

# To Conduct Pigeon Experiment to Study the Effect of Thiamine Deficiency

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**Abstract:** *This study explores the critical role of thiamine vitamin B1 in the diet and its implications on health, specifically through the lens of deficiency syndromes such as beriberi and Wernicke - Korsakoff syndrome. Using a detailed experimental setup involving pigeons as model organisms, the research demonstrates the effects of thiamine - deficient diets, particularly emphasizing the impact of polished versus unpolished rice on health. Historical context is provided, tracing the discovery and structural determination of thiamine, alongside a review of its biochemical properties, dietary sources, and physiological functions. The experimental section meticulously outlines the methodology, including diet formulation, observation protocols, and the assessment of thiamine's role in energy metabolism and nervous system function. The findings underscore the rapid onset of deficiency symptoms in pigeons fed a thiamine - poor diet and the swift recovery following thiamine supplementation, thereby highlighting the essential nature of thiamine in diet and its significant effect on neurological health. The study contributes to a broader understanding of nutritional science, the importance of dietary choices, and the foundational role of vitamins in health maintenance.*

**Keywords:** thiamine deficiency, beriberi, polished rice, dietary sources of thiamine, Wernicke - Korsakoff syndrome

## 1. Objective

- 1) To observe the growth and physical appearance of pigeon on various experimental diet (thiamine sufficient and deficient diet)
- 2) To observe the sign & symptoms of thiamine deficiency pigeon
- 3) To observe the process of the recovery from the deficient symptoms to normal stage on administration of thiamine.

## 2. Introduction

Thiamine is a member of the B - vitamin group. It is characterised by its most important property of being water soluble. The trial of research on thiamine crossed many international boundaries. Eijkman was a Dutch physician who was in Indonesia studying the problem of the high incidence of Beri - Beri among the military. By chance, he noticed that chickens eating the unpolished rice failed to develop a condition similar to beri - beri, whereas those that ate polished rice seemed to suffer from the disease. Eijkman then experimentally induced polyneuritis among the chickens

In 1936, Robert R Williams and his colleagues determined the structure of the anti - beri factor, which they called thiamine.

Thiamine is an extremely important substance in the body because of its role apart of co - enzymes essential for normal metabolism. The disease beri - beri, prevalent in many countries of the world particularly those in Asia – where milled or polished rice is staple in the diet has been recognised for centuries and extracted a great total in human suffering as death is due to thiamine deficiency.

Thiamine deficiency has been shown to cause dysfunction of the human central nervous system, as manifested by beri - beri and the Wernicke - Korsakoff syndrome. Degeneration of peripheral nerves occurs in Rats fed on thiamine – deficient diet and overt ultra structural changes occur in intra

muscular nerves of experimental animals maintain on thiamine deficient diets (Prineas *et al.*, 1970). Thiamine in its pyrophosphate form is an essential cofactor for several enzymes essential to the tri - carboxylic acid cycle, specifically Pyruvate dehydrogenase and alpha - ketoglutarate dehydrogenase.

**Chemistry of Thiamine:** The pure vitamin is identified as thiamine hydrochloride, a crystal compound. It is soluble in water and insoluble in fat. It has a characteristic odour described as “muddy and yeasty” Thiamine is stable in dry heat but destroyed rapidly in a neutral or alkaline solution. An acid solution enhances its stability.

### Forms of Thiamine in the Body

In animal the four different thiamine esters include free thiamine, thiamine mono phosphate (TMP), thiamine pyrophosphates (TPP) and thiamine triphosphate (TTP). Among these thiamine phosphate esters, thiamine pyrophosphate is the most abundant and includes about 80% of the total thiamine. 5 to 10% of the thiamine is in the form of thiamine triphosphate and free thiamine. These four forms of thiamine are inter convertible in the animal body by the interaction of various enzyme systems.

### Function of Thiamine

The most important function of thiamine seems to be its role in the utilization of carbohydrate. Glucose is broken down (oxidised) in the tissue to supply energy.

Another function is that the enzyme transketolase requires thiamine as a co - enzyme. This enzyme is present in red blood cells, the liver, kidney and another tissue except skeletal.

Thiamine seems to be necessary in maintaining a functionally normal nervous system.

### Sources of Thiamine

Thiamine is present in many foods but the amount supplied by any given one is relatively small. No single food in the American diet can be dependent upon to supply a major

portion of the individuals daily thiamine needs. Some foods contain more thiamine than others, and these primary sources include such a lean pork, organ meats, liver, sausage, lean meats, whole grains and enriched breads and cereals green leafy vegetables, nuts and legumes.

### Deficiency of thiamine

A thiamine deficiency may occur in one of two ways: a primary deficiency is the result of an inadequate dietary supply, or a secondary deficiency may come about as a result of conditioning factors that interfere with the proper utilization of thiamine or that increase the demand for thiamine, such as pregnancy and lactation.

A mild deficiency can be produced by a moderately low intake of thiamine. It is characterized by such symptoms as less of appetite, apathy, loss of knee jerk, fatigue, tendencies, nausea and numbness in the legs. Certain mental disturbances occur: moodiness, irritability and mild depression. An electro car - diagram may show variations for normal.

More advanced deficiencies result in the disease beri - beri. There are two types of beri - beri. The dry or peripheral neuritis is characterized by severe muscular wasting, loss of sensation in the skin, loss of weight and paralysis in the lower extremities. The wet form produces marked edema, which usually starts at the extremities and develops upward. When it reaches the trunk, they heart becomes involved, enlarged and frequently the end result is heart failure. Beri - beri in infants is characterized by edema, gastrointestinal difficulties, retarded growth and often cardiac failure.

Animals are used in nutrition research to explain phenomena affecting the human condition, human themselves cannot be used for reasons of ethics, time risk or expense.

When animal is used for obtaining information for a purpose other than for application with that species, it is known as "laboratory animal". Virtually any animal could serve as tool or vehicle on which the study is made.

### Purpose of Using Laboratory Animal

General purpose of using laboratory animals are-

- 1) Research - pure research investigates fundamental biological processes and generally employs a small number of animals with wide variety of different species.
- 2) Toxicological testing: To carry out research on toxicity of foods animals must be physiologically comparable to humans.
- 3) Diagnosis: Animals have also been used for diagnosis of disease and pregnancy.
- 4) Teaching: animals are widely used for teaching dissection and demonstration

Pigeons are one of the species most sensitive to thiamine deficiency. When these animals are placed on a diet of polished rice for 20 - 25 days, typical opistholonus - like convulsion occurs. In convulsive pigeons, a significant decrease of thiamine and calcium in the control nervous system rather than in peripheal nervous system. In subcellular fractions of the brain of the pigeons, thiamine

and calcium levels decreased in the member fraction. When dosages of the thiamine was injected into the muscle of the pigeons during convulsion after an hour started to recover. Hence the present study was done on the effect of thiamine deficiency on animals aim to obtain the following objectives:

- 1) To observe the utilization of thiamine from different food sources.
- 2) To observe the signs and symptoms due to thiamine deficiency in sequence.
- 3) To observe the recovery process

### 3. Review of Literature

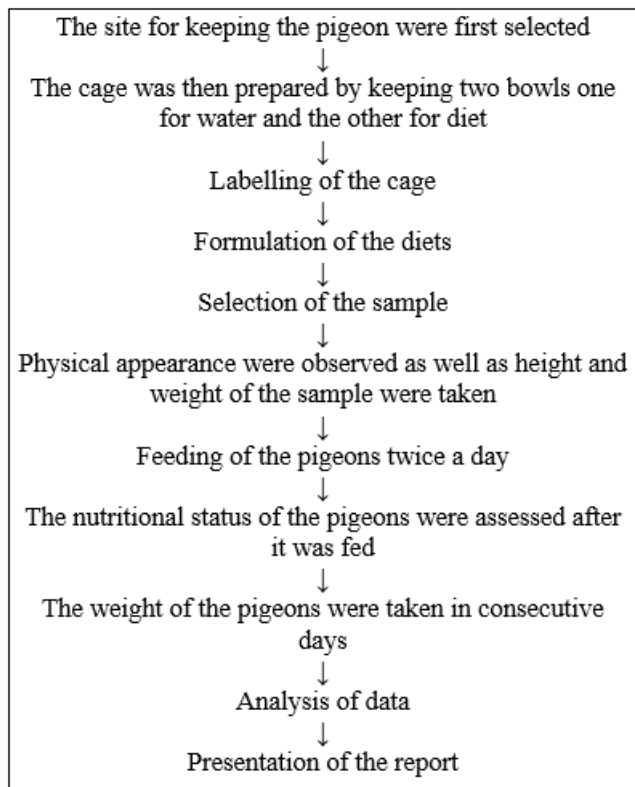
In 1890's Christian Eijkman, a Dutch medical scientist in java, initiated research and observed that the chickens housed in the hospital yard near his laboratory developed a disorder similar to human beriberi when fed a ration solely of polished rice. Because of the general paralysis and head reactions which occurred in the birds, he called this disease polyneuritis. Then he fed chickens with unpolished rice and embryo of the rice kernel and found that the symptoms of the polyneuritis disappeared.

In 1911, polished biochemist Funk, prepared a small amount of a crystalline material with biological activity from a rice bran extract, which he found cured experimental beriberi in birds.

The correct explanation for the etiology of beriberi was proposed in 1901 by Grijns. Grijns found that in addition to rice polishing, katjang hidioe, a green bean and meat could also prevent beriberi in fowl fed a diet consisting mainly of starch. As a result of these studies, he postulated that natural food - stuffs contained an unknown factor, absent in polished rice, which prevented the development the of polyneuritis.

Gries and scoff (1973) studied the comparative pathology of thiamine deficiency in growing chicken. This deficiency caused degeneration of the cells living and raculation is the pancreatic aciner cells. Thiamine act in coenzyme required for energy

### 4. Experimental design



## 5. Materials Required

Five diseased free domestic pigeons  
 Cages for housing the pigeons  
 Feeding bowls  
 Drinking water bowls  
 Wires  
 Weighing balance  
 Thiamine doses  
 Syringe  
 Containers  
 Papers

## 6. Methods

The experiment was conducted for a period of 17 days commencing from 10<sup>th</sup> November to 26<sup>th</sup> November, 2023.

The diet that was given are shown in the following table:

Ingredients	Normal diet I	Experiment diet II	Experiment diet III	Experiment diet IV	Experiment diet V
Unpolished rice	20	-	-	100	-
Polished rice	-	-	50	-	100
Lentil	20	25	12.5	-	-
Black gram	15	25	12.5	-	-
Bengal gram	15	25	12.5	-	-
Green gram	20	25	12.5	-	-
Mustard seed	10	-	-	-	-
	100	100	100	100	100

Firstly, the site for keeping the pigeon were selected and the cages for the respective pigeon was then prepared. The cages were washed and disinfected using dettol and water solution. It was then exposed to sunlight. After the cages were dried, the feeding and drinking bowls were tied tightly to the side of the cages with plastic rope. The cages were then labeled accordingly kept in a clean, well-lighted and ventilated area. Secondly, we formulate the diet for the pigeons. For this, we cleaned the fed, removed the foreign particles like clay, stones, bricks, etc. Polished rice was prepared by washing rice until clear water was obtained. It was then strain and sundried. The prepared fed was then stored in an airtight container and kept in a clean and dry place.

After preparation was ready, five free diseased domestic pigeons were randomly selected from local market. Initially, the weight of the were taken and their physical appearances were observed. They were then kept in their respective cages. The pigeons were kept in the following order:

Cage No 1: The 1<sup>st</sup> pigeon which was housed in the first cage, was fed on normal diet (unpolished rice, Bengalgram, Lentil, Green gram, and Mustard seed)

Cage No 2: The 2<sup>nd</sup> pigeon was housed in the second cage and was fed on the diet containing a mixture of all pulses (Bengal gram, lentil, green gram, and mustard seed)

Cage No 3: the 3<sup>rd</sup> pigeon was housed in the third cage and was fed on the diet containing polished rice and mixture of all pulses.

Cage No 4: The 4<sup>th</sup> pigeon was housed in the fourth cage and was fed on unpolished rice only.

Cage No 5: The 5<sup>th</sup> pigeon was housed in the fifth cage and was fed on rice which was highly polished.

In the experiment, the pigeon were first fed on normal diet for 3 days i. e. from 10<sup>th</sup> November to 12<sup>th</sup> November, 2023. From the fourth day onwards, the pigeon were fed with the experimental diet respectively. The feeds were given twice a day, i. e. in the morning and evening, 20g of feeds were given per serving for each pigeon. Apart from feeding, cleaning of the cages and the area where they were kept were done twice daily at the time of feeding.

The pigeons amount of consumption were recorded daily and their weight were taken every consecutive days, on a platform balance with the capacity of 1kg.

During the final stage of the experimental period, the pigeon being fed only on polished rice showed the symptoms of thiamine deficiency. The deficiency sign and symptoms were recorded were recorded with the help of the photograph taken. Thereafter, the recovery doses were provided by injecting B - complex injection at the rate of

1.5ml. The recovery stages were also keenly observed. After injecting the recovery doses, the experimental diets were discontinued and normal diets were given. Thus, daily observation on all the pigeons were recorded regularly without any lapses.

### 7. Results and Discussion

The present investigation is an attempt to assess the thiamine deficiency in pigeon of, Jorhat. Keeping in view the objectives of the present study, 5 pigeons were selected to maintain a represented sample size. Dietary data, on amount and types of food fed were recorded for 15 days. The impact of diet fed over the pigeons were observed and recorded.

The results of this experiment were given below. The diets of the pigeons were of following types:

**DIET I:** The first pigeon No.1 was fed on normal diet which is consists of a mixture of unpolished rice and four types of pulses.

**DIET II:** The second pigeon was fed on a diet consisting of mixtures of all the types of pulses.

**DIET III:** The third pigeon was fed on polished rice and mixtures of all pulses.

**DIET IV:** The third pigeon was fed on a diet consisting of only unpolished rice.

**DIET V:** The fifth pigeon was fed on a diet consisting of only rice which was highly polished.

### 8. Observation during the Experimental Period

The first pigeon fed on normal diet showed an increase in appetite. The average consumption of the pigeon was active and healthy.

The second pigeon was fed on diet consisting of all the pulses it is observed that the pigeon consumed only lentil but not the other pulses. At first the pigeon showed decrease in appetite but later on it increase gradually. The average consumption of the pigeon was around 21 gm.

The third pigeon was fed on polished rice and mixture of all pulses. The average quantity of consumption of this pigeon was around 30 gm. The pigeon was calm and aggressive in nature.

The fourth pigeon was fed on diet which consists of only unpolished rice. The average consumption of this pigeon was around 18 gm.

The fifth pigeon was fed on highly polished rice. It was observed that there was declination of both weight and appetite of this pigeon. From 10<sup>th</sup> day of the experiment, the pigeon showed the deficiency symptoms of thiamine. It was observed that the pigeon lose appetite and becomes weak. On the final stage, the head turned towards the back (twisted neck) and was unable to stand upright. When the pigeon shows thiamine deficiency, the recovery doses were provided by supplementing thiamine injection i. e. B - complex vitamin injection at the rate of 1.5 ml/dose. The recovery doses provided for 1 day. Then, the normal diets were restored for all the pigeons.

**Table: Changes in weight of the pigeons**

Pigeons	Weights (gm)		
	12 <sup>th</sup> Nov.2023	18 <sup>st</sup> Nov.2023	26 <sup>th</sup> Nov.2023
I	310	325	330
II	260	275	300
III	260	250	300
IV	260	245	260
V	270	220	250

### Physical appearance

After the pigeons were selected and kept in the cages. The physical appearances were observed. The pigeons were identified according to their cages label such as Pigeon I, II, IV and V.

**Pigeon I:** It was observed that the pigeon was healthy, i. e. the eyes were black in colour, its feet were reddish, beak was brown. The colour of the pigeon was black and slightly bluish.

**Pigeon II:** It was black in appearance. Its eyes were red and clear, it had reddish feet and its health was good.

**Pigeon III:** the pigeon was light grey in colour with light brown colour from head to neck portion. It was healthy. Its eyes were black, beak was pink, and feet was pinkish.

**Pigeon IV:** this pigeon was black in colour. Its eyes were black, its feet were reddish, beak was brown.

**Pigeon V:** this pigeon was white in colour. Its eyes were black, feet were reddish, beak was light pink in colour.

**Table: Physical Appearance**

Pigeon No	Sex	Eye	Beak	Behaviour	Feet	Appearance colour
I	Male	Black	Brown	Aggressive	Reddish	Bluish black
II	Female	Red	Brown	Normal	Reddish	Black
III	Female	Black	Pink	Normal	Pinkish	Grey
IV	Male	Black	Brown	Aggressive	Reddish	Black
V	Male	Black	Light pink	Normal	Reddish	White

**Table: Physical and Behavioural changes during the experimentation period:**

Days	Pigeon 1	Pigeon 2	Pigeon 3	Pigeon 4	Pigeon 5
1 <sup>st</sup> - 3 <sup>rd</sup> days	Aggressive	normal	Normal	Aggressive	Normal
4 <sup>th</sup> day	Normal with increase in weight	Normal in weight	Normal in weight	Aggressive in increase in weight	Normal in weight, change in stool colour
5 <sup>th</sup> day	Normal with constant weight	Normal with constant weight	Normal and increase in weight	Aggressive with constant weight	Normal with in appetite
6 <sup>th</sup> day	Normal with constant	Normal with increase	Normal in weight	Aggressive with	Normal with in appetite

	weight	in weight		decrease in weight	
7 <sup>th</sup> day	Normal with constant weight	Normal with increase in weight	Normal with constant weight and slightly change in stool colour	Aggressive and decrease in appetite	Normal with in appetite
8 <sup>th</sup> – 10 <sup>th</sup> days	Active with increase appetite and increase with weight	Normal increase in weight and appetite	Normal in weight	Aggressive and change in stool colour	Shows slight symptoms, long of appetite
11 <sup>th</sup> – 14 <sup>th</sup> days	Active with constant weight	Normal with constant weight	Normal and no changes in stool colour	Aggressive and active with constant weight, changes in stool colour	Severe symptoms like twisting of neck, loss of coordination of link. So 1.5 ml b - complex injection was injected.
15 <sup>th</sup> – 17 <sup>th</sup> days	Active with constant weight	Active with constant weight	Active with constant weight	aggressive with constant weight	Showed recovery

## 9. Summary and Conclusion

### 9.1 Summary

The experiment on effect of thiamine deficiency on animals can be summarized by the following points -

- The animals experiment continued for a period of 17days.
- The animal species selected for the experiment purpose were pigeons and were housed in separate cages by providing separate diets, i. e. normal and experimental diet.
- During the experimental period we observed feeding pattern, amount of consumption, weight of the animal, behavior and all other important aspect and recorded without any lapse.
- The fifth pigeon which was fed with only highly polished rice showed thiamine deficiency symptoms. The symptoms were twisting of the head towards the back and appear to be in a star gazing fashion, severe muscle and tissue wasting and lack of limb co - ordination due to which the pigeon could not stand on its feet.
- Recovery doses were provided in the form of thiamine injection viz, B - complex vitamin injection at rate of 1.5ml/dose.
- Normal diet was restarted for all the pigeons after administering the thiamine recovery doses to the affected pigeon, thus the experiment concluded.

### 9.2 Conclusion

On completion of the experiment, it was observed that, the pigeon which was fed only with highly polished rice showed the deficiency symptoms of thiamine. Therefore, this proved that thiamine is present in unpolished rice at a high range (about 0.06 mg present in raw milled rice)

Pigeons were selected for the experiment due to the fact that they show the symptoms of thiamine deficiency of at very rapid rate. Such deficiency symptoms are twisting of the head towards the back, gross muscle and tissue wasting, etc.

The recovery rate was also observed to be very fast after the administration of the B - complex injection at the rate of 1.5/ml dose. Thus, it can be opined that any type of rice should not be washed for a long period of time. Otherwise, there will be loss of nutrients including thiamine (vit. B<sub>1</sub>). Thus the entire experiment concluded.

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