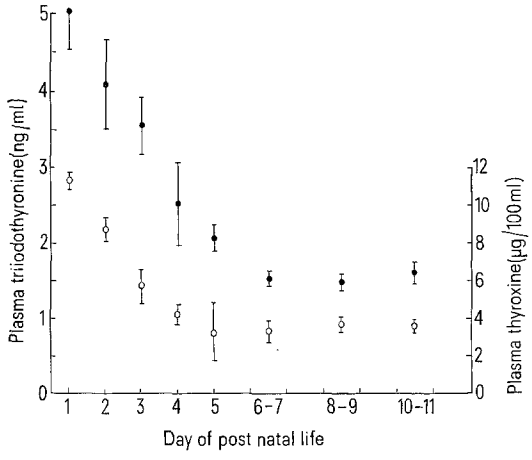
Plasma Triiodothyronine Concentration in the Newborn Calf

In the first days of extra-uterine existence rapid changes occur in the level of function of the pituitary-thyroid axis in many species. In the calf, plasma thyroxine concentration is high on the first day of life and falls rapidly in the early neonatal period. This decrease in plasma concentration is accompanied by a period of decreased turnover of exogenous labelled thyroxine injected into the circulation and a decreased peripheral utilization of thyroxine per kg body weight[1,2]. During this period there is also a decrease in the proportion of circulating thyroxine that is free or dialysable (A.L. THOMAS, unpublished observations).



Ordinates: plasma thyroxine (○) and plasma triiodothyronine (●) concentrations in the newborn calf. Abscissa: age after birth in days. Individual points are the means and S.E.M. of at least 7 calves.

Using a radioimmunoassay for triiodothyronine in plasma[3], plasma triiodothyronine concentration has been measured in the newborn calf (Figure). Plasma thyroxine was measured in the same samples by competitive protein binding[4]. The triiodothyronine: thyroxine ratio (expressed as ng/ml T_3: µg/100 ml T_4) is 0.43 ± 0.06 (mean ± sem; $n = 7$) on day 1. It rises to a maximum of 0.67 ± 0.08 on day 5, falling again to 0.45 ± 0.06 on day 8–9. The source of plasma triiodothyronine may be either direct secretion of triiodothyronine from the thyroid or peripheral conversion of thyroxine to triiodothyronine. Triiodothyronine is 3–4 times as potent as thyroxine in man[5]. It is therefore possible that more efficient production of triiodothyronine is responsible for the decline in the overall utilization of thyroxine.

Résumé. On a mesuré le taux de triiodothyronine et de thyroxine dans le plasma du veau nouveau né pendant ses premiers 11 jours. Le ratio d T_3:T_4 a eu son maximum au 5e jour.

P. W. NATHANIELSZ and A. L. THOMAS[6]

Physiological Laboratory, Cambridge CB2 3EG (England), 4 May 1973.

1 P. W. NATHANIELSZ, .J Physiol. *196*, 54P (1968).
2 P. W. NATHANIELSZ, J. Physiol. *204*, 43P (1969).
3 M. HUFNER and R. D. HESCH, Acta endocr. *72*, 464 (1973).
4 B. E. P. MURPHY and C. J. PATTEE, J. clin. Endocr. Metab. *24* 187 (1964).
5 K. STERLING, Recent Prog. Horm. Res. *26*, 249 (1970).
6 Acknowledgements. This work was supported by the Medical Research Council. The triiodothyronine antiserum was kindly provided by Dr. R. D. HESCH.

The Nutritional Status and Radiosensitivity of Some *Cicer arietinum* L. Cultivars

Amongst pulses; the chickpea (*Cicer arietinum* L.) plays an important economic role, with an acreage next to rice and wheat. It has, however, a surprisingly low yield[1], predominantly due to a poor genetic make-up in the existing varieties, poor cultural practices, and susceptibility to disease and insect pests. Induced mutation breeding for high yield and/or pod borer resistance in *Cicer arietinum* L. without adversely affecting the nutritional status appears promising as such applications have already led to improved yield potentials in several other crops[2–4]. This paper deals with estimating the nutritional status and radiosensitivity of the 3 predominant *Cicer arietinum* L. cultivars (C-612, Sanyasi, and Chhola) to acute γ-irradiation exposures before induced mutagenesis studies are undertaken.

Material and methods. One-year-old seeds of *Cicer arietinum* L. cultivars C-612, Sanyasi, and Chhola; obtained from Dokri Rice Research Station, Dokri, Pakistan; were used to determine the nutritional status and radiosensitivity.

Nutritional evaluation. 50 g seed samples of cultivars C-612, Sanyasi, and Chhola, were ground on a micro sample mill to pass through a 40 mesh sieve size and stored in air-tight containers. Standard procedures for moisture and Kjeldahl protein[5], sample hydrolysis[6] and amino acid analysis[7] were adopted.

Radiosensitivity evaluation. 8 seed lots of 100 seeds for each cultivar were prepared and given single γ-irradiation exposures of 2.5, 5.0, 7.5, 10.0, 15.0, 20.0 and 25.0 kR from a ^{60}Co 100 Ci source. Dose rate was 70 R/min at 10 cm distance. Lot 8 served as the control. The seeds were planted immediately in coarse sand and maintained under laboratory conditions. The planting design was a randomized complete block, with 25 seeds per treatment and replication. Observations for seed germination and seedling height were recorded 14 days from planting. D_{50} was determined according to OSBORNE and LUNDEN'S[8]

1 *F.A.O. Production Year Book* (Food and Agriculture Organisation of United Nations, Rome 1968), vol. 22, p. 166.
2 B. SIGURBJORNSSON and A. MICKE, IAEA/SM-21/60 (1969), p. 673.
3 K. A. MUJEEB and J. K. GREIG, Radiat. Bot. *13*, 121 (1973).
4 S. M. VASTI, S. H. SIDDIQUI, K. A. MUJEEB and G. MUSTAFA, Nucleus *9*, 55 (1972).
5 *Methods of Analysis*, Association of Official Agricultural Chemists, 1th edn. Washington 1970), p. 1015.
6 D. H. WAGGLE, D. B. PARRISH and C. W. DEYOE, J. Nutr. *88*, 370 (1966).
7 D. H. SPACKMAN, W. H. STEIN and S. MOORE, Analyt. Chem. *30*, 1190 (1958).
8 T. S. OSBORNE and A. O. LUNDEN, Int. J. appl. Radiat. Isotopes *10*, 198 (1961).

Table I. Protein content, amino acid composition and essential to total amino acid ratios of 3 *Cicer arietinum L.* cultivars

	C-612	Sanyasi	Chhola
Protein (moisture free)	20.6[a]	19.9[a]	22.6
Amino acids[b]			
Lysine	5.7	7.1	7.1
Histidine	2.7	2.6	2.6
Ammonia	1.9	1.7	1.8
Arginine	10.5	9.0	9.1
Aspartic acid	13.0	10.3	12.4
Threonine	3.6	3.6	3.9
Serine	5.3	5.9	5.2
Glutamic acid	18.2	18.8	17.5
Proline	4.0	4.1	4.0
Glycine	4.1	4.1	4.0
Alanine	4.3	4.4	4.2
Half cystine	1.8	2.3	1.9
Valine	4.2	4.5	4.4
Methionine	0.5	1.3	1.2
Isoleucine	4.1	4.2	4.2
Leucine	7.7	7.8	7.7
Tyrosine	2.7	2.8	3.0
Phenylalanine	5.6	5.7	5.6
Recovery nitrogen basis	103.6	100.4	102.9
Essential/total amino acid[b] ratio	0.45	0.41	0.46

[a] Values with same alphabet are not significantly different at $p \geqslant 0.05$. [b] Calculated to 100% recovery and to a moisture free basis.

Table II. Mean germination count (%) of *Cicer arietinum* L. cultivars

Cultivar	Dosages in kR								
	0	2.5	5.0	7.5	10.0	15.0	20.0	25.0	Mean[b]
C-612	85	80	85	85	83	78	85	83	83[a]
Sanyasi	80	78	71	75	69	52	57	48	66
Chhola	71	71	71	73	73	64	55	62	67
Mean[c]	79[a]	76[a]	76[a]	78[a]	75[a]	65	65	64	

[a] Values with same alphabet are not significantly different at $p \geqslant 0.05$. [b] Means for comparing cultivars. [c] Means for comparing treatments.

Table III. Mean seedling height (cm) of *Cicer arietinum* L. cultivars

Cultivar	Dosages in kR								
	0	2.5	5.0	7.5	10.0	15.0	20.0	25.0	Mean[b]
C-612	8.1	8.2	7.0	7.4	8.4	6.9	7.0	6.2	7.4[a]
Sanyasi	7.3	7.0	7.7	8.2	6.7	3.3	4.0	5.7	6.2
Chhola	7.2	7.3	6.7	7.0	6.4	4.2	3.3	3.0	5.6
Mean[c]	7.6[a]	7.5[a]	7.1[a]	7.5[a]	7.2[a]	4.8	4.8	5.0	

[a] Values with same alphabet are not significantly different at $p \geqslant 0.05$. [b] Means for comparing cultivars. [c] Means for comparing treatments.

procedure. The germination data (percentage) was transformed[9] (arcus sinus) prior to analysis. Dosage/germination, and dosage/seedling height correlations and regression equations were further calculated.

Results and discussion. The cultivars C-612, Sanyasi, and Chhola are the 3 leading cultivars of this ecological zone. C-612 is extensively consumed as a food grain and would be the preferred cultivar for induced mutagenesis studies. As literature in Pakistan completely lacks information of nutrition and radiation sensitivity investigations in *Cicer arietinum* L., both aspects have been evaluated for all 3 cultivars.

The protein content (moisture free basis) in these cultivars was 19.9% for Sanyasi, 20.6% for C-612 and 22.6% for Chhola. Though Chhola possessed a significantly greater protein content than Sanyasi and C-612, the amino acid compositions of all the cultivars did not offer any remarkable variations (Table I) except for low lysine (5.7) and high arginine (10.5) contents of C-612. These differences appear balanced out by the essential: total amino acid ratio (Table I). AMIRSHAHI and TAVAKOLI[10] had reported differences in protein content between different varieties in all species of the pulse crops which they studied in Iran. Accordingly genotypic effects on variations in amino acid compositions would be an obvious inference, a view earlier held by RAGHAVAIAH et al.[11].

The nutritional data indicates that together with the other objectives for the mutation breeding programme with *C. arietinum* L. cv. C-612, an initial selection pressure would have to be exercised so as to carry progenies into M_2 with favourable protein contents (higher than or

[9] W. G. SNEDECHOR, *Statistical Methods*, 5th edn. (The Iowa State Univ. Press, Ames, Iowa, USA 1956), p. 534.
[10] M. C. AMIRSHAHI and M. TAVAKOLI, *Improving Plant Protein by Nuclear Techniques* (I.A.E.A., Vienna 1970), p. 331.
[11] P. RAGHAVAIAH, V. P. AHUJA, A. K. KAUL and M. S. NAIK, Curr. Sci. *40*, 58 (1971).

Table IV. Mean seedling performance values for *Cicer arietinum* L. Cultivars

Cultivar	Dosages in kR								
	0	2.5	5.0	7.5	10.0	15.0	20.0	25.0	Mean[b]
C-612	100	95.0	86.4	91.4	101.9	78.4	86.4	75.5	89.4[a]
Sanyasi	100	93.5	93.7	105.3	79.2	29.4	39.1	46.9	73.4
Chhola	100	98.7	93.4	97.4	89.9	52.0	52.0	53.3	79.6
Mean[c]	100[a]	95.7[a]	91.2[a]	98.0[a]	90.3[a]	53.3	59.2	58.6	

[a] Values with same alphabet are not significantly different at $p \geqslant 0.05$. [b] Means for comparing cultivars. [c] Means for comparing treatments.

Table V. Dosage/germination, and dosage/seedling height correlations and regression

Cultivar	Dosage/germination		Dosage/seedling height	
	Correlation coefficient	Regression equation	Correlation coefficient	Regression equation
C-612	–0.406	$\hat{y} = 84.275–0.12\ X$	–0.742[a]	$\hat{y} = 8.092–0.067\ X$
Sanyasi	–0.944[b]	$\hat{y} = 80.339–1.326\ X$	–0.884[b]	$\hat{y} = 8.010–0.190\ X$
Chhola	–0.922[b]	$\hat{y} = 74.789–0.686\ X$	–0.994[b]	$\hat{y} = 7.788–0.204\ X$

[a] Significant at $p \geqslant 0.05$. [b] Significant at $p \geqslant 0.01$.

equal to control). The lower lysine content and fluctuation in other amino acids may also improve, as it is known[12] that mutants having high amino acid contents can be obtained after mutagenic treatments.

In the radiosensitivity studies, the seedlings emerged earlier in 2.5 and 5.0 kR in all cultivars, but the final germination count did not depict a significant stimulation for these treatments (Table II), where a significant ($p \geqslant 0.05$) germination depression occurred from 15.0 kR. The effect of 15.0, 20.0 and 25.0 kR was identical. Varietal germination differences indicated a significantly greater radioresistance for C-612 than Sanyasi or Chhola. The germination of the latter two was identical. Seedling height measurements (Table III) manifested the radiation effect by a significantly delayed seedling growth from 15.0 kR, with the higher dosages offering a seedling height nonsignificance. The seedling height varietal trend was also manifested by estimated seedling performance[8] (Table IV). Such data yielded D_{50} values of 24.0, 10.0 and 12.1 kR respectively for C-612, Sanyasi and Chhola. Related decreased germination and reduced growth characteristic results with increased radiation exposures have been earlier reviewed, and reported for *Phaseolus vulgaris* L. cv. Blue Lake by MUJEEB and GREIG[3].

Except for germination of cv. C-612, significant negative correlations with dosage of this characteristic were obtained for Sanyasi and Chhola. Seedling height and dosage were negatively correlated for all cultivars (Table V). These were significant. The significance level of Sanyasi and Chhola was greater than C-612. Nonsignificant correlation for such traits have been considered by SIDDIQUI and MUJEEB[13] to be a part result of growth stimulation. In the present study, the nonsignificant germination/dosage and 0.05% level seedling height/dosage correlation of cv. C-612 has been considered a function of greater radioresistance.

These initial findings for developing a mutation breeding programme with cv. C-612 provided a D_{50} value of 24.0 kR, a protein content of 20.6%, and a varied amino acid composition compared to cultivars Sanyasi and Chhola. This composition could be improved with greater emphasis attached to isolating variants depicting breaks in the negative high yield/protein/lysine correlations.

Résumé. Etat nutritif et radiosensibilité de 3 cultures de *Cicer arientum* L. (C-612, Sanyasi et Chhola). Le pourcentage de protéine était de l'ordre de 19.9, 20.6 et 22.6 pour Sanyasi, C-612 et Chhola respectivement. Chez C-612 le niveau de lysine est bas et celui d'arginine élevé. Le rapport de pourcentage entre le total des acides aminés et les protéines fut le même dans toutes les cultures. Le C-612 était significativement ($p \geqslant 0{,}05$) plus résistant aux rayons gamma, comme l'ont montré les relations: dosage/germination et dosage/hauteur de plantule.

K. A. MUJEEB[14] and S. H. SIDDIQUI

Plant Genetics Division,
Atomic Energy Agricultural Research Centre,
Tandojam (W. Pakistan), 28 May 1973.

[12] W. F. TONG, Y. E. CHU and H. W. LI, *Improving Plant Protein by Nuclear Techniques* (I.A.E.A., Vienna 1970), p. 71.

[13] S. H. SIDDIQUI and K. A. MUJEEB, Stimul. Newslet. *4*, 12 (1972).

[14] Acknowledgment. The help provided by Prof. C. W. DEYOE, Kansas State University, Kansas 66506, USA, for the nutritional estimations is gratefully acknowledged.

Identifying *Y* Chromosome in Interphase Nuclei of the Human Brain

A technique has been suggested for identifying certain specific species of heterochromatin by means of fluorescent staining with quinacrine mustard and quinacrine dihydrochloride. This technique permits us to identify the *Y* chromosomes in metaphase plates and in interphase nuclei (CASPERSSON et al.[1], PEARSON et al.[2]). In consequence it is possible to identify the fluorescent site of the *Y* chromosome among the nuclear structures, especially with respect to the nucleolus and the nuclear membrane.

There are advantages in using in neural tissues in such studies, for it is known that the neurons do not divide and this produces a relatively homogeneous mass of interphase nuclei with a very conspicuous nucleolus.

In order to obtain a monolayer, prints were produced by applying to slides surfaces of blocks obtained from the brain cortex, the hypothalamus, the medulla and the pontal tissue of 9 women and 8 men who have died 6–24 h before the material was taken. The material was fixed for 15 min in Carnoy's fluid. The slides were then washed with 96% alcohol and dried. Staining was carried out for 5 min in 0.5% solution of quinacrine dihydrochloride (National Institute of Health, USA), which was kindly provided by Professor M. GREEN, California University, USA, or quinacrine mustard (National Cancer Insitute, Bethesda, USA) prepared in a phosphate

[1] T. CASPERSSON, L. ZECH, C. IOHANSSON, I. LINDSTEN and M. HULTEN, Expl. Cell Res. *61*, 472 (1970).

[2] P. L. PEARSON and M. BOBROW, Nature, Lond. *226*, 79 (1970).