PROLAC-CHECK-1

Quantitative determination of Prolactin in whole blood, plasma or serum samples

Ref. 9091

- FOR EASY READER® AND EASY READER+® USE ONLY -

I- PRINCIPLE

Human Prolactin (hPRL), which is secreted by the anterior lobe of the pituitary gland (1, 2), is essential for breast development and lactation in women. High levels can be detected after the eighth week of pregnancy and continue until term. After birth, prolactin levels return to normal within three weeks in the absence of breast feeding (3, 4). Normal females show prolactin levels only slightly higher than males (2, 3). Abnormally high levels of hPRL are associated with infertility in men and women, male impotence and primary hypothyroidism.

The PROLAC-CHECK-1 is a rapid, quantitative assay for the detection of human prolactin in whole blood, serum and plasma samples. The method employs a unique combination of monoclonal-dye conjugate and polyclonal-solid phase antibodies to identify prolactin in the test samples selectively with a high degree of specificity.

As the test sample flows through the absorbent device, the labelled antibody-dye conjugate binds to the prolactin forming an antibody-antigen complex. This complex binds to the anti-prolactin antibody in the reaction zone (T) and produces a rose-pink coloured band. In the absence of prolactin, there is no line in the reaction zone (T). The mixture continues flowing through the absorbent device, past the reaction zone (T) and control zone (C). Unbound conjugate binds to the reagents in the control zone (C), producing a rose-pink coloured band, demonstrating that the reagents are functioning correctly.

II- PROLAC-CHECK-1 KIT COMPONENTS

Each kit contains everything needed to perform 10 or 20 tests:

1- PROLAC-CHECK-1 reaction devices	10	20
2- Disposable plastic pipettes	10	20
3- Diluent in a dropper bottle	2.5 mL	5 mL
4- Instructions leaflet	1	1

5- **Controls (Optional):**

Positive control ref. V9000 and Negative control ref. V9001: a freeze-dried preparation of a non-infectious compound in diluted human serum, tested and found negative for anti-HIV, anti-HCV and HBs antigen, containing 0.05 % sodium azide is optionally available as a positive and negative control (1x 0.25 mL). The concentration range is indicated on the vial label.

III- STORAGE AND STABILITY

1- All PROLAC-CHECK-1 kit components, including optional control before reconstitution, should be stored at any temperature between +4°C and +30°C in their original package.
2- ***Do not freeze the test kit***.
3- The PROLAC-CHECK-1 test is stable until the expiry date stated on the package label.

IV- PRECAUTIONS

1- This test is designed for *in vitro* diagnostic use and professional use only.
2- Read carefully the instructions before using this test.
3- Handle all specimens as if they contained infectious agents. When the assay procedure is completed, dispose of specimens carefully after autoclaving them for at least one hour. Alternatively, they can be treated with 0.5% to 1% solution of sodium hypochlorite for one hour before disposal.
4- Wear protective clothing such as laboratory coats and disposable gloves while assaying samples.
5- Do not eat, drink or smoke in the area where specimens and kit reagents are handled.
6- Avoid any contact between hands and eyes or nose during specimen collection and testing.
7- Do not use beyond the expiry date which appears on the package label.
8- Do not use a test from a damaged protective wrapper.

V- SPECIMEN COLLECTION AND PREPARATION

1- PROLAC-CHECK-1 is to be performed on human serum, plasma or whole blood.
2- The specimen should be collected under the standard laboratory conditions (aseptically in such a way as to avoid haemolysis).
3- If anticoagulant is needed, only EDTA or heparin should be used.
4- Each specimen should be treated as if potentially infectious.
5- Whole blood samples should be tested immediately (< 4 hours). Finger prick samples should be assayed just after collection.
6- If the test is to be run within 48 hours after collection the specimen should be stored in the refrigerator (+2°C to +8°C). If testing is delayed more than 48 hours, the specimen should be frozen. The frozen specimen must be completely thawed, thoroughly mixed and brought to room temperature prior to testing. Avoid repeated freezing and thawing.
7- In case of cloudiness, high viscosity or presence of particulate matter into the serum specimen, it should be diluted with equal volume (V/V) of diluting buffer (not provided but available upon request) before testing.

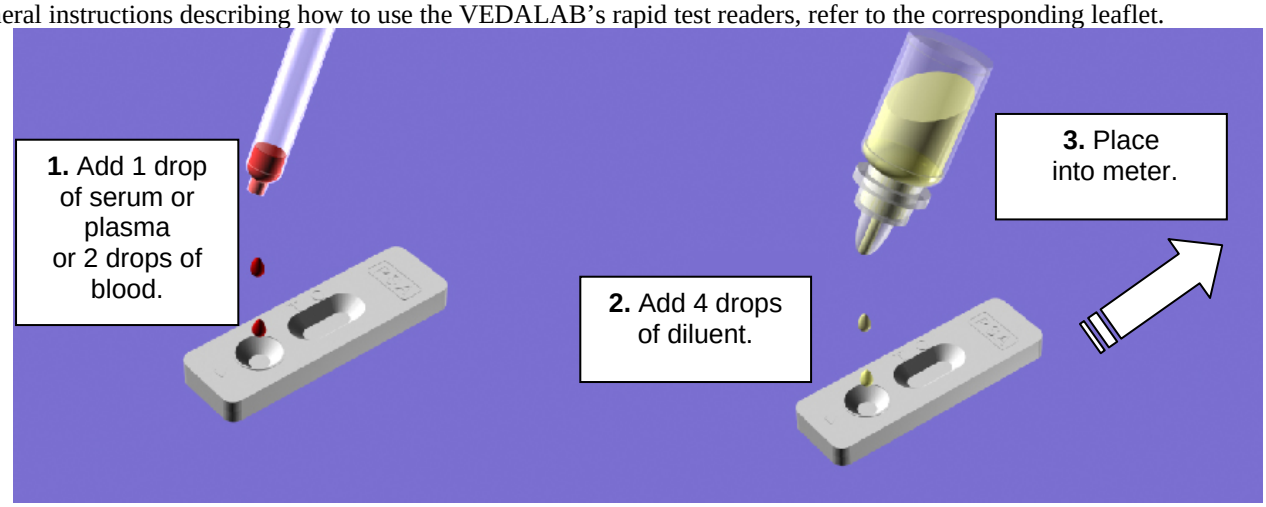
VI- ASSAY PROCEDURE

a) Controls testing
- Wait for 15 minutes after the freeze-dried control has dissolved.
- Add the requested volume (25µL) with **lab pipette (disposable tips)** into the sample well of the cassette and proceed in the same way as for a patient's sample.
- The expected concentration level (**in ng/mL**) is indicated on the vial label and obtained result must match the indicated value. The concentration level can change slightly depending on lot number.
- The reconstituted vial should be kept at +2°C to +8°C and should be used within 2 weeks after reconstitution.

b) Samples testing
Follow the below instructions or refer to the picture n°1.

1- Allow the sample and PROLAC-CHECK-1 test device to return to room temperature prior to testing.
2- Remove the "reaction device" from its protective wrapper by tearing along the line.
3- Label the device with the patient's name or control number.
4- Fill the serum dropper with specimen (serum or plasma) and by holding it vertically, dispense one drop (25 µL) into sample well. If whole blood is used, dispense 2 drops (50 µL) into the well (▷) **and wait for the blood sample to be completely absorbed before adding diluent**.
5- Add exactly 4 drops of diluent (150 µL) in the sample well (▷).
6- Read the result **(in ng/mL**) after 10 minutes either using the immediate or countdown reading mode (see corresponding leaflet).

For general instructions describing how to use the VEDALAB's rapid test readers, refer to the corresponding leaflet.



Picture n°1

VII- PERFORMANCES CHARACTERISTICS

a) Linearity
The measuring range is 20-350 ng/mL.
For Prolactin concentration below 20 ng/mL, the result will be given as "< 20 ng/mL".
For Prolactin concentration over 350 ng/mL, the result will be given as "> 350 ng/mL".
For samples whose concentration is higher than 350 ng/mL, dilute with saline and repeat the assay as per instructions of Part. VI.

b) Accuracy
A study has been performed using serum samples obtained from dilutions of the Prolactin international standard 84/500 (W.H.O.) covering a range of 0 to 350 ng/mL. Optical densities expressed as a function of Prolactin concentrations are described by following logarithmic curve:

$$Y = 7.959x / Ln (x) \quad (r = 0.961)$$

The results show a good correlation ($r > 0.95$) of the values obtained with PROLAC-CHECK-1 on VEDALAB's reader.

c) Sensitivity
Concentrations close to 10 ng/mL are detected by PROLAC-CHECK-1 test. In these cases, results will be rendered as "< 20 ng/mL". Levels higher than 20 ng/mL are generally considered as abnormal values.
The detection limit of the PROLAC-CHECK-1 rapid test has been checked according to W.H.O. 3rd international standard n° 84/500.

d) Precision

A panel of 40 human sera samples pre-assayed on the BAYER CENTAUR analyser (chemiluminescence) has been tested with PROLAC-CHECK-1 rapid test.

Results were read using the VEDALAB's reader. Results are shown in table I.

Table I

Human sera Identification SCIPAC panel (lot: 561-146)	Age	Sex	[PRL] in ng/mL Expected values BAYER CENTAUR	Confidence range		[PRL] in ng/mL Obtained values PROLAC-CHECK-1
				Lower Limit	Upper limit	
V011036475	33	F	1.4	1.12	1.68	<20
V011036482	40	F	5.5	4.4	6.6	<20
V011036483	38	F	6.1	4.88	7.32	<20
V011036480	30	F	8.3	6.64	9.96	<20
V011036481	34	F	11.4	9.12	13.68	<20
V011036477	36	F	11.7	9.36	14.04	<20
V011036476	42	F	12.6	10.08	15.12	<20
V011036479	33	F	12.9	10.32	15.48	<20
V011036478	26	F	13.5	10.8	16.2	<20
V011036474	34	F	13.6	10.88	16.32	<20
V011036492	**34**	**F**	**15.6**	**12.48**	**18.72**	**25.77**
V011036485	32	F	19.9	15.92	23.88	<20
V011036486	37	F	20	16	24	<20
V011036489	40	F	21.8	17.44	26.16	<20
V011036488	42	F	23.2	18.56	27.84	<20
V011036491	26	F	24.6	19.68	29.52	25.38
V011036490	26	F	27.2	21.76	32.64	24.6
V011036484	36	F	27.4	21.92	32.88	30.83
V011036493	25	F	27.5	22	33	24.6
V011036487	42	F	29.5	23.6	35.4	27.71
V011036503	38	F	30.2	24.16	36.24	30.05
V011036502	31	F	31.3	25.04	37.56	33.95
V011036499	34	F	31.9	25.52	38.28	36.68
V011036500	36	F	34.2	27.36	41.04	38.23
V011036501	34	F	34.4	27.52	41.28	27.71
V011036497	29	F	36.3	29.04	43.56	36.68
V011036498	30	F	39	31.2	46.8	37.84
V011036495	29	F	41.7	33.36	50.04	37.84
V011036494	41	F	46.3	37.04	55.56	37.06
V011036496	38	F	46.7	37.36	56.04	40.18
V011036508	33	F	50	40	60	42.91
V011036519	45	F	50.2	40.16	60.24	41.74
V011036516	33	F	50.5	40.4	60.6	48.75
V011036504	35	F	55.2	44.16	66.24	47.58
V011036517	35	F	55.2	44.16	66.24	46.81
V011036507	34	F	57	45.6	68.4	46.42
V011036505	31	F	60.8	48.64	72.96	50.78
V011036509	35	F	60.8	48.64	72.96	61.12
V011036518	31	F	60.8	48.64	72.96	49.53
V011036506	33	F	96.3	77.04	115.56	65.43
V011036506*****	33	F	73.70	58.96	88.44	65.43

*Sample pre-assayed by another method (ELFA VIDAS BIOMERIEUX) to check the titre.

A discrepancy is obtained with one serum sample (identified by bold typo):

-Serum V011036492: The result obtained by the both methods indicates probably the same clinical diagnosis profile (negative 15-26ng/mL).

Negative, borderline and positive samples are correctly detected (a correlation of 94.1% has been established between VEDALAB's rapid test and BAYER CENTAUR).

e) Intra-assay reproducibility

Within run precision was evaluated by using 26 replicates of two commercially available references containing 24.54 and 57.63 ng/mL of prolactin as determined with quantitative PROLAC-CHECK-1 for Easy Reader®.

The obtained CV (coefficient of variation) were respectively equal to 9.69% and 9.61%.

VIII- LIMITATIONS

1- PROLAC-CHECK-1 test is specifically designed to detect prolactin in whole blood, plasma or serum samples.

2- High levels of RF (Rheumatoid factor) or CRP (C-reactive protein) may create interferences and therefore lead to false positive results.

3- The test is designed to eliminate the potential interference of human antibodies to murine IgG (HAMA). However high level of HAMA could give falsely positive results.

4- **Use only fresh whole blood samples (< 4 hours) when test is performed with blood samples. Finger prick samples should be assayed just after collection.**

5- This format of test is to be only used with VEDALAB rapid test readers.

6- If the reading time (10 minutes) is not strictly respected, wrong results will be obtained.

7- This format of test should not be used for visual reading.

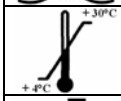
8- As it is true for any diagnostic method or for any measurements through analysers, there is a variability of the obtained result. Therefore, a confidence range of +/- 25% should be considered for the final value and for the clinical significance of the result.

IX- BIBLIOGRAPHY

1- Jeffcoate, SL, et al (1986), Assays for prolactin : guidelines for the provision of a clinical biochemistry service. Ann Clin Biochem, 2 3, 638-651.

2- Daughaday, WH (1985), The Anterior Pituitary. In Williams Textbook of Endocrinology. Ed Wilson, JD and Foster, DW, WB Saunders Co. Philadelphia, PA, 582-585.

3- Frantz, AG (1978), Physiology in Medicine : Prolactin . New Engl. J Med, 198, 201.

	Read the instructions before use		For *in vitro* diagnostic use
	Temperature limitations		Do not reuse
	Manufacturer		

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