



Seasonal dynamics of human retinol status in mobile pastoralists in Chad



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ABSTRACT

Vitamin A deficiency is a prevalent public health problem in Africa and South-East Asia, although national population based surveys are lacking in many countries. This study investigated seasonal variation of human retinol concentrations in Chadian mobile pastoralists to identify critical time periods for interventions addressing vitamin A deficiency. The repeated cross-sectional study design used convenience sampling during three seasons to include 327 Fulani, Gorane and Arab adult mobile pastoralists in nine camps in the Lake Chad area. Human blood and pooled cattle milk retinol concentrations were rapidly assessed by portable fluorometer (iCheck™). Linear regression models with random effects for correlation within camps were applied with human retinol concentration as outcome. Logistic regression models, with camp as random effect, were evaluated for the outcome human retinol deficiency. Human seasonal means were 2.14 $\mu\text{mol/L}$ (95% CI 1.82–2.46) in rainy, 0.99 $\mu\text{mol/L}$ (95% CI 0.91–1.07) in cold and 1.86 $\mu\text{mol/L}$ (95% CI 1.63–2.09) in dry season. Retinol concentration and deficiency varied according to season and ethnic group. Average values were highest in Gorane during rainy and in Fulani in the cold and dry seasons. Arabs had lowest average values in all seasons. Retinol deficiency (<0.70 $\mu\text{mol/L}$) was found in 15% of study participants in the dry, 25% in the rainy and 32% in the cold season. Retinol concentrations varied according to age, sex, milk consumption level and pooled cattle milk retinol concentration. Effect sizes varied and not all were statistically significantly different. Pooled cattle milk retinol concentrations varied seasonally and were positively associated to human retinol concentrations. This study establishes seasonal variation in human blood and pooled cattle milk retinol concentrations in Chad, demonstrating a linkage from animals to humans through milk. Rapid analysis using portable technology is feasible in remote populations under harsh climatic conditions.

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1. Introduction

Vitamin A is an essential nutrient for human beings, required in very small amounts for normal function of the visual system and development and growth of cells, including sustained immune function, epithelial cell integrity and reproduction. Because of its importance for growth and development, vitamin A status is espe-

cially relevant in pregnant women and children. Human dietary requirements for vitamin A are generally consumed as preformed retinol and pro-vitamin A carotenoids. Vitamin A deficiency (VAD) continues to be a central public health issue and underlying cause of disease in developing countries (World Health Organization, 2009). Low serum retinol concentration (<0.70 $\mu\text{mol/l}$) affects an estimated 190 million preschool-age children and 19.1 million pregnant women worldwide, corresponding to 33.3% of the preschool-age population and 15.3% of pregnant women in populations at risk for VAD, globally. Africa and South-East Asia are most affected by VAD (World Health Organization, 2009), although national population based surveys are sparse in many countries (Stevens et al., 2015). Even sub-clinical VAD likely increases mortality and severe morbidity from common diseases such as measles

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and diarrhea (Sommer, 1994; Tanumihardjo et al., 1994). Vitamin A supplementation is associated with large reductions in mortality and morbidity in young children in a wide range of settings (Awasthi et al., 2013; Mayo-Wilson et al., 2011).

In the Sahel, there are approximately 50 million mobile pastoralists, with estimates in Chad ranging up to 2 million (Rass, 2006; Thornton et al., 2002). Pastoralists migrate in a seasonal cycle managing livelihood risk through a mobile lifestyle highly adapted to utilize variable resources (Bille, 1997). Although the main diseases found among mobile pastoralists in Chad did not differ from those of Sahelian sedentary communities (Schelling et al., 2005), mobility and socio-cultural factors limit access of mobile populations to social services, in particular health services and education (Cohen, 2005; Münch, 2012). Barriers to access include lack of education and information, insufficient political empowerment and inadequate policies, mutual distrust, remoteness and constraints of nomadic daily life (Schelling et al., 2010; Zinsstag et al., 2006). In mobile populations, livestock milk is the primary source of vitamin A (Holter, 1988; Zinsstag et al., 2002). Although fruits, vegetables, fish, liver and *Spirulina* are good sources for vitamin A, these are rarely consumed by pastoralists. There are few studies on VAD in Sahelian pastoralist populations, and none which included men. Previous work in Chad found 20% prevalence of retinol deficiency among settled/semi-nomadic pastoralist women and 5% prevalence among nomadic women at the end of the rainy season, with similar results in children under the age of five (Bechir et al., 2012). An earlier study noted 60% prevalence of retinol deficiency in nomadic women sampled at the end of the rainy season and during the dry season (Zinsstag et al., 2002). This study further investigated the seasonal dynamics of retinol levels in adult mobile pastoralists in the Lake Chad area.

2. Materials and methods

2.1. Study area, design and population sample

The study area was located in the regions of Hadjer-Lamis, Lac and Bahr El Gazel, Chad, near the southern shore of Lake Chad. There is a rainy season from July to September. This study followed nine camps for three sampling intervals over eleven months: rainy (September 2012), cold (January 2013) and dry season (July 2013).

The mobile population in the study area consists mainly of three ethnic groups, Fulani, Arab and Gorane, which utilize different animal husbandry practices. Fulani stay close to the shore of Lake Chad and on islands within the lake, and also utilize subsistence agriculture growing millet and maize (Bechir et al., 2013). During the rainy season, they move eastward to access fresh pasture. Arab cattle breeders occupy similar areas but rarely cultivate crops (Krönke, 2001; Wiese, 2002). Gorane pastoralists utilize a larger transhumance area, moving primarily to the north, and do not farm crops (Weibel et al., 2008). Due to the nature of the study population, an accurate sampling frame was not available. The most recent census, done in 2009, registered about 387,800 nomads nationally, but evidence indicated many mobile people were not counted (Jean-Richard, 2013). A sample size of 300 was calculated, based on a prevalence of retinol deficiency of 20%, assuming a level of significance of 95% and a standard error of <1%. Ten camps, four Fulani, three Arab and three Gorane, were selected, considering logistic constraints, accessibility (projected location based on known transhumance patterns), availability and history of cooperation. Despite these considerations, one participant camp declined to participate before the first sampling interval, and one dropped out after the second sampling interval (reasons were insufficient incentive and change in transhumance route; both were Gorane). Eight camps participated during three sampling intervals and one camp

participated during two sampling intervals. In 2010–2012, median camp population in the study zone was 129 people (25–75% quartiles 65–210 people) (Jean-Richard et al., 2015), with a range of 3–7 households per camp (Jean-Richard et al., 2014).

2.2. Sample collection

In each camp, the study protocol was explained in the local language to the camp chief and to the heads of households, who were men, and the community willingness to participate was agreed. Seven men and seven women at least 15 years of age who presented voluntarily were selected from multiple households respecting the prevailing cultural customs. All participants were informed about the study procedure and verbal informed consent was obtained, according to the local norm. Each participant was interviewed to record demographic and dietary recall information (amount of milk consumed per day and number of times fruits or vegetables were consumed per week). Two ml of whole blood was collected by venipuncture, stored in EDTA tubes and protected from light and excessive heat until undergoing on-site analysis, which was performed within two hours. Records were kept anonymously using a number system to ensure patient confidentiality. Individual vitamin A outcomes were shared with participants immediately after testing, and those with severely deficient status (retinol <0.35 $\mu\text{mol RE/L}$), with the exception of women who stated that they were pregnant, were offered oral vitamin A supplementation without cost. Participants who received supplements were noted by participant number and excluded from the sample cohort at subsequent sampling intervals ($n = 28$). All deficient participants were advised to increase their milk consumption. All study participants were evaluated by the study nurse to ensure that they were not showing signs of clinical illness. All members of the camp community, regardless of participation in the study, were offered an on-site health consultation and examination by the study nurse. All camp members who were ill received basic treatments without cost, and two complicated cases were referred to a health center.

All cows in the study population were Zebu (*Bos indicus*) (Flury et al., 2009). Cow milk was pooled by household prior to consumption or marketing. Twenty ml of pooled morning milk from each household was collected into sterile tubes soon after milking and protected from light and excessive heat until analysis, which was performed within two hours.

2.3. Sample analysis

Portable iCheck™ devices (BioAnalyt GmbH, Teltow, Germany) were used to measure vitamin A (retinyl palmitate, retinyl acetate and retinol) and total carotenoids in blood and milk samples within two hours after collection. The iCheck™ FLUORO measures fluorescence to quantitatively determine retinol equivalent (RE) as $\mu\text{g/L}$ in biological fluids. The iCheck™ CAROTENE uses light emitting diode technology to photometrically measure total carotenoids as mg/L . These devices were previously validated against the gold-standard method of high-performance liquid chromatography (HPLC) using cattle blood (Raila et al., 2012). Manufacturer's instructions¹ for both devices were strictly followed, including calibration control. For human samples, a conversion factor was calculated (for women, 1.61; for men 1.79), based on published average hematocrit values (women: 38%, men: 44%) (Walker et al., 1990), and used to adjust the measured retinol concentrations

¹ iCheck™ FLUORO User Manual April 2013.EN.V15; iCheck™ MILA Instruction Guide Sept 2011

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