BLOOD GLUCOSE ESTIMATION : AN EXPERIENCE WITH GLUCOSE REAGENTS OF DIFFERENT COMPANIES.

Introduction: As is expressed very often, reports in blood glucose levels in the same sample are different in different laboratories. This is always explained to be because of different persons doing the tests with different instruments, with different companies reagents. It was thought to rule out these variations by doing the estimation 1. In the same laboratory, 2. By using one and the same instrument, 3 by one person and 4. Done at the same time. But, by using different reagents. The results are presented.

MATERIALS AND METHODS :

1. About 3 ml of clear unhemolysed plasma was pooled from different patients on one day. It was mixed properly and was used as the Test Plasma.

2. The instruments used for this test were Autodilutor of Transasia and Erba chem 5 Semiautoanalyser of Transasia, which are used in my lab. Both these instruments were serviced just before doing this test and the filters were cleaned.

3. The Semiautolnalyser was tested as follows : Thirty ml of Glucose solution of Accurex (GOD-POD) taken in a large test tube to which 300 muL of test plasma was mixed properly. The mixture was incubated for 15 mins at 37^0 C. Twenty serial readings were taken of the final colour, which did not vary at all. This showed that the counting instrument is working properly.

4. To test the autodilutor, the same plasma was submitted for 20 serial tests, as if doing 20 different tests of different patients. The results were charted, and calculated for coefficient of variation. The CV was 3.5 %, well within accepted range (i.e. below 4.5 %). This tested both the accuracy of the autodilutor and the expertise of the technician who carried out this test.

NO	GL %	DIFF FROM MEAN	SQ OF DIFF	
1	188.8	2.8	7.84	
2	194.5	2.9	8.41	
3	193.4	1.8	3.24	
4	192.5	0.9	0.81	

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5	193.7	2.1	4.41
6	189.9	1.7	2.89
7	190.0	1.6	2.56
8	191.2	0.4	0.16
9	189.6	2.0	4.00
10	193.0	1.4	1.96
11	193.1	1.5	2.25
12	190.6	1.0	1.0
13	191.2	0.4	0.16
14	189.6	2.0	4.0
15	194	2.4	5.76
16	191.0	0.6	0.36
17	189.6	2.0	4.0
18	188.6	3.0	9.0
19	194.0	2.4	5.76
20	192.8	1.2	1.44

TOTAL: 3831.1 70.01

MEAN 191.6

SD =   1.919, CV = 0.1 %

Steps No 3 and 4 minimized the instrumental and personal errors in this test.

5. Seven different companies. glucose kits (GOD-P0D) were tested during this test. The companies were Accurex, Biolab, Boehringer, Cadilla, Erba, Ranbaxi and Span. The reagents were prepared as per the company's instructions on one and the same day . The glucose estimation on the Test Plasma was carried out by using 1000muL of the reagent and 10 muL of the Test Plasma.

    i.   Seven racks containing 24 test tubes each were prepared bearing the name of the company to be tested.
    ii.   The same Test Plasma was used for the test.
    iii.   The same company's blank and standard were used to calibrate the instrument. Twenty tests were performed on the test plasma for each reagent. The last test tubes contained the same company's standard for confirmation.
    iv.   The results were tabulated . The mean was graphed with the other companies. means.
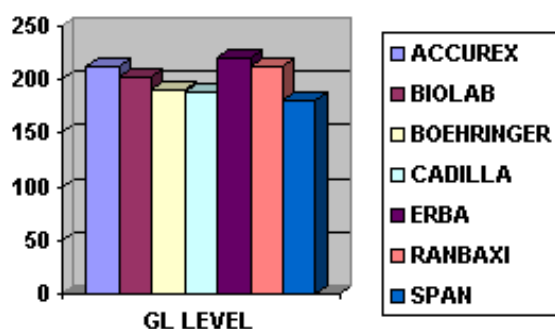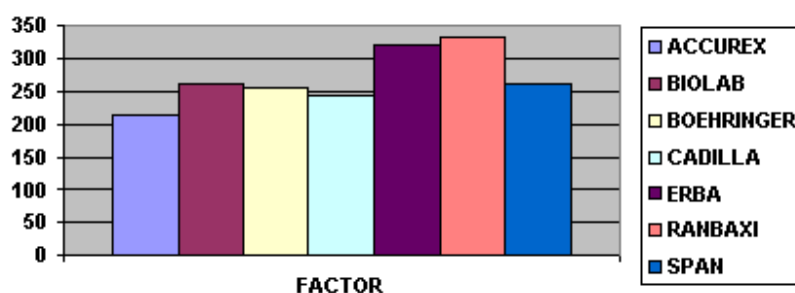
The means given by this method gave marked variation in results from 181 to 220 mg /dl.

6. Then I prepared my own standards by standard method. One gram of glucose (Analar) powder (after heating at 55^{0}C for 4 hrs) was weighed carefully. It was dissolved in 100 ml of 1 g/L Benzoic Acid solution. This was Stock Glucose

Standard. From this , the working standards containing 100, 150, 200, 250, 300, 400 and 500 mg /dl were prepared.

7. The step 5 was repeated again, substituting 100 mg working standard for the test plasma , with 20 tests for each reagent , and the same company's 100 mg standard for calibration. The mean value of the results were calculated.

8. This was followed for remaining 150 to 500 mg standards. All results were graphed.

OBSERVATION : There is a marked variation in the results, as seen from all these graphas. The personal, instrumental errors were reduced by doing the tests by only one and the same person, using the same instruments, and were done on one day.

	ONE SAMPLE 1	100 MG 2	150 MG 3	200 MG2 200 MGMGMMG	250 MG 5	300 MG 6	400 MG 7	500 MG 8
ACCUREX	212	102	169	222	291	341	645	525
BIOLAB	202	93	157	196	283	342	826	826
BOEHRINGER	190	93	151	226	281	353	637	935
CADILLA	189	94	132	175	228	277	385	468
ERBA	220	106	180	250	321	400	673	1111
RANBAXI	213	95	161	211	287	338	551	999
SPAN	181	90	144	206	251	319	524	527

IMPRESSION :

1. When the same test plasma was analysed, the results varied from 181 mg % to 220 mg % by different reagents.

2. For values between 100 to 300 mg %, the results agree *fairly* in Accurex, Biolab, Boehringer, Ranbaxi and Span , they are on lower side by Cadilla and on higher side by Erba.

3. In readings of 400 & 500 mg levels, NO REAGENT SHOWS ANY ACCURACY, whatever their claims are ! The 'Linearity Limit' printed is just for sake of printing. The real linearity limit appears to be between 200 and 250 mg % only. (After this experiment, I have challenged every company's representative to prove his company's claims about the Linearity Limit. NO COMPANY ACCEPTED THE CHALLENGE !)

NOTE : This is only one type of experiment, done to solve the problem of variation by different reagents. It also means that the above reagents ***at this time*** gave these reasults. ***THIS DOES NOT MEAN THAT A PARTICULAR REAGENT IS BETTER THAN OTHERS, OR A PARTICULAR REAGENT IS BAD !***

I will certainly welcome comments from other participants about such an experience of them.

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