High Levels of Natural Radiation 1996
Radiation Dose and Health Effects

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Experimental study of the genetic effects of high levels of natural radiation in South-France

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A survey made at the ground level in South-West France (Fig. 1) reveals that the gamma radioactivity ranges generally from 0.001 to 0.030 x 10^{-2} mGy/hr and that dose rates as high as 1 x 10^{-2} mGy/hr are not uncommon.

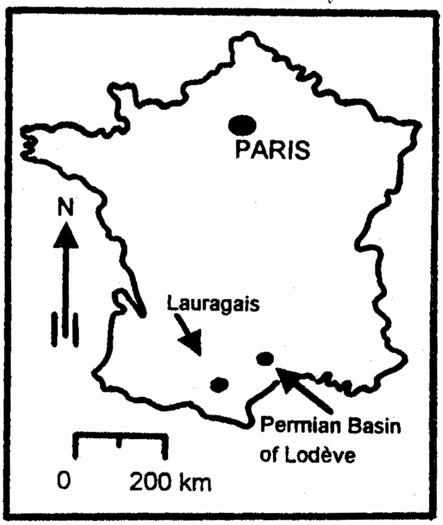

Figure 1. Geographical situation of uranous zones in South France

876 mGy/year

The Hérault Mission of the French Atomic Energy Commission discovered that the dose rate can reach 10×10^{-2} mGylhr at some "hot spots" located in the Pernian Bassin of Lodève and in the Laurageais. The ionizing radiations originate from the two natural uranium isotopes (99.3 % of ^{238}U and 0.7 % of ^{235}U) and from their 28 radioactive daughter products.

Plants that grow on the most radioactive substrates are apparently normal. However, some differences in germination rates and size of the plantlets, changes in color flowers, etc., transmissible to the progeny suggested, that mutagenic events had been produced by site components. To be able to discriminate the effects of the high natural radioactivity from the influence of other environmental factors, experiments were performed with a strain of Tobacco (*Nicotiana tabacum L.*) carrying genetic markers for leave color.

When evaluating the biological effects of high natural radioactivity on persons and animals living in those regions, one of the main difficulties encountered is the exact determination of the doses received and, in addition, to find appropriate controls. In order to overcome those difficulties, we performed observations on laboratory animals maintained in captivity at a "hot spot" in the bassin of the river Lodève. In the hut where the animals were placed, the y dose-rate amounted to about 8×10^{-2} mGylh.

700 mGy/year o.k.

1. EXPERIMENTS WITH A PLANT MARKER : THE α_1^+/α_1 α_2^+/α_2 SYSTEM OF TOBACCO (*Nicotiana tabacum L.*)

1.1. The genetic system [1 -5]

It consists of a double heterozygote α_1^+/α_1 α_2^+/α_2 of *xanthi* variety. The plants carrying this genetic system has greenish-yellow leaves. Either spontaneously or under the action of chemical or physical mutagens, the genetic composition of the system can be modified by mutations in α_1^+ op α_2^+ by reversion in α_1 or α_2, reversions being the most common event. In growing cells, each reverted cell will yield a clone that appears as a green spot in the palisade tissue of the greenish-yellow leaf. Since 1,000 to 2,000 cells can be analyzed per mm^2 of leaf; this marker system is particulary appropriate for detecting effects of low and very low doses of radiation with very good statistical precision.

The genetic effect of a given dose, can be expressed as average reversion rate per cell cycle on the basis of the number of reverted cells or, equivalently, of reverted leaf area. It has been showed that, for a given individual, there exists a simple relation between the totai leaf area *S* and the reverted area *Sg* on one hand, and the reversion rate on the other :

$$p = 1 - \left[\frac{S - Sg}{S}\right]^{1/t}$$

where t is the number of cell cycle that actually took place during the chronic irradiation:

$$t = \frac{\log N}{\log 2} - 7$$

N is the total number of cells observed that corresponds to the total leaf area S with cell density d (number of cells per unit area), so that :

$$N = S \times d$$

Several experiments were performed and the main results were as follows :

1.2. Results [6-10]

The main results of the numerous experiments performed with this system can be summarized as follows:

1.2.1. First experiment in the Permian Bassin of Lodève (Fig.2)

To study the reversion rate per cell cycle, one hundred plants, divided in two

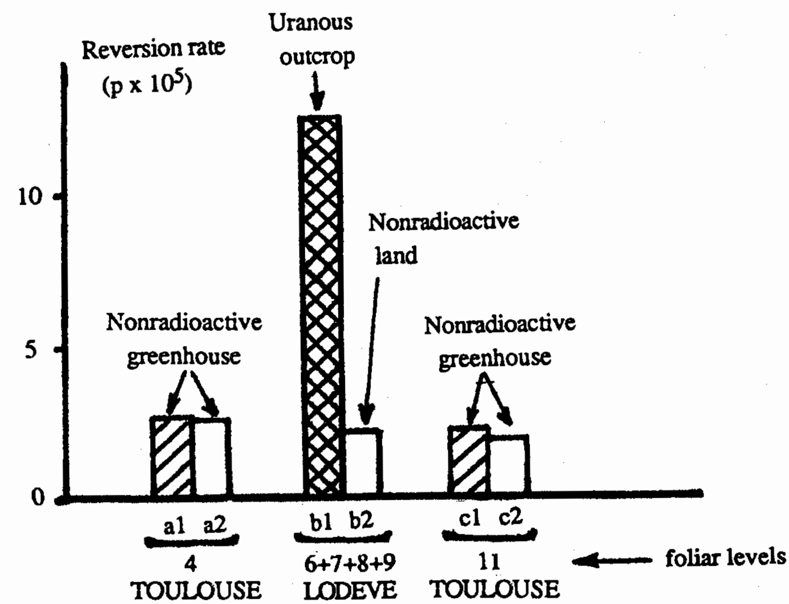

Figure 2. Reversion rates under different conditions of natural radioactivity (Mas d'Alary site)

sets reversion rates in the two sets of 50 pots were maintained in a greenhouse on the University campus in Toulouse and allowed to grow up to four or five leaves. No difference was found between the reversion rates in the two sets.

Thereafter, one set (set 1) was placed at the uranous site of Mas d'Alary where the dose rate ranged between 3 and 10 x 10^{-2} mGy/hr. The second set (set 2), was placed in the vicinity over a substrate yielding a very low dose rate of 0.030 x 10^{-2} mGy/hr considered as a control. Both sets, were allowed to grow up to nine to ten leaves. Whereas, a very large and significant increase in reversion rate was observed in the set 1, no difference was found between the control (set 2) and the plants grown in Toulouse.

Finally, the plants were taken back in Toulouse and placed under the same conditions as before their transfer to Lodève. The plants were allowed to grow a few more leaves. No difference was detected with respect to reversion rates between the latter leaves and the ones produced in the greenhouse at the start of the experiment.

It clearly appears from these data that the reversion rate in the plants maintained in the uranous site, was about five times larger than that of the average control.

1.2.2.Second experiment in the Permian Basin of Lodève (Fig. 3)

To study the dose effect relationship four lots of Tobacco plants were cultivated

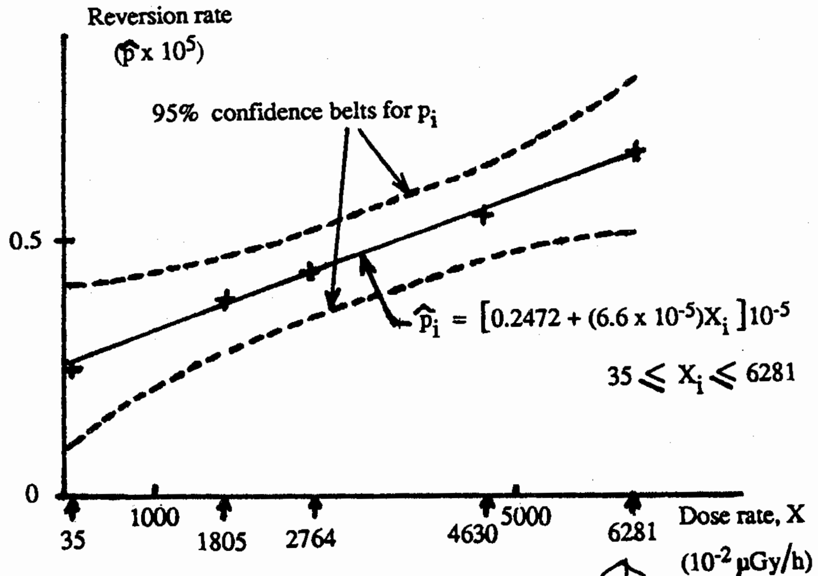

Figure 3. Dose-effect relationship over the Mas d'Alary uranous outcrop in the Permian Basin of Lodève

55021 m Gy/year

in areas yielding the following dose rates :

1.805, 2.764, 4.630 and 6.281 x 10^{-2} mGy/hr.

A fifth lot was placed in the vicinity over a substrate yielding the very low dose of 0.035 x 10^{-2} mGy/hr considered as a control. As previously, the response of the genetic system was expressed as reversion rate per cell cycle. The shape of the response curve was found to be linearly dependent of the dose.

1.2.3 Third experiment in Lauragais (Fig.4)

The study was perforryled at the uranous site of Lagravette where the dose rates are much smaller than in the second experiment above, ie:

0.065, 0.106, 0.179, 0.291 , 0.366 and 0.590 x 10^{-2} mGy/hr.

A threshold in terms of reversion rate per cell cycle was observed as long as the dose rate was no much larger than that from a normal natural background. For the two latter higher dose rates, the reversion rate increased significantly .

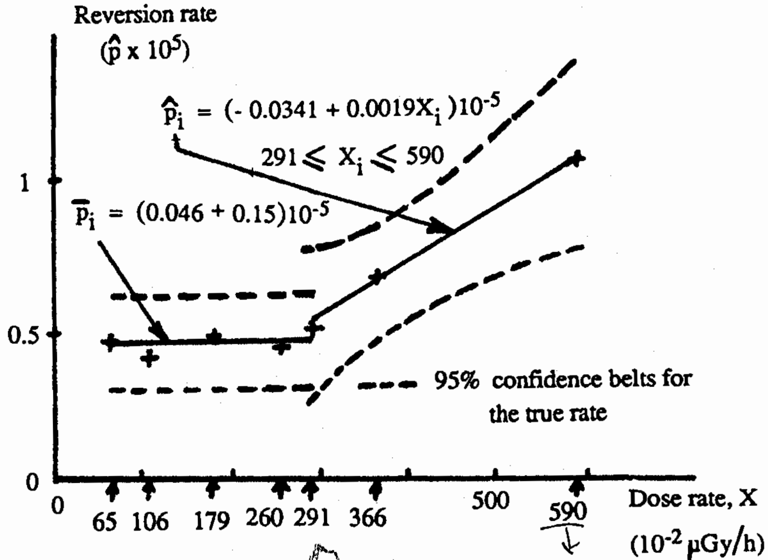

Figure 4. Dose-effect relationship over the Lagravette uranous outcrop in the Lauragais

1.3. Conclusions

1.This marker is very appropriate for detecting *in situ* the genetic effects of low and very low doses of natural radioactivity .

2.There exists a threshold dose rate for the mduction of this type of mutations in Tobacco leaves.

3. From the background to about 0.3 x 10^{-2} mGy/hr (2.63 x 10^{-2} Gy/year), we have a plateau.

4.From 0.3x10^{-2} mGylhr (2.63 x 10^{-2} Gy/year), the response is linearly dependent on the dose.

2. EXPERIMENTS WITH ANIMALS

To study the production of chromosome aberrations in mammalian somatic cells, male laboratory rabbits were kept, for 20 months, in plastic cages placed on the floor and gamma-ray dose determined by individual fluoride dosimeters put around the neck of each rabbit. Blood samples were taken at 4 months intervals to study the occurrence of structural chromosome aberrations in peripheral blood lymphocytes. The effects on male fertility were evaluated on groups of 50 three month old male mice of the BALB/C strain put also in plastic cages with two lithium fluoride dosimeters in each cage. Due to the sensitivity of the mice to low temperatures, the exposure was restricted to the summer period and was repeated with new animals during three successive years. After the exposure, each male mouse was mated in our laboratory to one three month old non-irradiated virgin female of the same strain for a 6 month period. Litter size was recorded at birth and the offspring sexed at weaning 20 days later. To correlate the effects of low doses of ionizing radiation with possible changes in germ cell populations, testicular damage was evaluated at the end of the main period by recording the weight of the testes and by histological examination of 100 carefully selected median cross sections of testicular tubules from one testis of each animal for the presence of spermatogonia, spermatocytes, spermatids and spermatozoa. This method provides mainly a qualitative analysis and allows only a semi-quantitative asssessment because germ cells may be numerous or searce in a given cross section. Meiotic preparations were made from the second testis of each animal and one hundred spermatocytes at the diakinesis-first metaphase stage of meiosis were analysed for the presence of reciprocal translocations.

Control rabblts and control mice maintained near the radioactive site under comparable conditions were handled and sampled in the same exposed animals.

2.1.Results

During the 20 months the exposed rabbits received from 36,300 up to 130,750 mGy according to the locations of their cages in the hut whereas the doses received during the same period by the controls arrounded to 2,400 mGy (Table 1).

Table 1
Dose(10^{-2}mGy) or γ-radiation recieved by the rabbits

Treatment	Rabbit n°	Duration of exposure (months)				
		4	8	12	16	20
Control	1	65	102	152	207	254
	2	40	95	140	195	240
	3	50	110	158	205	235
Irradiated	4	26,500	52,750	70,750	101,750	130,750
	5	22,300	44,300	57,300	76,800	95,800
	6	22,500	43,250	56,000	73,500	91,500
	7	13,000	26,500	38,700	51,200	*
	8	7,800	14,800	20,500	28,500	36,300

*Rabbit 7 died after 19 months

Figure 5 shows that the yield of structural chromosome anomalies typical of an exposure to ionizing radiations (chromosome fragments, dicentric chromosomes) increases initially but disappear completely after 20 months.

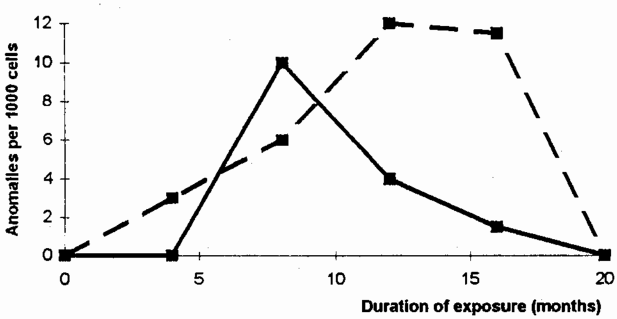

Figure 5. Relationship between duration of exposure and dicentrics (—) and cromosome fragments(-----) per 1,000 cells in rabbit peripheral blood lymphocytes

The male mice maintained in the area of high natural radioactivity received from 13,800 mrad up to 63,250 mrad and the concurrent controls from 34 to 127 mrad according to the duration of exposure (Table 2).

Table2
Doses of gamma radiation(mrad) recieved by the male mice

Experiment	Duration of exposure	Control	Irradiated
1 st year	3 months	34	13,800
2nd year	4 months	68	15,000
3rd year	6 months	127	45,080
			63,250

The number of litters and of offspring sired after exposure during 6 month period was clearly related to the dose of γ-irradiation received during the exposure period (Fig. 6). Both values increase in a dose-related way up to 45,080 and fall abruptly for the animals receiving 63,250 mrad.

The histological and cytological studies performed on the testes at the end of the mating period did not reveal any difference between the control and the exposed males (Table 3).

increase of fertility

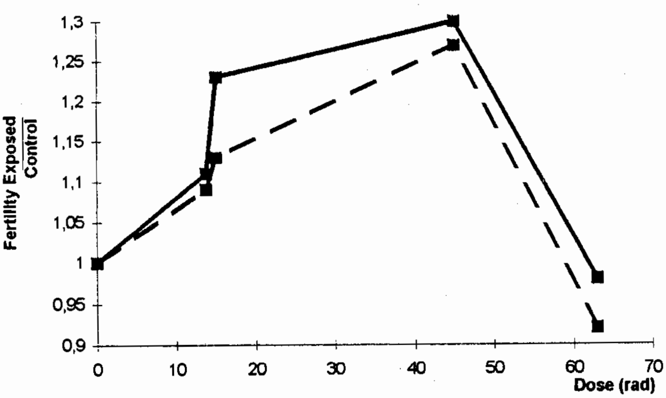

Figure 6. Relation between dose recieved and litters(---) and offspring(—) sired by male mice

Table 3
Results of the cytological and histlogical observations performed on male mice

Experiment	Dose received (cGy)	Weight of the testes (mg)	% of abnormal spermatocytes	Histological results Cross sections with			
				Spermatogonia (%)	Spermatocytes (%)	Spermatids (%)	Spermatozoa (%)
I	0.034	262	0	100	100	100	90
	13.8	270	0	100	100	100	90
II	0.068	264	0	100	100	100	87
	15.000	269	0	100	100	100	88
	0.127	254	0	100	100	100	88
III	45.08	268	0	100	100	100	90
	63.25	265	0	100	100	100	91

2.2. Discussion and conclusions

Our observations confirm that high natural radioactivity can result in the production of structural aberrations in mammals living in those areas. The drastic decrease in the incidence of anomalies occuring after a few months results probably from a selective elimination of the most sensitive stem-cells combined to the fact that the life-span of the rabbit peripheral blood lymphocytes does not exceed 2 months [11]. A short life-span of peripheral blood lymphocytes carrying unstable structural chromosome aberrations appears a common characteristics of the majority of mammals [12] and contrasts greatly with values exceeding three years reported in man it can be inferred, therefore, that one cannot consider that cytogenetic observations performed on wild mammals living in area of high radioactivity (natural or resulting from nuclear assays) would provide reliable informations on the doses received by the animals and on the level of damage produced to their genetic material.

The increase in male fertility observed for animals receiving up to 43,000 mrad could rpresent an example radiation hormesis (13) : stimulating effects of small doses of ionizing radiations is a controversial topic but it should be stressed that our results on the male mice are in agreement with the observations of Newcombe and [14] and [15] who reported an increase of embryo production after low doses of ionizing radiations administered to trout spermatozoa.

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