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THE BEST OF TEMPERATURE STORAGE ON MEASURED URINE ALCOHOL CONCENTRATION: A SYSTEMATIC REVIEW

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Abstract

Background*: Urinalysis is an examination that aims to detect and identify certain materials in urine such as alcohol. If an alcohol-containing urine sample cannot be analyzed immediately or requires shipping, it should be stored at an appropriate temperature. Pre-analytic is a stage that needs serious attention where at this stage the risk of errors often occurs. Error in storage samples can affect the results of alcohol concentration, so a laboratory technician needs to understand urine sample storage to measure alcohol concentration.* ***Aims****: Knowing the best storage of urine temperature in measuring alcohol concentration.* ***Methods****: This study used a systematic literature review of urine samples to measure alcohol concentration from PubMed and ScienceDirect obtained 24 data that according to the inclusion criteria*. ***The results****: The storage temperature on measured urine alcohol concentration was carried out from 1977 – 2023. It can be seen that in some studies regarding storage temperatures in urine using at least 5 temperatures: -20ºC, 4ºC, 6ºC, 14ºC, ±20ºC. The temperature storage at 4ºC can prevent the production of ethanol due to microorganism activity, it will maintain the stability of alcohol concentration in urine*. ***Conclusions****: A temperature of 4ºC is the best temperature for measuring alcohol concentration in urine samples.*

Keywords*: alcohol, pre-analytic, urine storage, urinalysis, temperature*

1. Introduction

Alcoholic beverages in the form of ethyl alcohol are one of the preferred drinks among the public, especially teenagers. Alcohol is one of the drinks commonly consumed by a person as a result of addiction and dependence[1]. According to Badan Pusat Statistik Indonesia, the number of alcohol drinkers aged ≥ 15 years in urban and rural areas in 2022 was 0.33%[2]. Alcohol concentration measurement can use samples from various body fluids including blood, breath, urine, and saliva. After alcohol is consumed, as much as 2-8% will be excreted through urine, saliva, tears, and breathing[3].

Urine alcohol measurement is carried out by social service agencies, the criminal justice system, or in alcohol rehabilitation programs[4]. Urine in the quantitative analysis of alcohol is particularly valuable when there is uncertainty about the validity of blood samples[5].

The use of appropriate temperatures is carried out if there is a delay in the

measurement of urine alcohol concentration[6]. Temperature storage is necessary because in some cases it has been shown that the increase in alcohol concentration may not be due to the consumption of alcoholic beverages, but rather due to the activity of microorganisms in the urine capable of producing ethanol from the glucose contained in the sample, especially people with Diabetes Mellitus[7]. The criteria for urine samples used were urine of alcohol drinkers and not suffering from Diabetes Mellitus. Temperature storage for alcohol concentration measurement is required before the sample is transported to the laboratory.

Considering the importance of storage temperature in the pre-analysis stage of determining urine alcohol concentration in case of delay in laboratory testing, it is necessary to conduct research with a Systematic Literature Review (SLR) approach. Based on the description above, the purpose of this article is to review the best storage temperature for measured the alcohol concentration.

2. Research Methods

Data search

For this data search, we combined through two online databases: PubMed and ScienceDirect. By combining Free Word with Medical Subject Heading (MESH), the search was made more sensitive. In Boolean expressions, operators like "Or," "And," and "Not" combine many terms. "Urine Alcohol Storage" and "Urine Storage Temperature" were the search terms. Journals produced after the data search was conducted in September 2024 were not included in the category. After searching on PubMed and ScienceDirect obtained 93 data searches, but at the end of the data selection, 24 data were obtained according to the inclusion criteria.

Data selection

I) The journals must be of an RCT type and written in English. ii) They must adhere to a complete writing structure that includes an introduction, methods, results, and discussion. iii) Journals must be accessed using keywords.

Data extraction

The main information extracted in the form of the temperature variation on measured urine alcohol concentration. In addition, information related to factors affecting the determination of alcohol concentration.

Data synthesis

Data synthesis was carried out qualitatively (meta-synthesis) concerning the best storage temperature on measured urine alcohol concentration. This article features milestones to describe studies into the storage of alcohol-containing urine samples using the temperature.

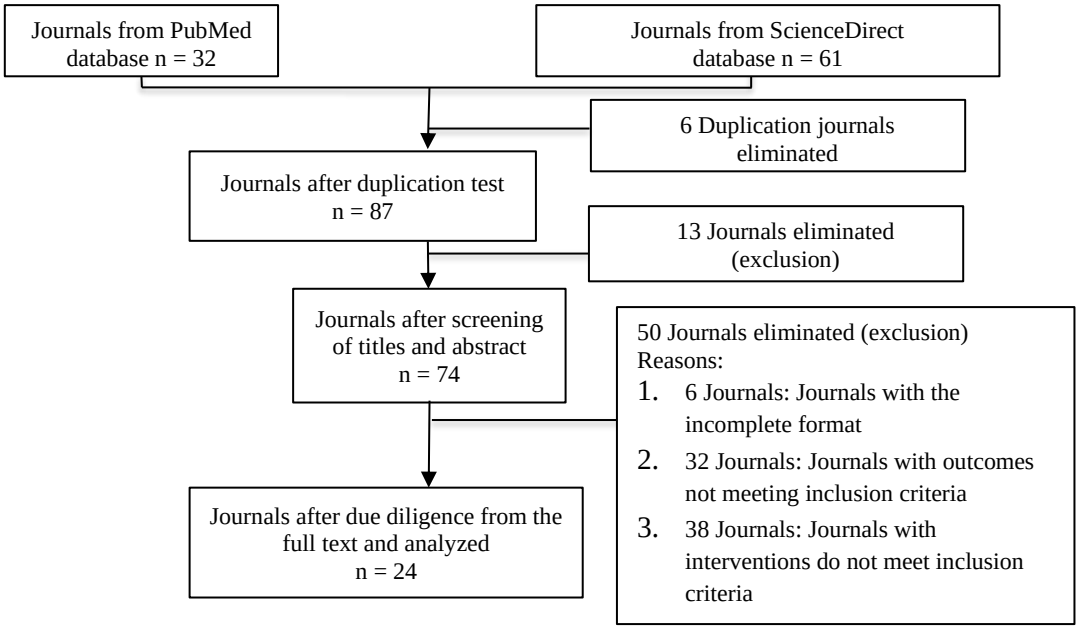

Figure 1. PRISMA flow diagram of systematic review

3. Results and Discussions

Results

A data search in this article uses 2 electronic databases, PubMed and ScienceDirect, because the combination of electronic databases has more optimal performance than single-use [8]. The total data obtained were 93 data searches on 01/10/2024. The data search results from this article are presented in Table 1.

The results of the data selection are summarized in the prism diagram of the study in Figure 1. The data went through 3 stages, namely: duplication test, selection of title and abstract, full text [9]. At the end of the data selection, 24 data were obtained according to the inclusion criteria and presented in Table 2.

Data extraction shows that storage temperature on measured urine alcohol concentration was carried out from 1977 – 2023. It can be seen that in some studies regarding storage temperatures in urine using at least 5 temperatures: -20^{o}C, 4^{o}C, 6^{o}C, 14^{o}C, ±20^{o}C. A summary of several temperature used for urine storage is presented in Table 3. This article presents a data synthesis in many sections, the first of which is: the evolution of urine samples containing alcohol in storage studies (Figure 2); the best storage temperature on measured urine alcohol concentration without preservative (Table 4); the best storage temperature on measured urine alcohol concentration with preservative (Table 5).

Table 1. Data Search Results

Databases	Search Term	Total
PubMed	"Urine Alcohol Storage"[Mesh] OR "Alcohol Urine"[tw] OR "Urine Storage"[tw] OR "Alcohol storage"[tw] OR "Alcohol concentration"[tw] AND ("Urine Storage Temperature "[Mesh]) OR "Urine Temperature"[Mesh]) OR "Alcohol Temperature"[Mesh] OR "Alcohol concentration"[tw]	32
ScienceDirect	("Urine Alcohol Storage" OR "Alcohol Urine" OR "Urine Storage" OR "Alcohol storage" OR "Alcohol concentration") AND ("Urine Storage Temperature" OR "Urine Temperature OR "Alcohol Temperature OR "Alcohol concentration")	61
	Total	**93**

Table 2. Data Extraction Results

Year of Journal Publication	References
1977	[10]
1985	[11]
1993	[12]
1994	[13]
1999	[14]
2001	[15], [16]
2002	[17], [18]
2003	[19]
2004	[20]
2007	[21]
2008	[22]
2009	[23]
2012	[24,25]
2013	[26]
2015	[27]
2016	[28]
2017	[29]
2019	[30,31]
2020	[32]
2023	[33]

Table 3. Summary of several temperatures used for urine storage

Temperature Urine Storage	Result	
	Advantages	**Disadvantages**
-20ºC	1. The pH values of urine specimens are relatively stable with a storage time of 2 weeks 2. Concentration drugs in urine were stable for 12 months, except for cocaine concentration 3. Recommended for quantitative analysis of 1P-LSD for up to five days	1. Not recommended for analysis of hCG 2. Decreased the urine specific gravity
4ºC	1. Catecholamines in urine are stable for 1 month in unpreserved, 4 months in urine preserved, and 1 year in acidified urine 2. Urine bacterial counts were well-preserved 3. Recommended for analysis hCG 4. Calcium oxalate, magnesium, phosphate, sodium, urea, or pH urine were well-preserved²⁷ 5. Urine alcohol concentrations were also stable for up to 1-4 weeks without preservatives, and 1 year with preservative	1. Decrease in clenbuterol concentration 2. Concentration of catecholamine in urine with format buffer preservative are declined 3. High risk of containing viable viruses 4. Creatine concentration increase for 15 days
6ºC	Did not have a significant effect on pH or specific gravity with a storage time of 6-24h	High risk of containing viable viruses
14ºC	Reduce bacteriophage and bacteria	High risk of containing viable viruses
±20ºC	1. The concentration of drugs (MDA, MDMA, and MDEA) is stable for 21w 2. Recommended for urine of 6 month 3. Urine-specific gravity was only modestly correlated	Increase in endogenous GBH levels in urine Reduce the concentration of a large list of drugs
References		
10,11,12,13,14,15,16,17,18,19,20,21,22,23,24,25,26,27,28,29, 30,31,32,33		

Table 4. Summary of production ethanol urine in 4ºC without preservative

Day	Ethanol Concentration (g/L)
0	< 0.5
2	< 0.5
4	< 0.5
6	< 0.5
8	< 0.5
10	< 0.5
12	< 0.5
14	< 0.5
34	< 0.5
References	
14	

Table 5. Additional information of production ethanol urine in 4ºC with preservative

1st analysis (mg/100ml)	40 days (mg/100ml)	1 year (mg/ml)	Difference in 40 days	Difference in 1 years
216	212	213	-4	-3
292	287	285	-5	-7
312	309	303	-3	-9
310	303	303	-7	-7
218	214	215	-4	-3
400	399	392	-1	-8
309	309	305	0	-4
289	290	287	+1	-2
257	253	253	-4	-4
References				
[10]				

Discussions

Urinalysis is an examination that aims to detect and identify certain materials in urine such as alcohol. If the sample cannot be analyzed immediately, it should be stored at an appropriate temperature because it will affect the result of alcohol concentration[34]. The appropriate storage of urine samples in the examination served to prevent changes, like color, pH, glucose, and other compounds that are very sensitive and can affect microscopic reading[35].

Based on Table 3. we can see the development of 46 years (1977-2023) of research about the advantages and disadvantages of temperature variation on measured urine alcohol concentration. Storage time and temperature led to a significant increase of Calcium Oxalate (CaOx) crystals in urine samples, due to changes in specific gravity and pH of urine storage [19]. Calcium oxalate, sodium, urea in urine samples were stable at 4ºC [21].

Alcohol is rapidly absorbed from the gastrointestinal tract within 30-60 minutes after ingestion and

will be distributed throughout the body and tissues, easily crossing the blood barrier and placenta. 90-98% of alcohol consumed is metabolized by the liver enzyme system into carbon dioxide and water. 2-8% is excreted through urine, saliva and respiration[3].

Based on table 4, shows that urine containing ethanol-alcohol stored at a temperature of 4ºC will remain stable for up to 34 days even without preservatives. This is because with storage at temperature 4ºC, urine bacterial counts were well preserved[17,22,14]. So, with storage at a temperature of 4ºC, urine alcohol concentrations were also stable without preservatives and can be stable until 1 year with preservatives (Table 4)[10,33]. Cause several microorganisms capable of producing ethanol-alcohol from glucose contained in the urine sample such as *Candida sp., Escherichia coli, Enterococcus, and Klebsiella sp.*[7,14].

4. Conclusions

Many variations of urine storage temperatures have been used since 1977-2023. The widely used urine storage temperatures are -20, 4, 6, 14, and ±20ºC. 4ºC is the best temperature for the measurement of alcohol concentration in urine samples. The storage at 4ºC can inhibit some microorganisms capable of producing ethanol-alcohol from glucose contained in the sample.

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