distance from both drinking bottles. Each drinking bottle was approximately 5 cm from the floor of the cage.

Environmental conditions included twelve hours of daylight, an average temperature of 68°F and a moderate noise level.

METHODS

Preliminary

I used five non-toxic food colorings to mark the backs of five animals. I left one white. All animals were then distinguishable.

I weighed each animal in grams. I then designated pairs of animals by making the pairs as equal in weight as possible.

Week 1:

Each pair of rats was housed in a separate cage. Abundant food (Eckholt's Feed and Grain) and water were available. Water was dispensed in both drinking bottles. Food consumption was weighed and recorded in grams on a daily basis. Water consumption was measured in ounces daily. I converted the ounce readings to milliliters. Each group of rats received 80 gm of food pellets and 8 ounces of water in each drinking bottle daily.
Week 2 and 3:

Groups 1 and 2 were designated as those to be fed a vitamin deficient diet. Group 3 was designated as the group to receive a vitamin supplemented diet.

Groups 1 and 2 were fed commercially baked bread, deficient in vitamins A and C. Each group was fed 50 grams of bread daily.

Group 3 continued to be fed 80 grams of food pellets each day. This group was also given 10 vitamin pellets per day.

Remaining food from the previous 24-hour period for each of the three groups was weighed and subtracted from the starting weight of food. This was used to determine the amount of food each group consumed daily.

One fluid dispenser in each group’s cage remained filled entirely with distilled water. The other dispenser in each cage was filled with an alcohol-water solution. The solution was composed of .5 ounces of ethanol in 7.5 ounces of distilled water.

I recorded water and alcohol/water consumption each day for each group. I reversed the position of the water and alcohol/water dispensers at the beginning of Week 3. I did this to prevent water or alcohol/water drinking by habit.

All six animals were weighed individually one time each week. I did not reassign groups to account for weight differences between groups.

Weeks 4 and 5:

Groups 1 and 2 remained on a vitamin deficient diet. Group 3 remained on a food pellet and vitamin supplemented diet. Food consumption was determined by weighing the food remaining after the 24-hour period.
and subtracting that amount from the starting weight. I recorded
daily food consumption for each group.

Groups 1 and 3 received the same fluids as in Weeks 2 and 3. Each
group received a dispenser containing only water. Groups 1 and 3 also
received a dispenser containing a solution of water and alcohol. The
alcohol/water solution was made from .5 ounces of ethanol plus 7.5
ounces of distilled water. The alcohol was 100 proof.

I designated Group 2 as the group to receive caffeine. I provided
Group 2 with one fluid dispenser filled only with distilled water. I
added approximately 10 mg of caffeine (1, 3, 7 - trimethylxanthine) to
fluid dispenser containing alcohol. The dispenser then contained .5 ml of
ethanol, 7.5 of distilled water plus 10 mg of 1, 3, 7 - trimethylxanthine.

I made daily records of fluid consumption for all three groups from
each of the fluid dispensers. Readings were taken in ounces and converted
to milliliters.

The six animals were weighed individually one time each week. The
weights were recorded in grams. Groups were not reassigned to compensate
for weight changes.
RESULTS

Average Water Intake

<table>
<thead>
<tr>
<th>Week</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>28.93</td>
<td>31.04</td>
<td>30.83</td>
</tr>
<tr>
<td>2-3</td>
<td>24.34</td>
<td>21.38</td>
<td>25.34</td>
</tr>
<tr>
<td>4-5</td>
<td>28.65</td>
<td>28.54</td>
<td>28.77</td>
</tr>
</tbody>
</table>

Week 1

All groups consumed between 26 and 31 ml of distilled water per day. The variations between the groups is due to randomness.

Weeks 2 and 3

Group 1 (vitamin deficient diet) and Group 3 (control) consumed between 24.34 and 25.34 ml of water, on the average, per day. Group 2 (vitamin deficient diet) consumed an average of 21.38 ml of distilled water per day. Using the t-test, I found the difference (almost 4 ml) in distilled water consumption has only a 20% probability of being due to chance error or randomness.

Weeks 4 and 5

All groups demonstrated a nearly equal average daily water intake per day. All groups consumed between 28 and 29 ml of distilled water in a 24-hour period.
### Average Alcohol Intake

**TABLE 2**

AVERAGE DAILY ALCOHOL INTAKE (ml/24 hours)

<table>
<thead>
<tr>
<th>Week</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-3</td>
<td>10.10</td>
<td>10.29</td>
<td>3.81</td>
</tr>
<tr>
<td>4-5</td>
<td>4.90</td>
<td>8.64</td>
<td>1.71</td>
</tr>
</tbody>
</table>

**Weeks 2 and 3**

Groups 1 and 2 (vitamin deficient diets) both consumed more ethanol per day, as compared to Group 3. The difference is shown to be statistically significant with the use of the t-test. There is less than a 5% chance that the increase in alcohol consumption by Groups 1 and 2 over Group 3 is due to randomness.

**Weeks 4 and 5**

Groups 1 and 2 again show a statistically significant high rate of alcohol consumption as compared to Group 3. There is a 99% probability that the differences recorded are due not to chance but to an actual variation. Group 2 (vitamin deficient diet and caffeine consumption) consumed more ethanol than either Group 1 or 2. The increase in voluntary ethanol consumption by Group 2 over Group 1 is also statistically significant.

All three groups decreased their total voluntary alcohol intake over a 24-hour period. The most substantial decreases occurred with Groups 1 and 3.
### Average Food Intake

#### TABLE 3

**AVERAGE DAILY FOOD INTAKE (gm/24 hours)**

<table>
<thead>
<tr>
<th>Week</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>38.1</td>
<td>39.9</td>
<td>38.1</td>
</tr>
<tr>
<td>2-3</td>
<td>37.2</td>
<td>38.6</td>
<td>38.5</td>
</tr>
<tr>
<td>4-5</td>
<td>42.3</td>
<td>41.9</td>
<td>39.5</td>
</tr>
</tbody>
</table>

**Week 1**

All three groups consumed approximately the same total weight of food. Group 2 consumed, on the average, 1.8 grams more food per day. This difference has a 75% probability of being due to randomness (t-test).

**Weeks 2 and 3**

Groups 2 and 3 showed similar food consumption averages (per day). Group 1 consumed less food, in grams, per day on the average than did Groups 2 or 3.

**Weeks 4 and 5**

The groups again showed a variation in the average amount of food consumed daily. Both Groups 1 and 2 increased food consumption. The increase is statistically significant, since the chance that this increase is due to randomness is only 10 to 15%, according to the t-test.
### TABLE 4

#### WEIGHT GAIN AND PERCENT INCREASE IN BODY WEIGHT

<table>
<thead>
<tr>
<th>Group</th>
<th>Week</th>
<th>Weight Gain (gm)</th>
<th>Percent Increase in Body Weight</th>
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<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>15.0</td>
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<tr>
<td></td>
<td>2</td>
<td>13.0</td>
<td>9.2</td>
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<td></td>
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<td>5</td>
<td>4.5</td>
<td>3.0</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>41.0 gm</td>
<td>31.8%</td>
</tr>
<tr>
<td></td>
<td>Average</td>
<td>8.2 gm</td>
<td>6.3%</td>
</tr>
<tr>
<td>3</td>
<td>1</td>
<td>20.5</td>
<td>18.6%</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>18.5</td>
<td>14.1</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>12.5</td>
<td>8.4</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>18.0</td>
<td>11.1</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>18.0</td>
<td>10.0</td>
</tr>
<tr>
<td></td>
<td>Total</td>
<td>87.5 gm</td>
<td>62.2%</td>
</tr>
<tr>
<td></td>
<td>Average</td>
<td>17.5 gm</td>
<td>12.5%</td>
</tr>
</tbody>
</table>

All rats consistently gained weight during all weeks of the experiment. Groups 1 and 2 animals, however, gained less weight than the rats of Group 3. Weight gain decreased in rats of Groups 1 and 2. Weight gain is fairly consistent for the rats of Group 3.
This graph shows the average daily water intake per group in milliliters and the change in consumption each week. All groups show an increase in water consumption after Week 2.

Group 1
Group 2
Group 3
This graph shows the average daily alcohol intake per group and the change in average consumption each week. Group 3 is a control group which received a non-vitamin deficient diet. Groups 1 and 2 received a vitamin deficient diet (deficient in vitamins A and C). During Weeks 4 and 5, Groups 2 received caffeine, also.

Group 1

Group 2

Group 3
Graph 3

WATER AND ALCOHOL CONTRIBUTIONS

to

AVERAGE TOTAL DAILY FLUID INTAKE PER GROUP

This graph shows the intake of water and alcohol and the contribution each of these makes to the total average daily fluid intake for each group each week.

Average Daily Water Intake Per Group (in ml.)

Average Daily Alcohol Intake Per Group (in ml.)

Groups: 1 2 3
Week: 1 2 3 4 5
This graph shows the average daily food intake per group. All groups increase their food consumption during the experiment. In comparison, all groups consume approximately the same amount of food daily.

Group 1
Group 2
Group 3
This graph shows the combined total weight of each group per week. It also shows the extent of the weekly weight gain for each group throughout the experiment.
**GRAPH 6**

**COMPARISON OF ALCOHOL AND FOOD INTAKES**

to

**WEIGHT GAIN**

This graph shows the correlation between consumption of a vitamin A deficient diet and alcohol and weekly weight gain of each group.

- Average Daily Food Intake per Group (in grams)
- Total Weekly Weight Gain per Group (in grams)
- Average Daily Alcohol Intake per Group (in ml)
DISCUSSION AND CONCLUSIONS

The purpose of this experiment is to test the effects of a vitamin A deficient diet and caffeine consumption on voluntary ethanol intake by rats.

The results from Table 2* show an increase in voluntary alcohol consumption when rats are fed a diet lacking vitamin A. The consumption of alcohol by Groups 1 and 2 is consistently higher than that recorded for Group 3.

Alcohol consumption by Groups 1 and 3 decrease during Weeks 4 and 5. Group 1 increases food intake during this period to maximize calorie consumption.

Group 2 increases ethanol consumption during Weeks 4 and 5, presumably because I added caffeine to their diet.

Water consumption for each group is similar during Week 1. During Week 2, Groups 1 and 2 consume much less water since they consume more alcohol. The total fluid intake is similar for each group. Water consumption for Weeks 3, 4 and 5 is similar for each group.

The increase in alcohol consumption by rats fed a vitamin-deficient diet is significant. A correlation exists. Rats fed a vitamin-supplemented diet do not voluntarily consume as much ethanol as rats fed a vitamin-deficient diet.

The effect of vitamin B complex deficiencies is known to cause an increased intake of ethanol by laboratory animals. I have shown the lack of vitamin A in a diet has a similar, but not necessarily as great, effect.

* See page 19, Table 2.
The conclusions from most vitamin-deficiency studies indicate that control mechanisms of the brains of rats are important for regulating alcohol consumption. These mechanisms require complete and fully adequate nutrition, vitamins and possibly the N factor from yeast. If any factor or vitamin is missing, the mechanism of control is off-balance.

A mechanism in the hypothalamus regulates drinking. With a deficiency in one vitamin, such as vitamin A, the hypothalamus is malnourished. Malnutrition may cause an alcohol craving which will have a cyclic effect. The more ethanol consumed, the greater the deficiency of vitamins. Calorie quotas may be maintained but the calories are empty—devoid of any vitamins and minerals.

From this experiment, a correlation may also be suggested for caffeine consumption and ethanol intake. Rats consuming a vitamin A deficient diet and caffeine consume more alcohol than rats fed either a vitamin-supplemented or vitamin-deficient diet. Group 2, which received caffeine in Weeks 4 and 5, maintained an alcohol consumption level of almost twice that of Group 1 and five times more than that of the control group, Group 3. Caffeine may stimulate the drinking in these rats.

Studies with rats have shown that the rats maximize caloric intake by eating a particular quantity of food and drinking a certain amount of alcohol. Each group maintains a fairly constant level of food consumption for Weeks 1, 2 and 3. Group 3 maintains this level also for Weeks 4 and 5. Groups 1 and 2 increase their food consumption in Weeks 4 and 5. Group 1 decreases its alcohol intake during this period and
increases its food consumption to continue a level of caloric intake.

The maximization of caloric intake for Groups 1 and 2, however, cannot compensate for the lack of vitamin A and important minerals, such as zinc and magnesium. Although Groups 1 and 2 consume more food, they gain significantly less weight than Group 3. Group 3 gains weight consistently throughout the experiment. Groups 1 and 2 gain weight initially, but with the continual consumption of "empty" calories, they begin to gain less weight than the animals of Group 3. Group 3 demonstrates the normal weight gain for rats which are not fully grown.

This experiment was conducted using a small sample of animals. Evidence for the effect of vitamin deficiency and caffeine consumption on voluntary ethanol intake is not as concrete as if a large sample of animals had been tested. An individual variation in one animal could potentially change the results of this experiment.

However, despite the small sample and individual differences, a generalized idea on alcoholism has emerged as a result of this experiment and many others before it. A generalized idea on alcoholism is as follows: Vitamin deficiencies and caffeine consumption may increase the tendency for rats to become alcoholic. Consequently, vitamin therapy and decaffeination of the diet may promote recovery from alcoholism.

The cause of alcoholism in humans may be linked to various social and psychological factors. However, to neglect physiological conditions, such as malnutrition, is to neglect a possible cause and eventual cure for alcoholism in some people.
Eckholt's Feed and Grain

# Hamster, Gerbil and Mouse Food

**Table 1**

<table>
<thead>
<tr>
<th>Guaranteed Analysis:</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude Protein</td>
<td>14%</td>
<td></td>
</tr>
<tr>
<td>Crude Fat</td>
<td>4%</td>
<td></td>
</tr>
<tr>
<td>Crude Fiber</td>
<td></td>
<td>15%</td>
</tr>
</tbody>
</table>

**Ingredients:**
- Expanded Corn
- Soybean Meal (dehulled)
- Cereal Fines
- Beet Pulp
- Yeast Culture
- Poultry Fat (preserved with Ethoxyquin)
- Dried Whey
- Dehydrated Cheese
- Choline Chloride
- Pyrooxidine
- Ascorbic Acid
- Folic Acid
- Vitamin B-12 Supplement
- Calcium Pantothenate
- Riboflavin Supplement (fermentation solubles)
- Vitamin E Supplement (with stability improved)
- Vitamin A Palmitate (with stability improved)
- Niacin
- Vitamin D-2 Supplement (with stability improved)
- Thiamine Hydrochloride
- Manganese Sulphate
- Potassium Iodide
- Iron Sulphate
- Cobalt Sulphate
- Copper Sulphate
- Zinc Sulphate
- Tri-Calcium Phosphate
- Ethoxyquin (preservative)
- Calcium Carbonate .5%
- Salt

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### Table 2

**Hartz Small Animal Vitamin Supplement**

<table>
<thead>
<tr>
<th>Guaranteed Analysis:</th>
<th>Minimum</th>
<th>Maximum</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crude Protein</td>
<td>16.0%</td>
<td></td>
</tr>
<tr>
<td>Crude Fat</td>
<td>2.0%</td>
<td>10.0%</td>
</tr>
<tr>
<td>Crude Fiber</td>
<td></td>
<td>11.0%</td>
</tr>
<tr>
<td>Moisture</td>
<td>1.0%</td>
<td>1.5%</td>
</tr>
<tr>
<td>Calcium</td>
<td>1.0%</td>
<td></td>
</tr>
<tr>
<td>Phosphorus</td>
<td>1.0%</td>
<td>1.5%</td>
</tr>
<tr>
<td>Salt</td>
<td>0.1%</td>
<td>1.1%</td>
</tr>
</tbody>
</table>

**Minimum Potency per Pound**

<p>| Vitamin A                          | 15,600 U.S.P. Units |
| Vitamin D₃                         | 1,560 U.S.P. Units  |
| Vitamin E                          | 2 Int. Units        |
| Niacin                             | 25.2 mg             |
| Vitamin C                          | 7.55 mg             |
| Pyrooxidine HCl (Vitamin B₆)       | 3.98 mg             |
| Riboflavin                         | 3.98 mg             |
| Thiamin HCl (Vitamin B₁)           | 3.77 mg             |
| Cobalt                             | 0.010%              |
| Magnesium                          | 0.280%              |
| Iron                               | 0.037%              |
| Copper                             | 0.022%              |
| Manganese                          | 0.004%              |
| Zinc                               | 0.003%              |
| Iodine                             | 0.00002%            |</p>
<table>
<thead>
<tr>
<th>Nutrient</th>
<th>Percentage of U.S. Recommended Daily Allowances Per Serving (2 oz.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein</td>
<td>8</td>
</tr>
<tr>
<td>Vitamin A</td>
<td>0</td>
</tr>
<tr>
<td>Vitamin C</td>
<td>0</td>
</tr>
<tr>
<td>Thiamin</td>
<td>15</td>
</tr>
<tr>
<td>Riboflavin</td>
<td>8</td>
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<tr>
<td>Niacin</td>
<td>10</td>
</tr>
<tr>
<td>Calcium</td>
<td>6</td>
</tr>
<tr>
<td>Iron</td>
<td>8</td>
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LITERATURE CITED


Fowler, Franklin, 1975. Listen. 5:11


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Yillsman, Tom. 1981. Boozers Aided by the Pill. Science Digest. 89:10 pp. 100