

Analytical Verification of the IDS-iSYS Intact Amino-terminal Propeptide of Type I Procollagen (PINP) Automated Immunoassay



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Background:

PINP circulates as different forms: larger intact trimeric PINP and several fragment monomers. In healthy individual, the circulating PINP form is predominantly the trimeric intact with almost non-detectable monomers. Under certain conditions, especially in renal impaired and bedridden geriatric patients, the proportion of monomeric form is elevated. The IOF and IFCC recommended s-PINP and s-CTX are used as reference BTMs in observational and intervention studies. We seek to validate the analytical performance and establish reference intervals for apparently healthy children, pre-menopausal, post-menopausal women, and men for the IDS-iSYS Intact PINP assay.

Materials and methods:

The accuracy profile was determined with 8 serum pool levels (9.4 – 217.2 ng/mL). The method linearity was verified with two sets of high/low serum samples. Serum specimens from overnight fasting, apparently healthy Caucasians subjects, normal Calcium, Phosphorus, Intact PTH and eGFR > 60, were selected to generate the preliminary reference interval: 132 males, 102 pre-menopausal, 124 post-menopausal, children (0.3 – 20 years, 281 males, 295 males).

Results

Accuracy

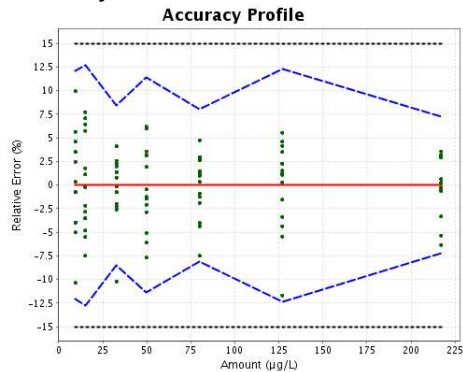


Fig. 1: IDS-iSYS Intact PINP accuracy profile.

The plain red line is the relative bias, the dash lines are the β expectation tolerance limits and the dotted lines represent the acceptable limits. The data points represent the relative error of the back-calculated concentrations and are plotted with respect to their targeted concentration.

The accuracy profile showed the risk that the possibility of a sample result outside of the $\pm 15\%$ acceptance limit was <5%.

Linearity

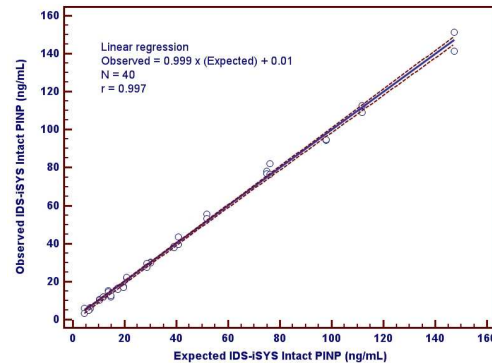


Fig. 2: IDS-iSYS Intact PINP linearity profile.

Two elevated samples were diluted 9/10, 8/10, 7/10, 6/10, 5/10, 4/10, 3/10, 2/10 and 1/10 with a samples containing very few amounts of PINP. The plots of the observed vs. expected PINP concentrations show a perfect linear relationship up to the 1/10 dilution.

Reference Intervals

Populations	No. of Subjects	Age (year) (Mean \pm SD)	Median (ng/mL)	SD (ng/mL)	Mean (ng/mL)	95% CI. (ng/mL)
Pre-Menopausal	107	36.8 \pm 8.0	30.6	12.1	33.5	7.4 - 57.1
Post-Menopausal	124	67.4 \pm 6.7	36.1	20.2	38.5	10.0 - 83.7
Males	138	57.4 \pm 16.4	36.7	18.5	39.8	11.4 - 78.6

Table 1: IDS-iSYS Intact PINP reference intervals for pre-menopausal, post-menopausal and males.

Age Group	No. of Subjects	Median (ng/mL)	SD (ng/mL)	Mean (ng/mL)	5 th - 95 th (ng/mL)
0 - <2	23	879	256	817	364 - 1188
2 - <4	28	556	186	512	194 - 757
4 - <6	35	580	178	587	304 - 966
6 - <8	31	596	159	616	390 - 937
8 - <10	31	712	237	727	394 - 1108
10 - <12	25	644	223	700	388 - 1094
12 - <14	29	272	184	332	82.0 - 650
14 - <16	29	102	77.4	130	53.8 - 300
16 - <18	33	70.0	31.9	75.0	25.1 - 148
18 - <20	31	48.2	28.1	54.7	24.1 - 118

Table 2: IDS-iSYS Intact PINP observed ranges for females <20 years.

Age Group	No. of Subjects	Median (ng/mL)	SD (ng/mL)	Mean (ng/mL)	5 th - 95 th (ng/mL)
0 - <2	30	740	161	748	472 - 1039
2 - <4	31	604	157	575	271 - 765
4 - <6	33	492	128	461	248 - 660
6 - <8	30	594	108	610	314 - 897
8 - <10	33	624	155	642	410 - 882
10 - <12	25	618	243	643	328 - 1169
12 - <14	24	772	301	764	194 - 1146
14 - <16	24	303	260	421	145 - 1015
16 - <18	25	144	125	196	66.7 - 436
18 - <20	26	92.4	83.6	110	27.2 - 34.3

Table 3: IDS-iSYS Intact PINP observed ranges for males <20 years.

Conclusion:

- The IDS-iSYS Intact PINP assay offers a reliable automated method for clinical laboratories.
- The pre-menopausal, post-menopausal, and males reference intervals will be useful tool for the monitoring of osteoporosis therapeutic treatment.
- Values from more than 500 children classified according to age and gender can be helpful to clinical laboratories and pediatricians.
- Further data are needed to fully assess the distribution of Intact PINP in children, especially between 8 – 14 years of age.

The 95% confidence level reference intervals for pre-menopausal, post-menopausal, and males was calculated (Table 1).

The 5th – 95th percentile observed ranges for females and males <20 years of ages were stratified by every 2 years (Tables 2 and 3).

Similar to other bone turnover markers, the Intact PINP concentration changes with age and menopause (Figure 3).

The levels are much higher in children than in adults, with peak between the ages of 8 to <12 for females, and 8 to <16 for males.

In this set of samples, the Intact PINP levels in females start the declining after 12 years of age. In males, the decreasing trend occurs 2 years later.

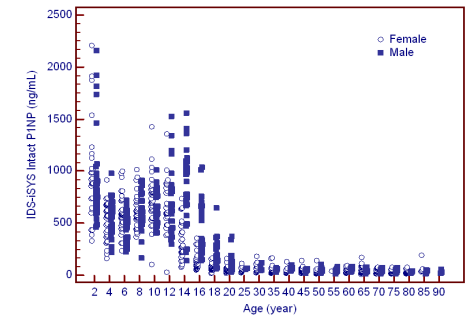


Fig. 3: Age and gender related changes in Intact PINP.