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Flame Photometric Determination of Potassium in Irradiated Alfalfa (*Medicago sativa* L.)

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ABSTRACT

Alfalfa is often grown in fields by farmers for pasturage and forage. With respects to the nutritional properties and information of Alfalfa plant is rich in calcium, sodium, potassium, carotene, zinc, iron and vitamins such as A, C, B_1, B_6, E and K as well as proteins. It is extensively cultivated in Maharashtra as a fodder crop and also having a medicinal value. In the present investigation, the seeds of Alfalfa were irradiated with different doses of gamma radiation (5 KR, 10 KR, 15 KR, 20 KR, 25 KR, 30 KR, 35 KR, 40 KR, 45 KR, and 50 KR) and treatment combination were arranged in a randomized block design with eleven replication to determine potassium content. Data were determined with the help of Flame photometry method. This method was properly validated by using standard chemicals and can be applied to formulation. The proposed method was found to be simple, specific, inexpensive and less time consuming. Result obtained in the present investigation, it can be determined that the mutagens are highly energetic in inducing genetic variability with considerable variation in potassium content of Alfalfa.
Keywords: Potassium, Alfalfa, Fodder, Flame photometry.

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INTRODUCTION

Alfalfa is important forage crops worldwide due to its high forage yield, excellent forage quality in a wide range of environments, and high malleability to different climatic conditions [13]. From the point of view of farmers and world's agricultural scientist is considered to be the "Queen of Fodder Herbs" [19]. Due to high nutritive value and high proteins content, alfalfa is widely used in animal fodder. It contains minerals like Calcium, potassium, phosphorus, Magnesium and zinc as well as chlorophyll, organic acids, saponins, isoflavins, sterols, coumarins and alkaloids [14]. In general, Cobalt-60 and Cesium-137 are the main sources of gamma rays used in mutation induction [10]. Gamma rays are characterised in ionizing radiation because these radiations cause alterations in the anatomy, morphology, Physiology and biochemistry of the plants [12]. Gamma radiation can induce useful as well as harmful effects on crops so there is a need to predict the most beneficial dose for improvement of specific traits of crop plants [8]. Potassium is not only performs the important physiological functions as, but it improves nitrogen use efficiency. Potassium (K) increases crop yield and improves quality. It is required for numerous plant growth processes [9]. Flame Photometry also recognized as flame atomic emission spectrometry that observed the species in form of atom, mostly it works on principle of ionization of alkali metal salt drawn into a non-luminous flame. Alkali metal salt absorb energy from flame and emit the light of characteristic wavelength which is observed by change in intensity of color, the energy absorb was enough to vaporize alkali metal salt [7]. Flame photometer is an optical device to measure the colour intensity of substances, such as sodium, potassium that have been aspirated into a flame [3]. The objective of this study was to determine the Potassium content of Alfalfa which were treated with different doses of gamma radiation by flame photometry method.

MATERIAL AND METHODS

Method of Sample Collection:

Alfalfa plant samples were collected from all the treated and untreated concentration, which were planted in field by RBD method with eleven replication.[5].

Preparation of dried Alfalfa powder:

For preparation of dried alfalfa powder, fresh Alfalfa leaves with stem of various doses was dried at room temperature in a dark room. The dried Alfalfa after 3-4 days was powdered in a grinder [16].

Preparation of various extract for dried Alfalfa plant:

Wet digestion of sample: For wet digestion of sample, exactly (1.000gm) of the powdered sample was taken in digesting glass tube. Twelve milliliters (12ml) of HNO3 was added to the samples and mixture was kept for overnight at room temperature. Then 4.0 ml perchloric acid (HClO4) was added to this mixture and was kept in the fumes block for digestion. The temperature was increased gradually, starting from 50ºC and increasing up to 250-300 C. The digestion completed in about 70-85 min as indicated by the appearance of white fumes. The mixture was left to cool down and the contents of the tubes were transferred to 100 ml volumetric flasks and the volumes of the contents were made to 100 ml with distilled water. Stored the digest and used it for mineral determination [1].

Determination of Potassium (K) in Alfalfa by flame photometer:

Principle: The flame photometer measures the emission of radiant energy when atoms of an element return to their ground state after their excitation by the high temperature of the flame. The degree of emission is related to the concentration of the element in the solution [1].

Procedure: Potassium analysis of the sample were done by the method of flame photometry. Standard solution of potassium (K) were prepared by dissolving 190.6 mg of Potassium chloride (KCl), so it gives 100 ppm of K [1]. Standard solutions of 20, 40, 60, 80 and 100 milli equivalent/L were used both for K. The same wet digested sample solutions were used for the determination of potassium [4].

Measurement procedure:

Sample of irradiated Alfalfa extracts were analyzed for mineral detection using standard and distilled water as a reference. For various extract of Alfalfa, standards prepared in distilled water was used. Here instrument has given concentration of minerals in millimole/100ml. From this, amount of minerals present in terms of mg/g Alfalfa were calculated [6].

RESULTS AND DISCUSSION

Result for analysis of potassium in various irradiated extract of Alfalfa in terms of mmol/100ml, ppm and mg/g Alfalfa is given in following table:

Table 1.1- Standardization of Potassium solution:

S. No.	ml. of standard (ppm) sol^n	Dilution	Conc. of Standard (ppm)	Meter Reading (Potassium)
1	20	100	20	16
2	40	100	40	37
3	65	100	60	58
4	80	100	80	78
5	100	100	100	100

Repeatability of method is checked by repeating the same procedure by preparing standard solution for three times on three different days. This is mean of three meter reading for Potassium which are taken on three different days.

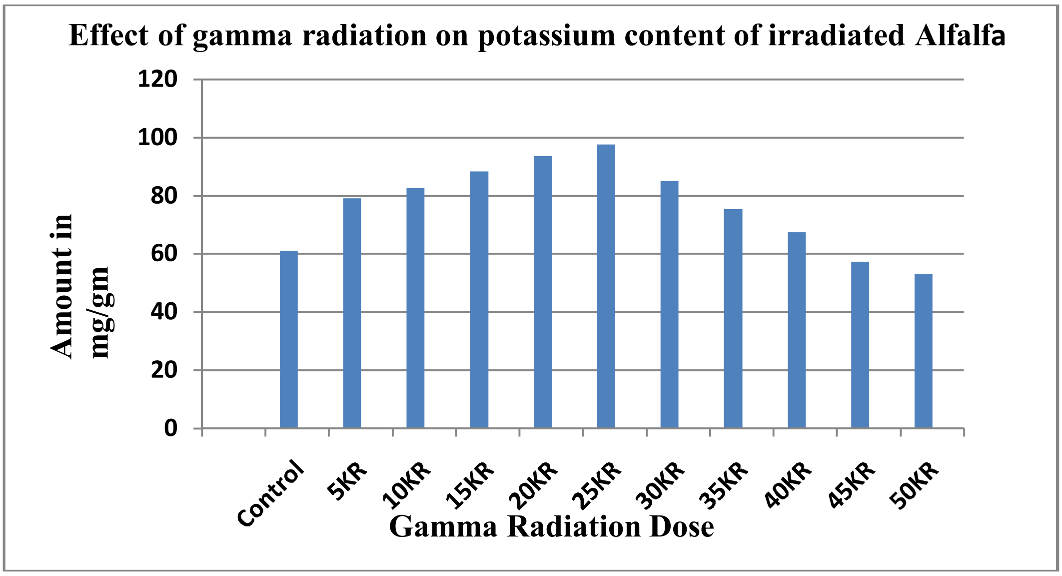
Effect of Gamma Radiation on Potassium content of Irradiated Alfalfa:

The result reveals that average maximum content of Potassium in the Alfalfa was 97.6 mg at 25KR dose of gamma radiation. However, minimum average content of Potassium was reported in 50KR and control with 53 mg and 61 mg respectively. It is quite noticeable that lower doses of gamma radiation shows maximum content of Potassium while higher doses of gamma radiation shows minimum content of Potassium. 20KR and 25KR doses of gamma radiation shows maximum content of Potassium. When increasing the dose of gamma radiation from 30KR to 50KR the amount of Potassium goes on decreasing. The amount of Potassium in Alfalfa shows variations in all the doses of gamma radiation and control.

Table 1.2-Effect of Gamma Radiation on Potassium content of Irradiated Alfalfa

Sr. No.	Sample of Irradiated Alfalfa	Amount of Potassium in mg/gm			Mean	S.D.
		1st Day	2nd Day	3rd Day		
1	Control	62	60	61	61	1.00
2	5KR	78	80	79	79	1.00
3	10KR	83	82	83	82.6	0.57
4	15KR	89	88	88	88.3	0.57
5	20KR	93	94	94	93.6	0.57
6	25KR	98	97	98	97.6	0.57
7	30KR	86	85	84	85	1.00
8	35KR	76	74	75	75.3	1.00
9	40KR	68	67	67	67.3	0.57
10	45KR	58	57	57	57.3	0.57
11	50KR	54	53	52	53	1.00

(Three times with the same extract on three different days by repeating the calibration with freshly prepared standards.)



Graph 1. Effect of Gamma Radiation on Potassium content of Irradiated Alfalfa

However, the method of sample preparation by wet digestion method shows relatively variation. Similar result also obtained by Sanni T.A., [15] studied the effect of gamma Irradiation on mineral, vitamin and cooking properties of Sorrel (*Hibiscus Sabdariffa* L.) and recorded that Potassium was significantly increased with increasing radiation dose to 2.0KGy but sodium was significantly reduced at 2.5KGy than at any other dose level. Mohajer *et.al*, [12] also reported that lower doses of gamma radiation have stimulatory effects on nutritional composition of Sainfoin. Borzouei *et.al*, [2] also observed that lower doses will be significant as compared to higher doses. Along with the stimulation of cell division and enzymatic activity there is an increase of mineral and water uptake, which can explain the increase if assimilatory pigments in plants derive from seeds exposed at lower doses [11]. Low doses of Ionizing radiation have modulatory role in the metabolic and biochemical processes of seedling [18].

CONCLUSION

It is evident from this study that lower doses of gamma radiation i.e. 5KR, 10KR, 15KR, 20KR and 25KR is mostly effective as compared to higher doses with 35KR, 40KR, 45KR and 50KR and control. The stimulatory effect at a lower dose is due to the fact that mutagens at lower concentration stimulate the role of enzyme and growth hormone responsible for growth and inhibitory effect is due to the fact that biological damage increased at a faster rate in higher concentration of mutagen. It is concluded from this study that appropriate mutation or variation in Potassium content of Alfalfa can be created through gamma rays and it can be improved in various genotypes through various gamma ray doses. The proposed flame photometric method was successfully employed to estimate the amount of potassium in eleven different treatments of Alfalfa. The proposed method was found to be simple, specific, accurate and precise. Hence, the proposed method can be employed for routine analysis of mineral composition in various plant extracts.

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