SELECTION AND STANDARDIZATION OF A METHOD FOR QUANTITATIVE FRUCTOSE DETERMINATION IN HUMAN EJACULATES

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Key-words: fructose — human sperm — quantitative determination — methods

D-fructose is a product of the seminal vesicles. It is proved to be a constant component of the ejaculate of some mammals and man (5). It is the basic sugar in the seminal plasma and plays a main role in spermatozoon metabolism and vitality. Fructose secretion and accumulation in the seminal vesicles is under the hormonal control of androgenic factors produced by testicular Leydig cells. Fructose is a basic energy source for spermatozoon motility and plays an important role in the fertilizing capacity. That is why it is necessary to choose the most suitable method for its quantitative determination with the view of diagnosis and prognosis of male infertility.

Material and methods

In the present study three methods were applied and adapted. All of them were based on the same principle namely Selivanov's reaction (1) — i. e. after heating with mineral acid and resorcinol fructose underwent acetylation and dehydratation resulting in W-hydroximethylphurphurol. The latter stained red in the presence of resorcinol. The following methods were compared in our study:

1. The method described in "Clinical Laboratory Methods (3)."
2. Bistop's et al. method (4).
3. Schirren's method (6).

Two basic fructose standard solutions with concentrations of 27,8 and 55,5 mmol/l, respectively, were used as samples. There were also controls with definite fructose concentration of 11,10 mmol/l. The samples were read by using of calculating spectrophotometer LKB at 501 nm wave length.

Results and discussion

In the case of application of the first method (3) a calibration curve was constructed starting from a basic fructose standard of 27,8 mmol/l (fig. 1). Five dilutions were done. Simultaneously, controls' samples containing 11,10 mmol/l fructose processed as patient's samples after the same method were assessed. The results were read on the calibration curve and showed a maximal deviation of $\pm$0,278 mmol/l.

The second method (4) was applied by using the same basic fructose standard of 27,8 mmol/l for elaboration of the calibration curve (fig. 2). Five dilutions were done. Control samples processed after this method contained 11,10 mmol/l fructose. The results were read on the calibration curve and showed a maximal deviation of $\pm$0,278 mmol/l.

Fructose solution of 555 mmol/l was used to elaborate the calibration curve when the third method (6) was concerned. 15 dilutions were done. An enlarged standard curve was constructed (fig. 3). Control samples were read on the calibration curve and showed a maximal deviation of ± 0,278 mmol/l.

These three methods for quantitative determination of human ejaculate fructose showed reliability and preciseness. However, there are certain methodical

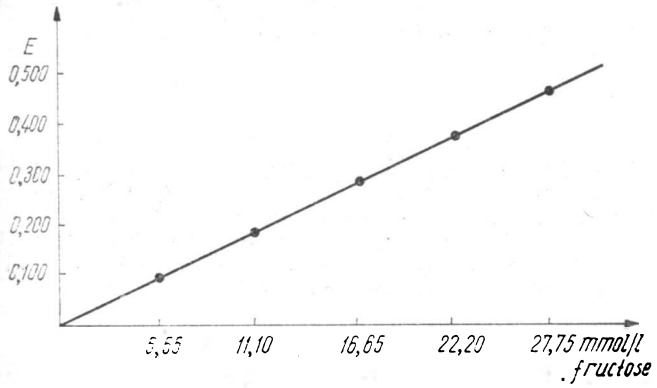

Fig. 1

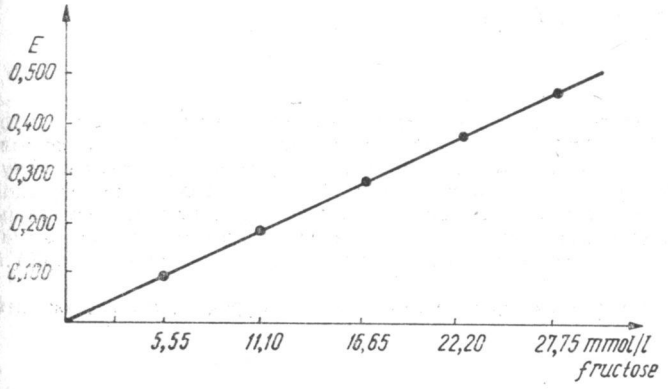

Fig. 2

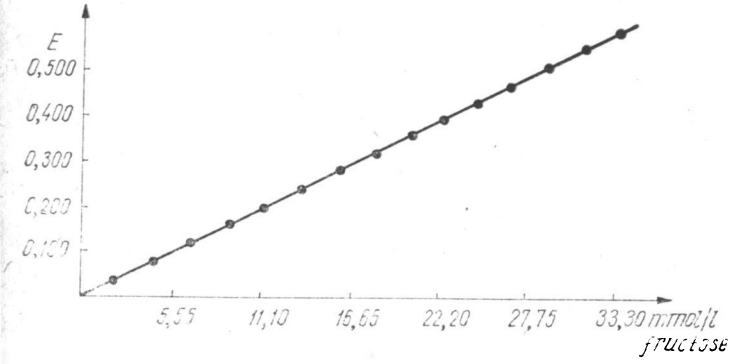

Fig. 3

differences related to the following aspects: technology of material deproteination; degrees and duration of temperring; chemical components of control (empty) samples of reagents. Schirren's method was proved to be the most suitable one from the technological point of view. That is why a statistical processing of the control data was done when reproducibility from day to day was concerned. The results were as followed: n=20; x=11,10 mmol/l; S=8,1; VC=4,05 %.

This allowed us to choose this method for assessment of human ejaculate concerning fructose quantity and to apply it in the andrological diagnostics.

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ВЫБОР И СТАНДАРТИЗАЦИЯ МЕТОДА КОЛИЧЕСТВЕННОГО ОПРЕДЕЛЕНИЯ ФРУКТОЗЫ В ЭЯКУЛЯТЕ ЧЕЛОВЕКА

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РЕЗЮМЕ

Д-фруктоза является постоянной составной частью семенной плазмы некоторых млекопитающих и человека. Ей принадлежит существенная роль в метаболизме сперматозоидов; она оказывает влияние на их подвижность. Количественное определение фруктозы в эякуляте человека имеет важное диагностическое и прогностическое значение. В работе обсуждаются методы, опирающиеся на один и тот же принцип согласно указанным литературным источникам. Существуют некоторые различия в технологии депротеинизации материала, в степенях и длительности темперирования и в химическом составе контрольных проб с реактивами.

Авторами рекомендуется метод Ширена для количественного определения содержания фруктозы в эякуляте человека.