

TOXICOLOGICAL AND MOLECULAR ANALYSIS OF WORKERS GAS STATIONS EXPOSED TO THE BENZENE IN RIO DE JANEIRO

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INTRODUCTION

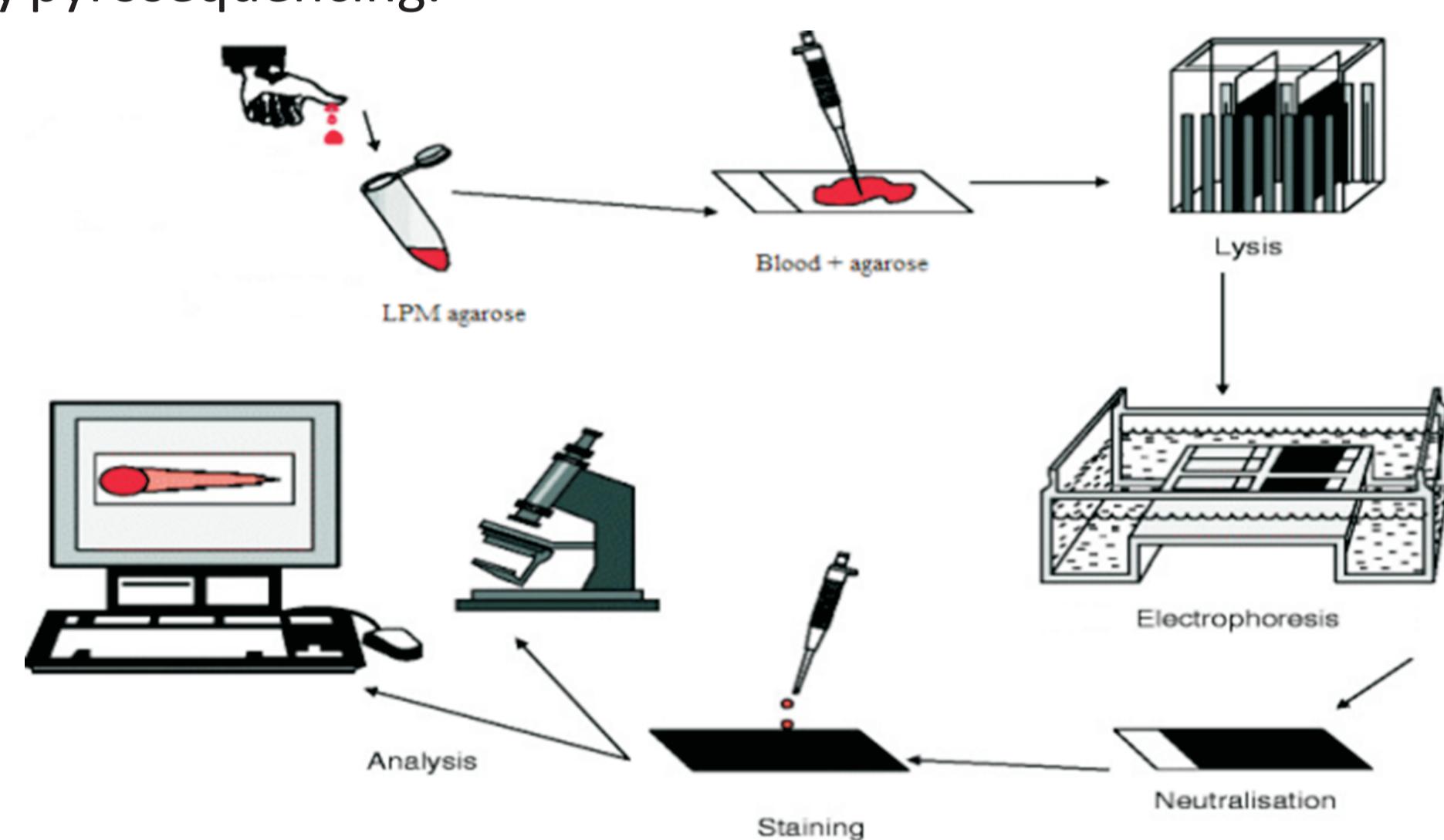
Benzene is a polycyclic aromatic hydrocarbon classified by the *International Agency for Research on Cancer* (IARC) as carcinogenic to human beings, presenting toxicity to hematopoietic and central nervous systems. This compound is present in gasoline, resulting in the exposure of millions of people who work and circulate in gas stations. Therefore it is crucial to monitor the health conditions of petrol station employees to access potential benzene damage to them.

OBJECTIVE

To evaluate potential health indicators and markers of benzene toxicity in gas station employees exposed to benzene present in gasoline at petrol stations in Rio de Janeiro, investigating DNA lesions by Comet Assay (genotoxic effects), the methylation profile of the transposable elements, and of the genes involved in cell proliferation control and DNA repair.

METHODOLOGY

A cross-sectional study carried out on workers at petrol stations, either occupationally exposed or not to benzene, from Downtown and South Zones of Rio de Janeiro city. All participants are older than 18 years old and signed an informed consent form prior to study enrollment. Clinical, demographic and economic data were obtained by answering a standardized questionnaire. Blood samples were collected to evaluate the hematological and biochemical alterations, the genotoxic and epigenetic (methylation) effects. Hemogram and automated biochemical analysis were performed according to the hemogram protocol of the hematology section of the clinical pathology laboratory from HCl/INCA. The genotoxic effects were evaluated by the Comet Assay, following the FIOCRUZ institutional protocol. Epigenetic alterations will be evaluated through the analyses of the methylation profile of the transposable elements (*LINE1* and *Alu*) and *p15*, *p16*, *PARP-1* and *MSH3*, by pyrosequencing.



RESULTS

A total of 406 benzene occupationally exposed workers were interviewed. 323 samples were collected of which 210 were directly exposed (attendants, managers) and 113 were exposed indirectly (convenience stores), whereas 166 office labor individuals were considered as the control group.

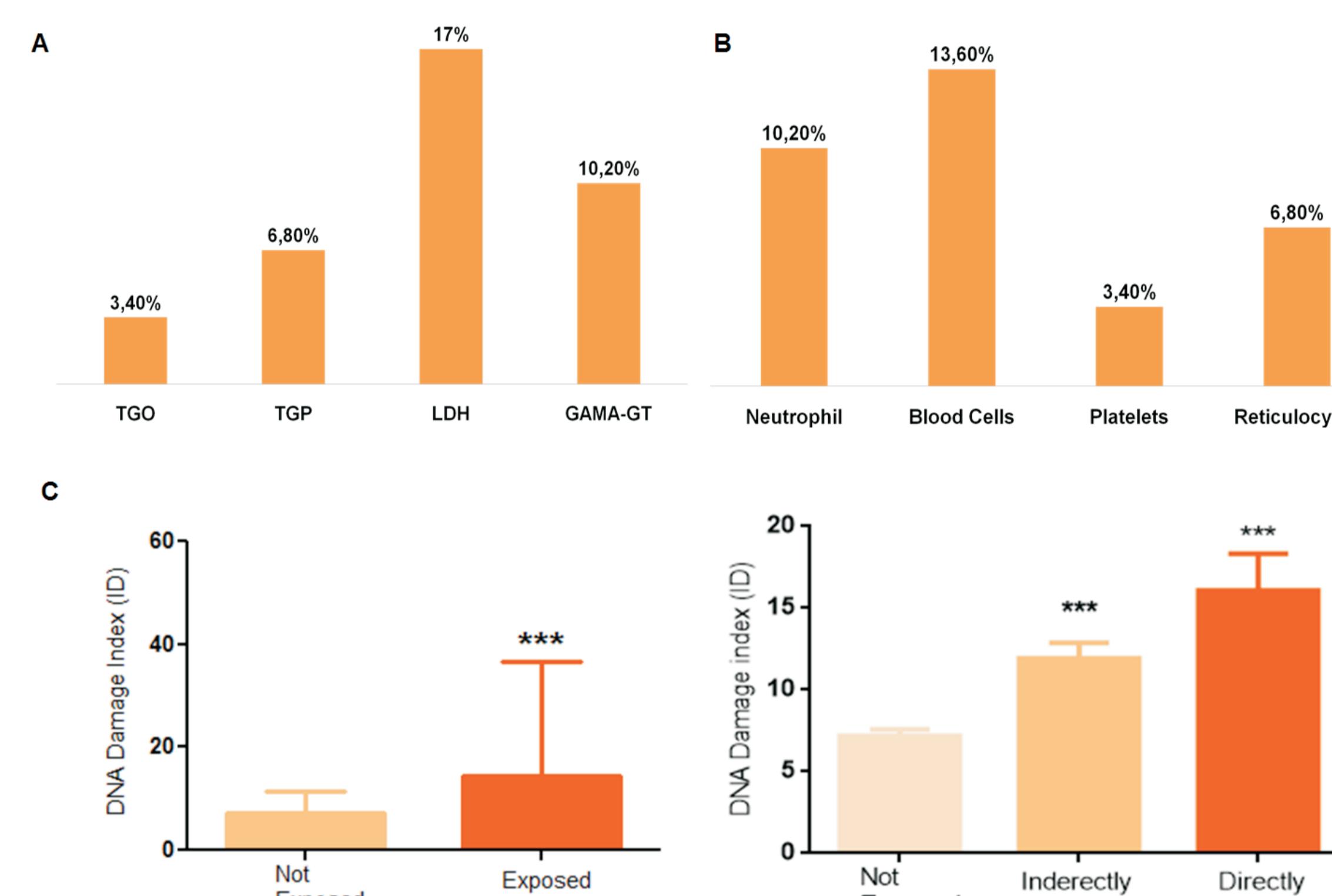
Table 1 - Demographic and socioeconomic characteristics of workers exposed and not exposed to benzene by means of gasoline.

Demographic characteristics	Directly Exposed		Indirectly Exposed		Not exposed		p Value	
	N	%	N	%	N	%		
Gender	Male	212	88,3	49	31,6	87	47,5	<0,01
	Female	28	11,7	106	68,4	96	52,5	
Age	Up to 24 years	35	14,6	51	33,3	06	3,3	<0,01
	From 25 to 34 years	74	31,0	51	33,3	54	29,5	
Skin color	35 years and over	130	54,4	51	33,3	123	67,2	<0,01
	Not white	187	77,9	118	76,1	101	55,2	
Marital Status	White	53	22,1	37	23,9	82	44,8	<0,01
	Married	143	59,6	60	38,7	94	51,4	
Education	Widowed or Divorced	23	9,6	11	7,1	16	8,7	<0,01
	Single	74	30,8	84	54,2	73	39,9	
Financial Support: PPSUS / Faperj; OPAS; INCA / MS	Elementary School	66	27,5	39	25,2	12	6,6	<0,01
	High school	157	65,4	100	64,5	32	17,5	
	Higher education	17	7,1	16	10,3	139	76,0	

Table 2 - Means and standard deviation of hematological and biochemical parameters in workers from the group not exposed and exposed directly and indirectly to the benzene that performed blood collection. Reference values based on the Brazilian Society of Hematology (SBH).

Biochemical Parameters	Not Exposed (Mean e DP)	Indirectly Exposed (Méan e DP)	Directly Exposed (Mean e DP)	p-Value	Reference Value
TGO (UI/L)	17,00(- 7,37)	23,28(- 8,35)	21,32(- 8,62)	0,683	Até 40
TGP (UI/L)	20,00(- 14,32)	27,34(- 15,61)	21,37(- 16,30)	0,340	Até 41
Gama-GT (UI/L)	25,00(- 28,60)	48,91(- 75,93)	31,27(- 36,13)	0,738	10 a 71
Creatinine (mg/dl)	0,80(- 0,96)	1,00(- 0,21)	0,90(- 0,18)	0,478	0,3 – 1,3
Total Bilirubin (mg/dl)	0,39(- 0,39)	0,43(- 0,21)	0,41(- 0,17)	0,042	Até 1,2
Direct Bilirubin (mg/dl)	0,16(- 0,10)	0,17(- 0,07)	0,13(- 0,05)	0,777	Até 0,2
Indirect Bilirubin (mg/dl)	0,21(- 0,13)	0,29(- 0,15)	0,22(- 0,13)	0,935	Até 0,8
LDH (UI/L)	351(- 103,16)	384(- 73,25)	362,0(- 109,23)	0,005	240 à 480
C-reactive Protein (mg/dl)	0,19(- 0,22)	0,16(- 1,60)	0,21(- 0,40)	0,884	<0,5
Hematological Parameters					
Red blood cells (milhões/	4,70(- 0,95)	5,04(- 0,45)	4,60(- 0,52)	0,311	4.50-6.50
Hemoglobin (g/dl)	13,90(- 2,94)	14,7(- 1,10)	13,10(- 1,10)	0,608	13.5-18
Hematocrit (%)	42,10(- 10,55)	44,60(- 3,41)	40,40(- 3,16)	0,776	40-54
VGM (fL)	89,10(- 16,75)	88,7(- 5,36)	89,20(- 16,03)	0,011	76-96
HGM (fL)	29,70 (- 43,12)	29,7(- 2,26)	36,70(- 2,30)	0,818	27-32
CHGM (g/dL)	33,10 (- 51,45)	33,15(- 1,17)	32,70(- 1,14)	0,916	32-36
	7145 (- 2266)	6985(- 2012)	7558(- 1947)	0,028	4000 – 10000
Leukocytes(mm ³)					
Neutrophil (uL)	4953(- 1666)	5094(- 995,4)	5265(- 874,2)	0,066	1600-7500
Eosinophils (uL)	1,80 (- 2,88)	2,30 (- 3,72)	2,72 (- 3,96)	0,466	40 - 600
Basophils (uL)	0,43 (- 0,24)	0,46 (- 0,68)	0,52 (- 0,90)	0,029	0- 100
Typical lymphocyte (uL)	29,33 (- 23,90)	31,16 (- 15,92)	31,59 (- 27,64)	0,139	800 – 4500
Monocytes (uL)	4,69 (- 2,66)	7,40 (- 2,99)	6,65(- 1,84)	0,651	80 – 100
Platelets (mil/uL)	242 (- 76,77)	247 (- 60,56)	267 (- 56,79)	0,369	150 – 400
Reticulocytes (%)	0,99(- 0,59)	1,18(- 0,32)	1,29(- 0,49)	0,392	0,5 – 2,00

The frequency of main altered parameters on the group exposed to benzene at gas station.



Altered Biochemical (A) and Hematological (B) parameters on exposed group and DNA damage index (DI) (C) in workers exposed (directly or indirectly) or not to benzene at gas stations using Comet Assay. ***p<0,001

Table 3 - Distribution in the percentage of the class of DNA damage in the participants with direct, indirect occupational exposure and without exposure to gasoline.

DNA Damage Class	Groups		
	Not Exposed (n=100)	Indirectly Exposed (n=100)	Directly Exposed (n=150)
Class 0	95,5±0,2	92,6±0,6	89,9±1,4
Class 1	4,2±0,2	7,1±0,6	9,6±1,3
Class 2	0,2±0,0	0,2±0,0	0,4±0,1
Class 3	0,1±0,0	0,1±0,0	0,1±0,0

CONCLUSION

This work has shown that the exposure to benzene in workers at gas stations causes different damages, such as genetic, hematological and biochemical changes leading to a health damage in these workers.

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Projeto Gráfico: Serviço de Edição e Informação Técnico-Científica / INCA