



ISSN (E): 2277-7695  
ISSN (P): 2349-8242  
NAAS Rating: 5.23  
TPI 2023; 12(3): 2709-2710  
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Received: 16-12-2022

Accepted: 20-01-2023

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## Feasibility of urine collection from buffaloes for biomarker assay

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### Abstract

Urine is a non-invasive bio-fluid used for biomarker detection. The present study aims to assess the feasibility of urine collection in buffaloes. Total six healthy buffaloes were observed for urination for seven days. If animal did not urinate within 20 minutes, buffaloes were stimulated for urination by pouring water on rump, tying or offering water. Out of 84 attempts, only 60 times urine was collected, suggesting ~72% chances of urine collection. Time-wise study showed that 4.76% urination was within 0-5 min, 3.57% urination within 5-10 min, 8.33% urination within 10-15 min and 21.42% of urination within 15-20 min. 8.33% buffaloes urinated by pouring water on rump. If animal did not urinate after 20 min, 3.57% buffaloes showed urination when they were offered water and 21.42% urination was seen when animals were tied in the horns. In conclusion, there is nearly 72% feasibility of urine collection for biomarker analysis.

**Keywords:** Urine, buffaloes, collection, biomarkers, feasibility

### 1. Introduction

Urine is a non-invasive fluid which is excreted in large volumes which serves as an ideal source for biomarker detection (Pedroza-diaz and Rothlisberger, 2015) [2]. It is excreted in large volume. It is having 95% of water portion containing electrolytes and metabolites proteins, nucleic acids and organic components (Bock *et al.*, 2010) [1]. Urine can be used to identify biomarker like proteins and microRNA. For instance, luteinizing hormone (LH) in urine acted as a biomarker to predict the ovulation in buffaloes (Selvam *et al.*, 2017) [3]. Some of the urinary microRNAs were found as biomarkers for hepatotoxicity, cancer studies and kidney injury (Wang *et al.*, 2012) [4]. For analyzing the biomarkers collection of the sample is important. Urine collection can be controlled in human patients. But in veterinary patients' collection can't be controlled. Till now there is no report of feasibility of urine collection from buffaloes. To check feasibility of urine collection in buffaloes, present study was conducted.

### 2. Materials and Methods

Total six healthy buffaloes were taken for the study. The institutional ethic committee of ICAR-National Dairy Research Institute has approved these animals for sample collection under the IAEC approval No. 42-IAEC-18-2. These animals were managed as per the standard managemental conditions at Livestock Research Centre (LRC), ICAR-National Dairy Research Institute, Karnal. These animals were observed for urination twice daily for seven days. Urine was collected in the morning time between 8.15 a.m to 9.15 a.m. and in the evening from 3.30 p.m to 4.30 p.m. 20 minutes were decided as standard time for observation. If the animal urinated within 20 minutes, that time was noted down. If the animals were not urinated within 20 minutes, then animals were stimulated for urination by pouring water on the rump region or by tying the animal or by offering the water to the animal. Thus total 84 times tried to collect urine.

### 3. Results and Discussion

With all efforts as described above, out of 84 attempts, only 60 times urine could be collected. Therefore, in the present study, results showed that 71.42% chances of collection of urine. The details of time taken and different effort made for urine collection has been summarized in Table 1. Urination data obtained without any external stimulation, time wise study showed that 4.76% urination was within a period of 0-5min, 3.57% urination within a period 5-10 min., 8.33% urination within a period of 10-15 min and 21.42% of urination within 15-20 min. However, 8.33% buffaloes were urinated by pouring water on the rump region.

Maximum possibility of urination was seen when the animals were tied by putting an extra rope in the horns (21.42%) and 3.57% buffaloes showed urination when they were offered water.

Here we have found that 8.33% buffaloes were urinated by pouring water on the rump region. By pouring cool water on the rump region, urinary bladder might be undergoing contraction in order to emptying the urine outside. 3.57% buffaloes urinated when they were offered water for drinking. Sound produced by the time of water supply may cause stimulant to the buffaloes, it may urinate. Splash of water, movement of the workers or movement of pipe or water pump sound may cause stimulation to bladder and animal may urinate. Sometimes if the animal is lying down on the floor, but for drinking water it may get up and may urinate. However, this probability may increase further at farmer level. The close association with buffaloes further helps in understanding the behaviors of the animal and predicting the expected timings of urination. But we found the maximum possibility (21.42%) of urination when the animal is made to

tie applying an extra rope to horns. Application of rope may frighten animal and cause activation of sympathetic system thereby it may lead to urination. Sometimes if we apply an extra rope to one animal, the neighboring animal may get up and urinate. If we study in a large herd approaching the one animal may cause some sort of fear in the neighboring animal and it may urinate. In a large herd by seeing the one animal urinating the other animal may urinate. But to confirm this study must be done in large population. In the field conditions, the farmers are closely associated with the animal and understand the behavior of the animal. They might better predict the possibility of animal urination rather than in an experimental set up. This may further enhance possibility of urination. We understand this was first study where the feasibility of urination to collect urine has been studied. Since there is no information available and therefore it was not possible to corroborate present data with literature. This analysis can help in calculating efficiency of any methods where time dependent urine sample is required.

**Table 1:** Time taken and efforts done for the urine collection in buffaloes

Animal No.	10/11/17		13/11/17		14/11/17		15/11/17		16/11/17		17/11/17		18/11/17	
	M	E	M	E	M	E	M	E	M	E	M	E	M	E
7201	20min	5min	13min	*	20min	**	*	**	17min	**	15min	N	5min	**
7203	15min	10min	5min	N	**	15min	***	**	***	**	5min	**	N	**
7204	18min	*	10min	**	20min	**	N	N	20min	N	20min	**	N	N
7206	20min	N	***	N	13min	13min	20min	N	N	N	20min	N	*	N
7207	N	*	N	*	10min	*	20min	N	20min	**	N	N	N	18min
7208	15min	N	20min	**	16min	16min	N	**	20min	**	N	**	20min	**

\* Water pouring on the rump, \*\*Tying the animal, \*\*\*Water is offered min, Minutes; M, Morning; E, Evening & N, Urine could not collect applying all effort as described

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