Table II. Protein and significant amino acid compositions of *Cicer arietinum* L. cv. Chhola as influenced by γ-radiation seed treatment

Characteristics	Dosages (kR)								LSD (0.05)
	0	1	2	3	4	5	7.5	10	
Protein (moisture free)	19.5	19.3	19.4	19.5	19.5	19.5	19.0	19.0	NS
Amino acids[a]									
Arginine	9.22 ± .04	9.43 ± .07	8.49 ± .04	9.25 ± .04	9.16 ± .11	9.04 ± .22	8.68 ± .04	8.91 ± .10	.37
Aspartic acid	12.02 ± .01	12.01 ± .03	12.24 ± .10	12.12 ± .02	11.96 ± .02	12.21 ± .02	12.17 ± .04	12.16 ± .03	.13
Recovery (nitrogen basis)	93.09	94.25	93.22	92.34	93.53	91.89	96.31	94.42	

[a] Amino acid values were corrected to 100% recovery of KJELDAHL protein and to a moisture free basis.

nonsignificant correlation with dosage. The significant growth reductions so characteristic of high dosages, existed for height and seedling performance. The negative correlations of seedling height and performance with dosage have provided additional support for this growth trend (Table 1). Such reductions have been earlier reported[7–10].

Significant variations in amino acid compositions were obtained for arginine and aspartic acid. The latter provided a definite increasing trend from 5.0 kR while erratic decreases were obtained for arginine at 2.0 and 7.5 kR (Table II). The nutritional parameters studied[11] suggest that up to 10.0 kR no adverse trends developed to create possibly deleterious first generation effects that may enhance the frequency of not overcoming the negative yield/protein/nutritional quality correlations. Current mutation breeding programmes have attached importance to improvement of protein content and quality. This trait has been given due importance as the final selection sieve for mutants in advanced generations[12] or as an M_1 selection parameter[13]. The merits and demerits of either approach have been considered[13] earlier, with the essentiality of such qualitative assessment put forward by JOHNSON et al.[14] and GOTTSCHALK and MULLER[15]. For *C. arietinum* L., a diploid, the author feels that the nutritional aspects should form the initial sieve. Subsequent efforts may then be directed towards isolating progenies depicting maximum combination of desirable characters and possessing the adverse negative-correlation breaks.

The morphological data in general did not manifest adverse irradiation effects up to 4.0 kR, and has provided information for controlling treatments that a greater desirable frequency of mutability is achieved in order to facilitate making multidirectional selections. This may lead to blending in an 'ideal variant' the improved desired trait/s coupled with the carry through of the nutritive quality genetic pool as is or be positively directed. Although the nutritional status was not adversely effected up to 10.0 kR, negative trends did appear for the growth parameters. It may hence be advantageous not to use the higher range for induced mutation studies, considering the greater possibility of altering the otherwise optimum cultivar characters.

The influence of acute γ-radiation exposures upon the nutritional and growth parameters of *Cicer arietinum* L. cv. Chhola was studied. Consistent adverse variations in protein and amino acid compositions did not occur up to 10 kR. Growth parameters of germination and seedling height provided varying degrees of significant ($p \geqslant 0.01$) stimulatory responses and growth reductions. Interpretations from the nutritional and morphological data have been made for developing a mutation breeding programme.

Zusammenfassung. Nachweis, dass es mittels Röntgenstrahlen bei *Cicer arietinum* L. cv. Chhola gelingt, günstige Mutationen bezüglich Aminospuren und Proteinen herzustellen.

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The Effect of Starvation on the Chemical Composition of Red and White Muscles in the Plaice (*Pleuronectes platessa*)

Recent studies[1,2] have shown that red and white muscles respond differently to prolonged starvation. In the present study on the plaice (*Pleuronectes platessa*) the levels of nucleic acids have been investigated in the red and white myotomal muscles during starvation. The previous investigations[2] concerning water, glycogen and protein content of the two muscles have also been extended to fish starving for up to 30 weeks.

Materials and methods. The method of capture and treatment of fish has been described previously[2]. Fish

Table I. Changes in the concentration of red and white muscle water, glycogen and protein nitrogen

	Condition index	Water (%)		Glycogen (mg glycogen/100 g muscle)		'Insoluble' protein nitrogen (g PN/100 g muscle)		'Soluble' protein nitrogen (g PN/100 g muscle)	
		Red muscle	White muscle	Red muscle	White muscle	Red muscle	White muscle	Red muscle	White muscle
Concentration (0 weeks)	10.50	$82.4^{+0.4}_{-0.5}$	$81.7^{+0.5}_{-0.6}$	635.5 ± 51.6	181.6 ± 28.2	1.67 ± 0.11	2.70 ± 0.16	0.56 ± 0.06	0.34 ± 0.0^4
Concentration (30 weeks)	7.67	$85.6 \pm^{1.3}_{1.5}$	$95.1^{+1.1}_{-1.7}$	329.7 ± 37.6	41.1 ± 10.8	0.90 ± 0.19	0.41 ± 0.05	0.09 ± 0.01	0.11 ± 0.02
% Change	—	+	+	—	—	—	—	—	—
Significance (P)	—	< 0.05	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

The values given represent the means and standard errors of 8 determinations.

were killed by plunging them into liquid nitrogen (−170 °C). Red muscle was dissected whilst the fish was still frozen (−20 °C) from both dorsal and ventral sides. White muscle was dissected from the epaxial musculature adjacent to the dorsal fin. Frozen (−170 °C) samples of dorsal and ventral red muscle were finely chopped and mixed to obtain a homogenous sample. Quick freezing the fish minimizes changes resulting from struggling or post mortem metabolism. The initial samples were taken 3 days after the fish were captured and the final sample after 30 weeks of total starvation. The SCHMIDT THANNHAUSER procedure[3] was adopted to extract RNA and DNA following pulverization of 100 mg samples of the muscle by a freeze grinding technique[4]. DNA was assayed by a modification of the Ceriotti method[5], and RNA by the orcinol method[6]. Glucose contamination in the RNA assay was corrected for by reading at 2 wavelengths. Water contents, glycogen and nitrogen concentrations of protein fractions were determined as described previously[2]. The significance between groups of data was calculated using the students t-test. In cases of unequal variance between sets the Brehrens-Fischer test was utilized[7]. Results for water content which were in a percentage form were subject to arc sine transformation before statistical analyses.

Results and discussion. The results for condition index (weight/length$^3 \times 1000$), water content, glycogen, protein nitrogen concentration and RNA, DNA concentrations are given in Tables I and II respectively. The changes in the concentrations of glycogen, water and proteins 'insoluble' at low ionic strength were found to be considerably more marked in the white than in the red muscle (Table I). In contrast the 'soluble' protein component of the red muscle was found to be more reduced in percentage terms than in the white muscle. The concentrations of RNA and DNA in non starved fish were found to be considerably higher in the red muscle. Expressed as wet weight of tissue the concentration of RNA and DNA were reduced by similar extents in both muscles. When expressed in terms of dry weight of tissue there was an increase in the concentration of RNA and DNA in the white muscle and a decrease in the red muscle. These changes may in part be influenced by changes in the greater rate of protein mobilisation (Table I) from the white muscle and by changes in water content. The present results are, however, consistent with changes observed in recent histological and ultrastructural studies on the effect of starvation on the red and white muscles of

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Table II. Changes in the concentration of red and white muscle DNA and RNA during starvation

	Wet weight of tissue (μg/mg) Red muscle		White muscle		Dry weight of tissue (μg/mg) Red muscle		White muscle	
	DNA	RNA	DNA	RNA	DNA	RNA	DNA	RNA
Concentration (0 weeks)	0.58 ± 0.01	2.07 ± 0.13	0.26 ± 0.02	1.22 ± 0.09	3.32 ± 0.06	11.74 ± 0.72	1.43 ± 0.13	6.67 ± 0.50
Concentration (30 weeks)	0.30 ± 0.02	0.62 ± 0.05	0.16 ± 0.01	0.43 ± 0.06	2.08 ± 0.23	4.33 ± 0.60	4.76 ± 1.21	11.23 ± 1.93
% Change	—	—	—	—	—	—	+	+
Significance	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.05	< 0.05

The values given represent the means and standard errors of 8 determinations.

teleosts[2,8]. The greater fall in the 'insoluble' protein fraction, which consists largely of myofibrillar and connective tissue proteins, in the white muscle reflects the noticeable atrophy of these fibres during starvation[2,8]. In contrast to the white muscle little degeneration of the red muscle myofibrils was observed at the ultrastructural level in carp starving for 16 weeks[8]. The marked loss of low molecular weight proteins and decrease in RNA concentration in the red muscle probably parallels the considerable degeneration of mitochondria in this muscle during starvation[8]. The changes in RNA and DNA levels found in the plaice would seem to be correlated with the observed loss of euchromatin material from the nuclei of both red and white fibres of the carp[8], but whether this alone could account for the reductions is uncertain.

The present study indicates that the white muscle contractile proteins are preferentially utilized by the fish during starvation. There would, therefore, appear to be a differential response by the two principle muscle types of teleosts to inanition. It seems possible that severe changes in the nutritional state of a fish might influence the division of labour between the myotomal muscles. Such temporal changes are already thought to occur with respect to seasonal changes and migrations[9,10].

Zusammenfassung. Die Wirkung von experimentellem Hunger auf rote und weisse Muskeln der Scholle, *Pleuronectes platessa*, wurde untersucht. In roten wie in weissen Muskeln wurde eine starke Verminderung von Protein und Glycogen gefunden, während der RNS- und DNS-Gehalt in beiden Muskeln herabgesetzt war.

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Free Amino Acids of *Hevea brasiliensis* Latex

Very little information is available on the free amino acids of *Hevea* latex. Former investigations[1-4] showed a very limited number of these compounds present in latex. In the more recent works of SOEI[5] and of D'AUZAC and PUJARNISCLE[6,7], this number has been extended to include most of the classic amino acids. However, the evaluation of the respective quantities of each compound was based on paper chromatography and could therefore only be considered as approximative. The only quantitative results were those concerning the total amino acid content of latex[7,8] and the sum of aspartic and glutamic acids[7]. In the present work, the amino acid composition of both cytoplasmic and lutoidic serums were investigated.

Materials and methods. Two batches of trees of clone PR107 were chosen on experimental plots of the I.R.C.A. (Institut de Recherches sur le Caoutchouc en Afrique), in Bimbresso, Ivory Coast. The trees were tapped, on full spiral, twice a week (d/3, d/4). The latex was allowed to run for 1 min in order to eliminate the fraction containing the organelles which had suffered from the traumatic effects of tapping. A 35 ml subsequent fraction was collected in a tube immersed in ice. Half-an-hour later the latex samples were centrifuged for 30 min at 40,000 rpm (Spinco, rotor 50 Ti) at 0 °C. The tubes were pierced, and the clear cytoplasmic serum was withdrawn. The upper part of the tube was cut and discarded with the rubber. The pellet consisting mainly of lutoid particles was recovered and resuspended in water to which ethanol was immediately added. Ethanol was also added to the cytoplasmic serum to a final concentration of 85%. After breaking of the lutoides and precipitation of the proteins of the 2 fractions (lutoidic and cytoplasmic), the ethanol extracts were centrifuged. All operations were carried out at 2–4 °C.

Free amino acids of the 2 fractions were determined quantitatively with a Technicon Autoanalyzer using the one-column technique and an elution system composed of 3 buffers: pH 2.875, 3.8 and 5.0 forming a continuous gradient[9,10]. The temperature of the column was maintained at 60 °C. Under these conditions the 2 amides (glutamine and asparagine) and threonine run together. As shown by paper chromatography, glutamine and threonine are the ever present constituents of both cytoplasmic and lutoidic serums of *Hevea* latex, whereas asparagine is virtually absent. In order to evaluate each of these 2 compounds separately, the aliquots were hydrolyzed in 1 *N* HCl at 40 °C for 24 h and then chromatographed. Threonine was measured directly. Glutamine evaluation was based on the increase of glutamic acid or on the decrease of the 'threonine' peak.

Results and discussion. The determinations of the free amino acids have been carried out on 16 latex samples coming from 12 trees, taken during the period from July to October 1972. The data in Table I represent the average values of these determinations.

The free amino acid content varies, of course, between the samples taken at a given moment from different trees, as well as the samples taken from the same tree at

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