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Intervention to diminish dehydration and kidney damage among sugarcane workers ¹

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- 1 Supplementary material
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Handling and analysis of blood and urine samples

Workers were given urine collection flasks two days before and asked to bring their first void on the day of our field visit. Participants were instructed how to supply clean mid-stream urine samples. Workers brought the first void of urine the day of our visit but, if the sample was older than 2 hours upon arrival to the field, workers were asked to provide a new sample. Urine was transferred to 3 separate 8 mL tubes using a vacuum system. Two tubes were stored directly in an icebox at 4-10°C and transported to the Centro de Hemodiálisis, San Salvador where they were stored in a freezer logging -20 to -25°C within 4 hours of collection. The third tube was assessed by dipstick (Bayer 10-SG MultiStix Urine Dipsticks) and then the sample was centrifuged directly on-site for immediate microscopic urinalysis on sediment. Dipstick results were read optically using a Siemens CLINITEK Status®+ Analyzer. Urine osmolality was analyzed using an Atago PAL-mOsm, a digital hand-held compact refractometer (ATAGO Co. LTD).

Venous blood samples were drawn from participants using a BD vacutainer system, one 8 mL SST serum tube with gel separator and two 4 mL EDTA tubes. The serum tube was centrifuged after 30-50 minutes in ambient temperature and thereafter stored in the icebox before being transported to the laboratory where serum was transferred to cryo-safe tubes and stored in the freezer within 4 hours of collection. EDTA tubes were stored in the icebox and one tube was used for same-day hemogram at Laboratorio CECIAM Escalon, San Salvador while the second tube was stored at the Centro de Hemodiálisis, San Salvador in the freezer within 4 hours of collection.

The frozen samples of urine, serum and blood were later shipped on dry ice to Sweden together with a temperature logger confirming a temperature lower than -20°C, stored temporarily at -80°C and thereafter thawed, aliquoted and analyzed at the Department of Clinical Chemistry, Skåne University Hospital in Lund, Sweden. Biomarkers were analyzed on a Cobas 701-instrument (Roche Diagnostics, Basel, Switzerland). The serum level of creatinine was determined by an enzymatic colorimetric assay and by use of an isotope dilution mass spectrometry (IDMS)-traceable calibrator.

Table A: Baseline Socio-demographic, health and work characteristics for participants seen at both baseline and end of harvest and for the subset with largest cross-harvest eGFR changes

						FR	Dath are			
		and		tland		pers ^a	Both groups n=80			
	n=	40	n=	40	n=	20	n=	80		
Work history in:										
Cotton	1	3	14	35	4	20	15	19		
Other agriculture	36	90	27	68	15	75	63	79		
Construction	13	33	15	38	8	40	28	35		
Frequently worked with										
agrochemicals (past 12 months)										
Fertilizers	8	20	7	18	5	26	15	19		
Pesticides	16	40	9	23	7	35	25	31		
Smoking										
Current										
Male	11	28	6	21	4	27	17	25		
Female ^b	-	-	=	-	-	-	-	-		
Ex-Smoker										
Male	6	15	8	29	4	27	14	21		
Female ^b	-	=.	=.	=	-	=	-	-		
Alcohol consumption										
Males										
None	24	62	15	54	9	60	39	58		
Sometimes	14	36	13	46	6	40	27	40		
Often/every day	1	3	0	0	0	0	1	1		
Females ^b										
None	-	-	10	83	4	80	11	85		
Sometimes	-	-	2	17	1	20	2	15		
Often/every day	-	-	0	0	0	0	0	0		
Marihuana use (last 30 days)	2	(20	0	0	0	0	2	15		
Acetaminophen	15	38	15	38	8	40	30	38		
Metamizole	0	-	4	10	3	15	4	5		
Aspirin	4	10	6	15	4	20	10	13		
Antibiotics	1	3	2	5	0	_	3	4		

^a Quartile with largest negative eGFR changes during the harvest (Inland 8; Coastland 13)

^b Only one female participant in the Inland group – data not presented for privacy

Supplementary Table B: Medians and interquartile ranges of biomarkers of hydration and kidney function among Inland (subtable A-1) and Coastland (sub-table A-2) cane cutters, at baseline and at three points during the harvest 2013-2014, and mean cross-shift changes of these markers at three points during the harvest.

									B-1:	NLAN	ID												
		_	BASELII			/Dr		ANUARY ntervention)				/ N	FEBF ⁄lid-inte	RUARY	on)		APRIL (End of harvest)						
		(F)	re-harvest) AM				1		M	Δ		<u> </u>	M	PM		Δ		AM		PM		Δ	
	Units	N	Q ₂	IQR	N ^a	Q ₂	IQ R	Q ₂	IQR	am- pm	• • • • • • • • • • • • • • • • • • • •	Q ₂	IQR	Q ₂	IQR	am- pm	N	Q ₂	IQR	Q ₂	IQR	am- pm	
Biomarkers							l						1		I								
Calculated eGFR	ml/min/ 1.73kg/m²	40	119	17	35	115	22	100	31	- 13*	37	115	22	112	32	-6	40	116	19	108	37	-11*	
Serum uric acid	μmol/L	40	297	63	35	283	69	338	58	38*	37	286	58	309	76	23	40	307	65	326	56	24*	
Serum CPK	μkat/L	40	2.9	2.4	35	3.1	1. 5	3.6	1.7	0.5	37	3	1.5	3.4	1.9	0.5	40	2.7	1	3.2	1.3	0.6	
Serum creatinine	μmol/L	40	71	8	35	73	10	86	12	13	37	75	8	80	15	6	40	73	10	82	17	13	
Serum chloride	mmol/L	40	101	3	35	100	3	101	3	1*	37	99	3	100	3	1	40	99	2	98	5	0*	
Serum sodium	mmol/L	40	141	2	35	140	2	142	2	2** *	37	140	2	142	3	2**	40	140	2	141	3	1***	
Serum albumin	g/L	40	43	5	35	42	4	43	4	0	37	43	4	43	4	0	40	41	4	43	5	1*	
Urine osmolality	mOsm	_b	-	-	35	470	29 0	845	200	300 ***	37	650	410	820	310	204	40	500	340	810	245	259* **	
Hemoglobin	g/L	40	149	10	35	144	15	137	11	-6*	37	140	11	135	12	-5*	40	146	13	140	13	-5*	
Hematocrit	%	40	44	3	35	46	5	44	3	-2*	37	43	3	42	4	1	40	46	4	44	5	-1	
eGFR by health																							
Hyper-tension ^c	Yes	6	112	21	5	115	11	103	31	-12	6	108	19	97	31	-9	6	108	23	86	29	-11	

	No	33	121	13	29	118	22	98	35	-18	30	119	16	114	30	-5	33	116	19	110	29	-9
Self-rated	≥good	15	118	16	13	119	18	103	33	-15	14	120	18	112	35	-5	15	124	22	108	41	-8
health	≤regular	25	119	17	22	115	16	98	30	-12	23	114	27	107	36	-6	25	114	19	97	33	-10

^a Pre-shift n = 35. One worker in January participated only in pre-shift testing, therefore, n for post-shift values and am-pm values is 34

 Q_2 = median

IQR = inter-quartile range

 Δ = mean difference by paired t-test (*= p<0.05; **= p<0.01; ***= p<0.001)

eGFR = estimated glomerular filtration rate

CPK = creatine phosphokinase

^b Refractometer instrument arrived after baseline, therefore no urine osmolality measures at baseline

^c One subject's measurement of blood pressure is missing for unknown reason.

								B-2: C	OASTI	AND													
			BASELIN		JANUARY (Pre-intervention)							(1)		RUAI	RY ntion) ^d		APRIL (End of harvest)						
		(Pre-harvest)				A			PM	Δ			AM	1	PM	Δ		AM		PM		Δ	
	Units	N	Q ₂	IQR	N	Q ₂	IQR	Q ₂	IQR	am- pm	N	Q	IQR	Q IQF	IQR	am- pm	N	Q ₂	IQR	Q ₂	IQR	am- pm	
Biomarkers									•														
Calculated eGFR	ml/min/ 1.73kg/m²	40	108	30	11	108	30	93	34	-12	-	-	-	-	-	-	40	100	37	86	34	-9	
Serum uric acid	μmol/L	40	324	124	11	288	77	347	97	34	-						40	319	122	359	140	29	
Serum CPK	μkat/L	40	2.2	0.9	11	2.9	1.8	3.8	2.5	0.9	-	-	-	-	-	-	40	2.6	1.3	3.3	1.8	0.7	
Serum creatinine	μmol/L	40	76	31	11	78	12	85	38	12	-	-	-	-	-	-	40	81	30	91	37	11	
Serum chloride	mmol/L	40	101	3	11	99	4	100	3	1	-	-	-	-	-	-	40	101	4	100	4	0	
Serum sodium	mmol/L	40	140	2	11	139	2	141	3	1	-	-	-	-	-	-	40	140	2	140	4	1	
Serum albumin	g/L	40	40	5	11	40	5	41	6	1	-	-	-	-	-	-	40	40	3	41	5	0.5	
Urine osmolality	mOsm	_e	-	-	11	570	480	720	400	78	-	-	-	-	-	-	40	420	330	690	265	203	
Hemoglobin	g/L	40	139	16	11	135	21	130	18	-6	-	-	-	-	-	-	40	132	18	127	16	-4	
Hematocrit	%	40	42	4	11	45	8	43	6	-2	-	-	-	-	-	-	40	42	6	41	5	-1	
eGFR by health											-	-	-	-	-	-							
Hyper-tension	Yes	6	103	51	0	-	-				-	-	-	-	-	-	6	91	41	75	35	-11	
riyper-terision	No	32	108	31	11	108	30	93	34	-12	-	-	-	-	-	-	32	101	39	89	36	-9	
Self-rated health	≥good	25	105	27	5	106	18	79	34	-14	-	-	-	-	-	-	25	101	27	82	33	-10	
Jen racea nearth	≤regular	15	109	45	6	110	35	93	31	-12	-	-	-	-	-	-	15	98	46	92	40	-8	

^d Coastland group did not participate in February sampling (see Methods)

Q₂ = median

IQR = inter-quartile range

= mean difference by paired t-test (*= p<0.05; **= p<0.01; ***= p<0.001)

eGFR = estimated glomerular filtration rate

CPK = creatine phosphokinase

^e Refractometer instrument arrived after baseline, therefore no urine osmolality measures at baseline

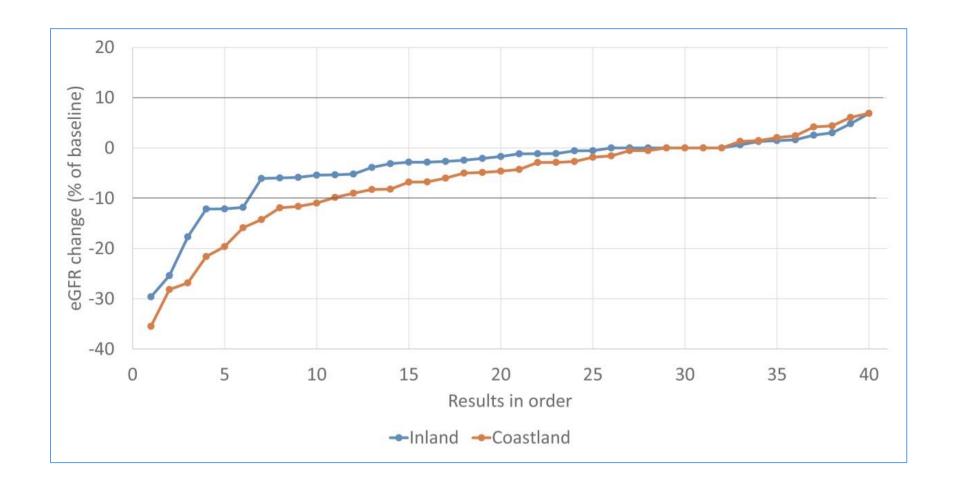


Figure A: eGFR changes during the harvest season for the Inland intervention group and the Coastland group without intervention (ordered highest to lowest). The threshold value of 10% is indicated in the figure. An eGFR decrease of ≥10 % was seen in 28% of the individuals in the Coastland group but only 15% of the individuals in the Inland intervention group.