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Vitamin A Status of Populations in Three West African Countries

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Summary: The results of food consumption surveys, epidemiological surveys (clinical and biochemical) undertaken in three sub-sahelian countries have established:

- the existence of xerophthalmia in Upper Volta
- a transitory vitamin A deficiency during the dry season in South Mali, without serious clinical signs
- a good vitamin A status in Casamance, in South Senegal.

A combined biochemical indicator which takes account of plasma carotenoids and vitamin A is recommended to assess the importance of vitamin A deficiency in a population.

Introduction

Few epidemiological surveys have been carried out in the Sahel region to assess vitamin A status of populations. Some clinical surveys have been reported [14, 25]. These surveys are rarely representative of the whole population of considered regions. Several authors have shown that measles is frequently associated with xerophthalmia [10, 23, 31]. Few results of plasma carotenoids and vitamin A in populations of the Sahel have been published [7, 14].

Our objective has been to evaluate the vitamin A status of populations by clinical examination and blood determinations in areas where vitamin A deficiency may exist. The results of the study undertaken in Senegal will be discussed parallelly to those of the food consumption surveys carried out during the same period:

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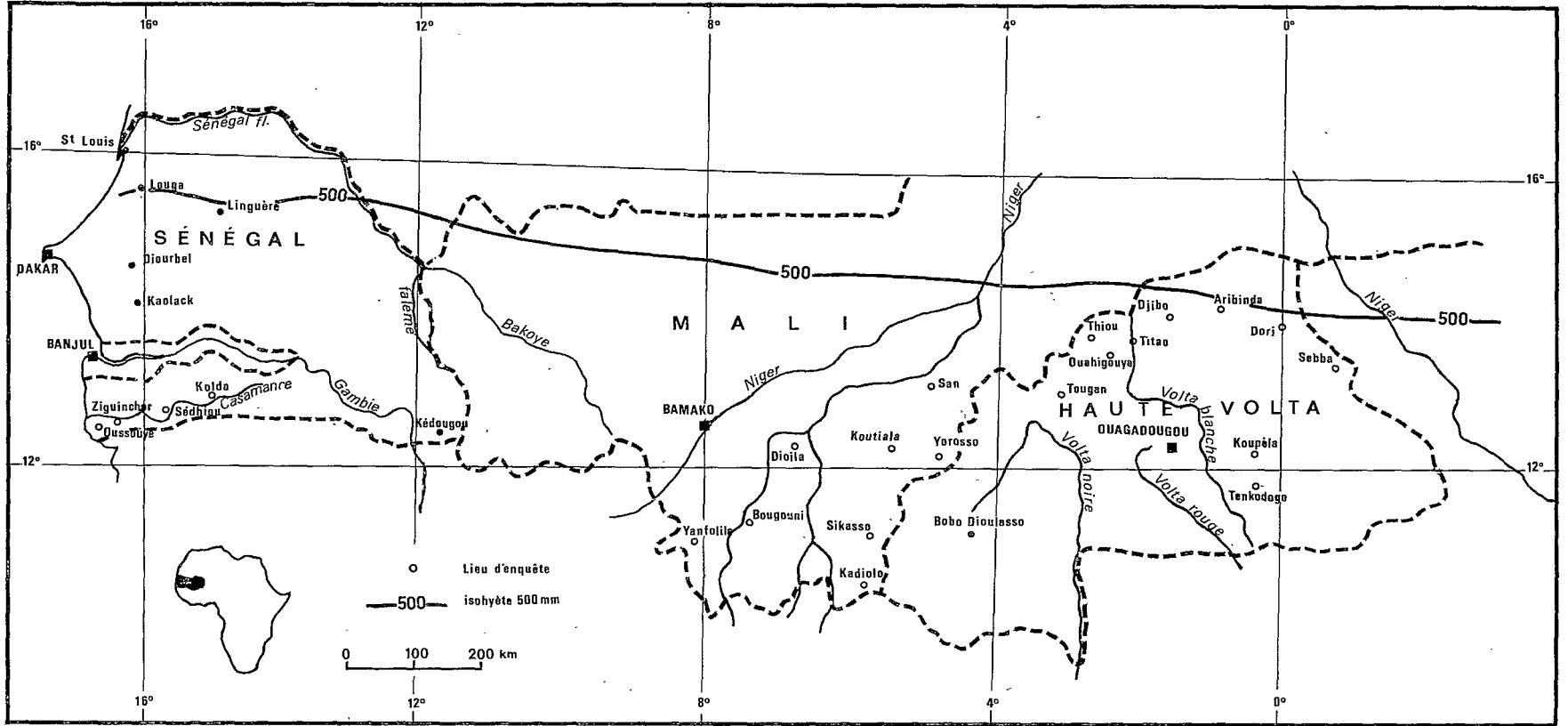
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Fig. 1: Geographical situation of the principal villages.



Methods

a) Food consumption surveys

Household food consumption of selected families was measured during 3 to 5 days by weighing the foods. Meals basically consist of a cereal (rice or millet) with a sauce. For the calculations we used the recommended daily intakes of FAO [8] and primarily the food composition table for Africa [9]. The results of these food consumption surveys have already been published [6].

b) Clinical and biochemical surveys

We represent on figure 1 the geographical situation of the principal villages. The isohyet 500 mm which is marked corresponds to that of the period preceding the recent years of drought.

The examined individuals were obtained by random sampling with two levels on the basis of the results of the last census. The fraction of the sampling was 1/1000; firstly the villages were chosen at random and secondly the families. The global results of these nutritional surveys were published elsewhere [4]. Clinical signs of xerophthalmia were assessed according to the recommendations of WHO [21].

Veinous blood samples were collected from the arm with evacuated blood collecting tubes. Few samples were taken in preschool children because of technical difficulties. The plasma was centrifuged on the field, transferred in polypropylene tubes and kept in liquid nitrogen. The samples were stored at -70°C in the laboratory.

Plasma carotenoids and vitamin A were determined on average 3 months later. Preliminary assays had not shown any loss of carotenoids and vitamin A after several months of storage at -70°C and a unique thawing.

Plasma carotenoids and vitamin A were assayed by the colorimetric method of NEELE and PEARSON [20] using trifluoroacetic acid. The samples were saponified and then extracted with hexane as ROELS *et al.* [27] recommended. We did not notice any loss of vitamin A in our standard curve due to the saponification. The samples were usually determined once only. Vitamin A standards prepared with capsules of USP reference standard of vitamin A¹ were added to every batch of samples. The carotene standard curve was prepared with synthetic β -carotene Merck. In our conditions, the coefficients for the calculations were:

$$\begin{aligned} \text{Carotenoids} & \quad (\mu\text{g}/\text{dl}) = 742 \text{ DO}_{450} \\ \text{Vitamin A} & \quad (\mu\text{g}/\text{dl}) = 292 (\text{DO}_{620} - 0.169 \text{ DO}_{450}) \end{aligned}$$

The calculations were processed with a IBM 370 computer of the Ministry of Finance (Dakar) with Fortran language for the food consumption surveys and Osiris programs for the other studies.

Results

a) Food consumption surveys

The results of the food consumption surveys undertaken in Senegal are reported in table I. Vitamin A requirement is satisfied in the regions of Diourbel and Casamance. On the contrary, the requirement is only partially covered at 54% in Louga, 40% in Linguere, at the end of the dry season (two towns located in the north of the country, fig. 1). On average, the requirement of vitamin A is covered at 81% in Dakar, 87% in Kedougou.

The role of provitamin A in the diet is important because 84 to 98% of ingested vitamin A is of vegetable origin, that is as carotenoids; vegetables and

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Tab. I: Results of the food consumption surveys in Senegal

Region and date of survey ¹	Duration and total cases	Average requirement ²	Average intake ²	% of requirement covered	% of individuals with their requirement covered at less than		% of vitamin A of vegetable origin	
					50 %	100 %		
<i>Dakar</i> May - July 1977	3 days 88 families 1031 individuals	634	516	81	49 %	81 %	91 %	vegetables 51 % fruits 6 % oil 28 %
<i>Kedougou</i> June - July 1977	5 days 69 families 666 individuals	614	533	87	29 %	68 %	98 %	vegetables 87 % fruits 5 % oil 3 %
<i>Louga</i> March - May 1978	5 days 87 families 1102 individuals	608	328	54	60 %	93 %	84 %	vegetables 64 % fruits 6 % oil 13 %
<i>Linguere</i> March - May 1978	5 days 25 families 280 individuals	611	244	40	91 %	100 %	90 %	vegetables 47 % fruits 13 % oil 28 %
<i>Diourbel</i> July 1979	3 days 88 families 987 individuals	620	882	142	5 %	38 %	97 %	vegetables 89 % fruits 4 % oil 3 %
<i>Casamance</i> July 1979	3 days 66 families 912 individuals	626	2425	387	18 %	37 %	98 %	vegetables 12 % fruits 10 % oil 75 %

¹ These surveys have been carried out for the Ministry of Planification. The first 4 for the FAO planification project the last 2 for the nutrition project of the World Bank.

² µg of retinol/day/average individual.

fruits supply the main part of vitamin A, except in Casamance, where palm oil provides $\frac{3}{4}$ of the vitamin A intake.

The cover of the energetic requirement as lipids is important as the percentage of calories of fat origin varies from 20 to 34% between the regions.

b) Clinical surveys

We have summarized in table II the results of the clinical surveys.

Tab. II: Results of the clinical surveys

Survey	Season	Number of individuals			Clinical signs xerophthalmia
		Children		Adults	
		0-5 yrs	6-14 yrs		
<i>Upper Volta</i> March - April 1978	dry season	197	318	492	- 1 girl aged 2 yrs with conjunctival xerosis (XIA) - 1 boy aged 3 yrs with keratomala- cia (X3B) and marasmus
<i>South Mali</i> July - August 1978	rainy season	118	149	278	-
<i>South Mali</i> March - April 1979	dry season	300	293	548	-
<i>Senegal</i> Casamance Nov. - Dec. 1979	dry season	248	328	686	- 1 woman aged 40 yrs with corneal scars (XS)

- The main nutritional problems affecting the population at risk (0-5 yrs) are:
- anaemia which affects from 30 to 57% of the children according to the region (hemoglobin level less than 11 g/dl)
 - protein-energy malnutrition: 8 to 17% of the preschool children were found to have a weight for height less than 80% of the standard:

Tab. III: Results of the plasma determinations of vitamin A ($\mu\text{g}/\text{dl}$)¹

Country and age	Mean \pm S.D. (number of cases)	Range	Vitamin A level ²				% of individuals with deficient vitamin A level	% of individuals with deficient vitamin A level and low carotenoid level
			0-9 deficient	10-19 low	20-49 acceptable	\geq 50 high		
<i>Upper Volta</i>								
0- 5 years	16.6 \pm 7.1 (17)	8-32	3	9	5	0	3/17	3/17
6-14 years	17.5 \pm 6.7 (178)	7-36	13	105	60	0	7%	7%
Adults	24.9 \pm 11.6 (316)	4-86	12	110	184	10	4%	3%
Pregnant or lactating women	19.9 \pm 9.4 (78)	4-53	5	40	31	2	6%	5%
<i>South Mali</i>								
rainy season								
0- 5 years	16.5 \pm 7.5 (16)	8-35	2	10	4	0	2/16	2/16
6-14 years	19.1 \pm 6.5 (119)	10-54	0	69	49	1	0	0
Adults	24.2 \pm 8.1 (253)	4-56	7	69	175	2	3%	2%
Pregnant or lactating women	22.0 \pm 6.5 (60)	9-36	1	22	37	0	2%	2%
<i>South Mali</i>								
dry season								
0- 5 years	16.7 \pm 6.6 (57)	3-34	8	33	16	0	14%	14%
6-14 years	21.1 \pm 7.2 (197)	8-53	1	89	106	1	1%	1%
Adults	27.3 \pm 10.4 (388)	3-69	13	75	292	8	3%	3%
Pregnant or lactating women	24.2 \pm 10.2 (92)	6-69	4	28	59	1	4%	4%
<i>Senegal</i>								
Casamance								
0- 5 years	20.0 \pm 7.0 (51)	7-38	3	20	28	0	6%	0
6-14 years	22.4 \pm 7.8 (231)	6-50	4	92	134	1	2%	0
Adults	31.2 \pm 11.1 (620)	4-86	4	62	515	39	1%	0
Pregnant or lactating women	29.8 \pm 9.0 (113)	11-55	0	11	98	4	0	0

¹ The median has not been marked because it is very close to the mean value.

² ICNND [1963].

Tab. IV: Results of the plasma determinations of carotenoids ($\mu\text{g/dl}$)

Country and age ¹	Mean \pm S.D. (number of cases)	Range	Median	Carotenoid level ²				% of individuals with low carotenoid level
				0-19 deficient	20-39 low	40-99 acceptable	\geq 100 high	
<i>Upper Volta</i>								
0- 5 years	29 \pm 18 (17)	7- 69	23	6	6	5	0	12/17
6-14 years	34 \pm 20 (178)	8- 164	29	33	90	54	1	69 %
adults	36 \pm 18 (318)	6- 151	32	50	164	98	6	67 %
<i>South Mali</i>								
rainy season								
0- 5 years	40 \pm 17 (16)	13- 74	38	2	7	7	0	9/16
6-14 years	55 \pm 32 (119)	14- 168	46	6	43	55	15	41 %
adults	50 \pm 28 (253)	7- 179	41	14	101	119	19	45 %
<i>South Mali</i>								
dry season								
0- 5 years	36 \pm 24 (57)	7- 152	29	13	27	16	1	70 %
6-14 years	41 \pm 25 (197)	5- 146	34	26	95	70	6	61 %
adults	32 \pm 34 (388)	4- 333	23	138	172	67	11	80 %
<i>Senegal</i>								
Casamance								
0- 5 years	174 \pm 142 (51)	30- 732	145	0	2	16	33	4 %
6-14 years	224 \pm 151 (231)	41-1154	182	0	0	41	190	0
adults	248 \pm 163 (621)	22-1221	199	0	6	60	555	1 %

¹ We have not included a separate category for the pregnant or lactating women as their carotenoid levels are no different from those of the other women.

² ICNND [1963].

c) Biochemical determinations

Carotenoids and vitamin A increase with age (tab. III, IV). According to ICNND [1963], the average levels of vitamin A are low in the children of Upper Volta and Mali but acceptable in adults; they are also acceptable in Senegal (tab. III). The average carotenoid levels are low during the dry season in Upper Volta and Mali; they are acceptable in Mali during the rainy season and high in Senegal (tab. IV). The percentages of individuals with low carotenoid levels give comparable results.

This difference in plasma carotenoids is explained by a difference in food consumption. Upper Volta and South Mali are indeed partially in the Sahel where vegetables and fruits are rare, especially during the dry season. On the contrary in Casamance, near the Guinea zone, the vegetation is much more luxuriant. Vegetables, fruits and above all palm oil very rich in carotenoids are available nearly all the year long, which explains the elevated carotenoid levels.

There is a problem of vitamin A deficiency in a population when more than 5% of the preschool children have vitamin A levels lower than 10 µg/dl [21].

By examining the results of the Casamance survey, we see that 6% of the preschool children have deficient levels of vitamin A (tab. III). We could conclude that there exists a problem of hypovitaminosis A in this region. However, this conclusion would be contradictory with the results of the food consumption and clinical surveys.

Therefore we suggest a new biochemical indicator to assess vitamin A status in a population. Individuals who have deficient levels of vitamin A (less than 10 µg/dl) and low levels of carotenoids (less than 40 µg/dl) will be considered as deficient in vitamin A. We will justify the utilisation of this indicator in the discussion.

By using this combined indicator for our results (tab. III), we can see that vitamin A deficiency exists in Upper Volta and Mali in the dry season, but that it is not a problem in South Mali in the rainy season. Hypovitaminosis A is then totally absent from the south of Senegal. These results are strengthened by the clinical surveys which showed 2 cases of xerophthalmia in Upper Volta (tab. II). On the other hand, in South Mali, no cases were seen, neither in the dry season, nor in the rainy season. (We did not evaluate the incidence of night blindness – clinical sign not very objective in areas of onchocerciasis).

Therefore hypovitaminosis A which occurs in South Mali in the dry season is transitory as it does not appear to give any serious sign of xerophthalmia. At the beginning of the rainy season, the intake of carotenoid rich foods such as mangoes would allow to restore the level of the vitamin A liver stores.

We have noticed a significant liaison between plasma carotenoids and vitamin A levels (tab. V). The coefficient of correlation higher in children may mean

that their vitamin A status is more dependent on their carotenoid intake and that their liver stores of vitamin A are lower than in the adults. A liaison between vitamin A and hemoglobin or plasma iron levels has also been observed. Vitamin A is carried in the plasma bound to RBP, which is also bound to prealbumin, that is why the correlation between prealbumin and vitamin A is so high (tab. V).

Tab. V: Correlation coefficients between vitamin A and some other plasma parameters

Country and age		Correlation coefficient between vitamin A and:			
		Carotenoids ¹	Prealbumin	Hemoglobin	Iron ¹
<i>Upper Volta</i>	194 children	0.520 ***	0.558 ***	0.228 **	0.188 **
	316 adults	0.251 ***	0.496 ***	0.270 ***	0.205 ***
<i>South Mali</i> rainy season	135 children	0.550 ***	0.681 ***	0.303 ***	0.088 NS
	253 adults	0.219 ***	0.720 ***	0.164 *	0.010 NS
<i>South Mali</i> dry season	254 children	0.524 ***	0.790 ***	0.094 NS	0.045 NS
	388 adults	0.361 ***	0.766 ***	0.293 ***	0.230 ***
<i>Senegal</i> Casamance	282 children	0.188 **	-	0.276 ***	0.153 *
	620 adults	0.170 ***	-	0.286 ***	0.241 ***

¹ Plasma carotenoid and iron levels have been transformed into logarithm to obtain a normal distribution.

Statistical significance: NS = not significant, * P < 0.05, ** P < 0.01, *** P < 0.001.

Discussion

In Black Africa and in many developing countries where vitamin A deficiency can be found, most of the vitamin A intake is of vegetable origin, that is as carotenoids [22, 24, 29, 32].

Plasma carotenoids are a good indicator of the recent dietary intake of carotenoids and therefore, indirectly of the vitamin A status.

Furthermore, in Africa vitamin A deficiency does not exist in forest areas where carotenoid intake and therefore plasma carotenoids are high. On the contrary, xerophthalmia is found in savanna regions where carotenoid intake and plasma levels are low.

For these reasons, we recommend a combined biochemical indicator for vitamin A deficiency which takes account of carotenoids. We will consider as possibly deficient in vitamin A the individuals with plasma vitamin A less than 10 µg/dl and plasma carotenoids less than 40 µg/dl.

SAUBERLICH *et al.* [30] noticed that when low serum β-carotene levels are found in association with low serum vitamin A levels, the evidence for inad-

equate vitamin A nutriture is quite strong. We use total carotenoids in this indicator, there is therefore a certain error but which should be allowed because in this way this indicator can be easily used in epidemiologic surveys.

This combined indicator would also have the advantage to eliminate the individuals with a deficient level of vitamin A for another reason than an insufficient intake. Indeed, subjects with deficient vitamin A and acceptable or high carotenoid levels are sometimes found, either because of a defective determination or a transitory deficiency of transport.

When carotenoids and vitamin A are assayed simultaneously by a colorimetric method, AWDEH [3] showed that the vitamin A concentration may be falsely lowered if carotenoid level is high. This is why this author recommends a chromatographic separation of carotenoids before dosing the vitamin A. This does not seem very realizable to us for epidemiologic surveys, but by using the combined indicator, it is possible to eliminate the most erroneous results.

Besides the vitamin A level is often transitorily impaired during infections in general [2, 19], measles [18, 33]; although carotenoid level decreases less during infectious diseases [2, 18, 19, 26]. This could also justify the utilization of this combined indicator.

These surveys have shown that xerophthalmia exists in Upper Volta; the plasma determinations confirmed vitamin A deficiency in this population. RAOULT [25] also saw cases of xerophthalmia in this country. Furthermore, BLEIBERG [5] noted that during the dry season, only 45% of the vitamin A requirement is satisfied in a group of voltaic children. Vitamin A intake is low in Upper Volta and in the north of Mali where cases of xerophthalmia are found [22].

In South Mali we have established a seasonal hypovitaminosis A, perceptible on the biochemical level but which does not appear clinically. In a damp area (Casamance), the vitamin A status of the population is good, owing to an abundant intake of provitamin A in the diet.

INGENBLEEK and DE VISSCHER [13] found acceptable and high average vitamin A levels varying from 43 to 67 $\mu\text{g}/\text{dl}$ in the inhabitants of Casamance 7-21 years old. Our results are lower, perhaps because we did not use the spectrophotometric method.

In South Cameroon, in forest areas, we also observed a good vitamin A status due to high carotenoid levels [1]; only 1.6% of the 257 preschool children examined had a deficient vitamin A level [15]. If we use this combined indicator, no child can be considered as deficient in vitamin A in this population.

PATWARDHAN [24] only, found a strong correlation between seric carotenoids and vitamin A by collecting 48 average values of surveys which included 5520 subjects from 11 countries: $r = 0.755$. We have also noticed a highly significant liaison between these two parameters in our surveys in Upper Volta

and Mali, except in Casamance. The common point between these studies is that they were undertaken in areas where vitamin A intake is mainly provided by carotenoids; the difference is that vitamin A intake may be limited in Upper Volta and Mali, but not in Casamance.

Besides, in 257 preschool children in South Cameroon we also noticed a high correlation between carotenoid and vitamin A levels: $r = 0.526$ [15].

Why is xerophthalmia relatively rare in Africa compared to Asia? From our studies we think that the vitamin A intake is generally sufficient, at least during a specific period of the year – the rainy season.

Prealbumin serves as a limiting factor for RBP binding which in turn acts as the limiting factor for retinol transport [12]. Then a deficiency of these proteins may lead to an inadequate transport of retinol.

The minimal theoretical quantity of RBP to carry 10 μg of retinol/dl is 0.8 mg/dl. Recent studies undertaken in Africa showed that this minimal carrying capacity for RBP is generally adequate, except in certain serious cases of protein-energy malnutrition [12] and in children infected with measles [16; 28, 31]. In 55 children with measles from the north of Senegal, 9 had a level of RBP lower than this limit; 2 weeks later the average RBP concentration had returned to normal [16]. It is possible that in certain preschool children this low level of RBP precipitates xerophthalmia, especially if the liver stores of vitamin A are depleted. All the controls – apparently healthy – presented a level of RBP higher than this limit, even the malnourished children [16].

Several authors have noted a liaison between vitamin A and hemoglobin or iron levels [11, 17]. Vitamin A may play an important role in hematopoiesis; the effect of vitamin A may not be directly on hemoglobin, but on the availability of iron for synthesis of this heme protein [17].

Clinical and biochemical surveys carried out in these countries have shown a high incidence of nutritional anaemia which is probably due to multiple deficiencies (iron, protein, folates, etc. ...), and of protein-energy malnutrition in the preschool children.

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