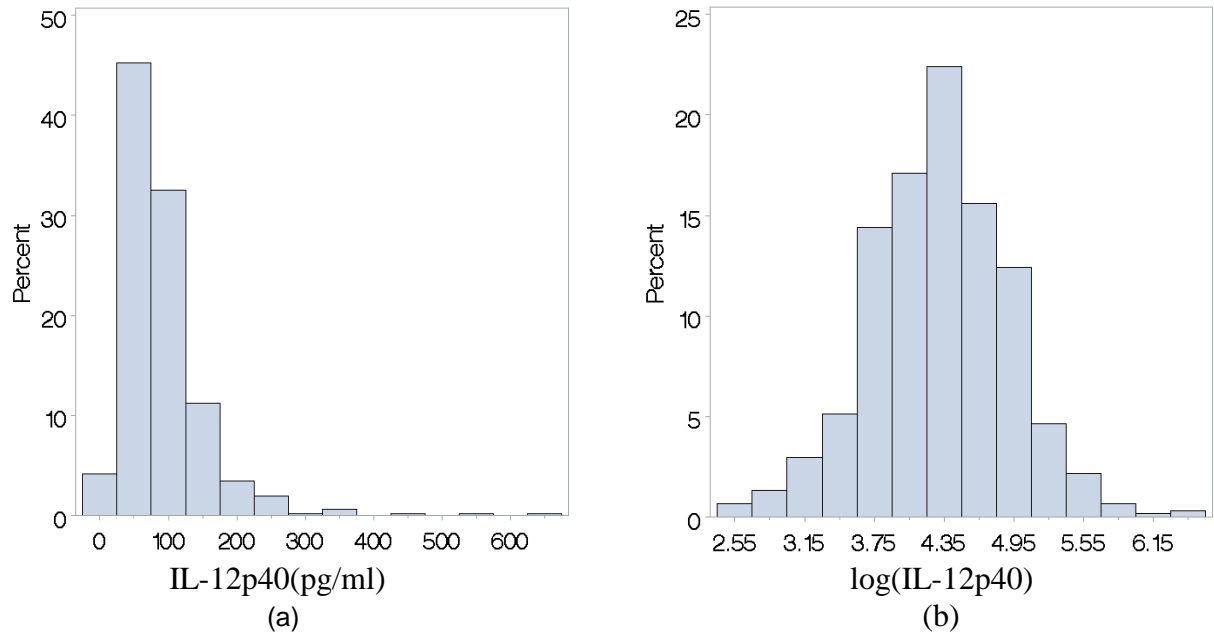
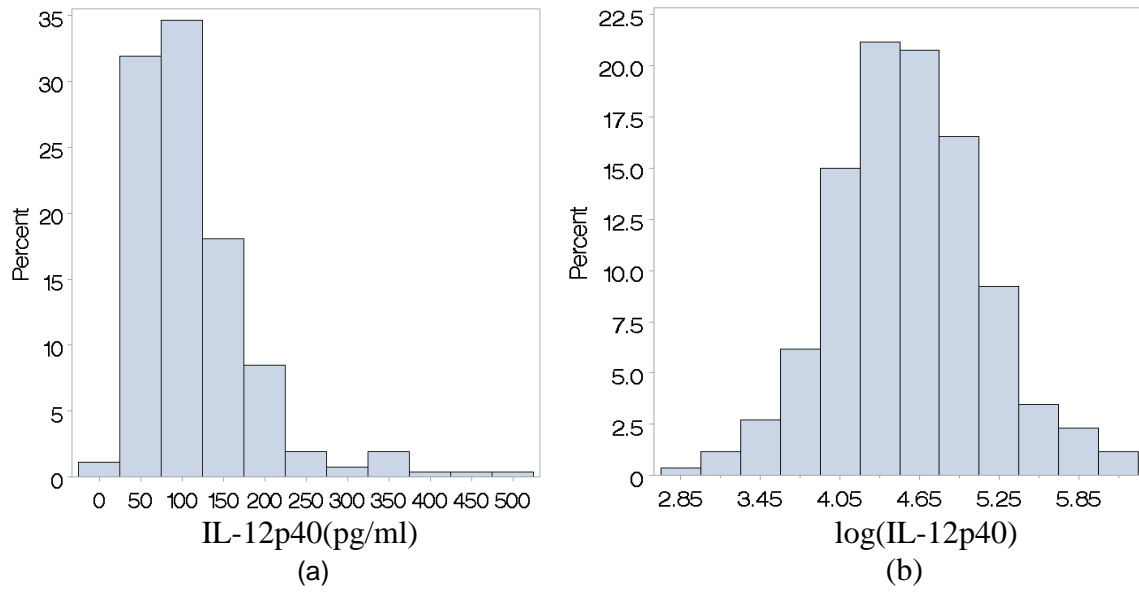


**The relationship between blood IL-12p40 level and melanoma progression**

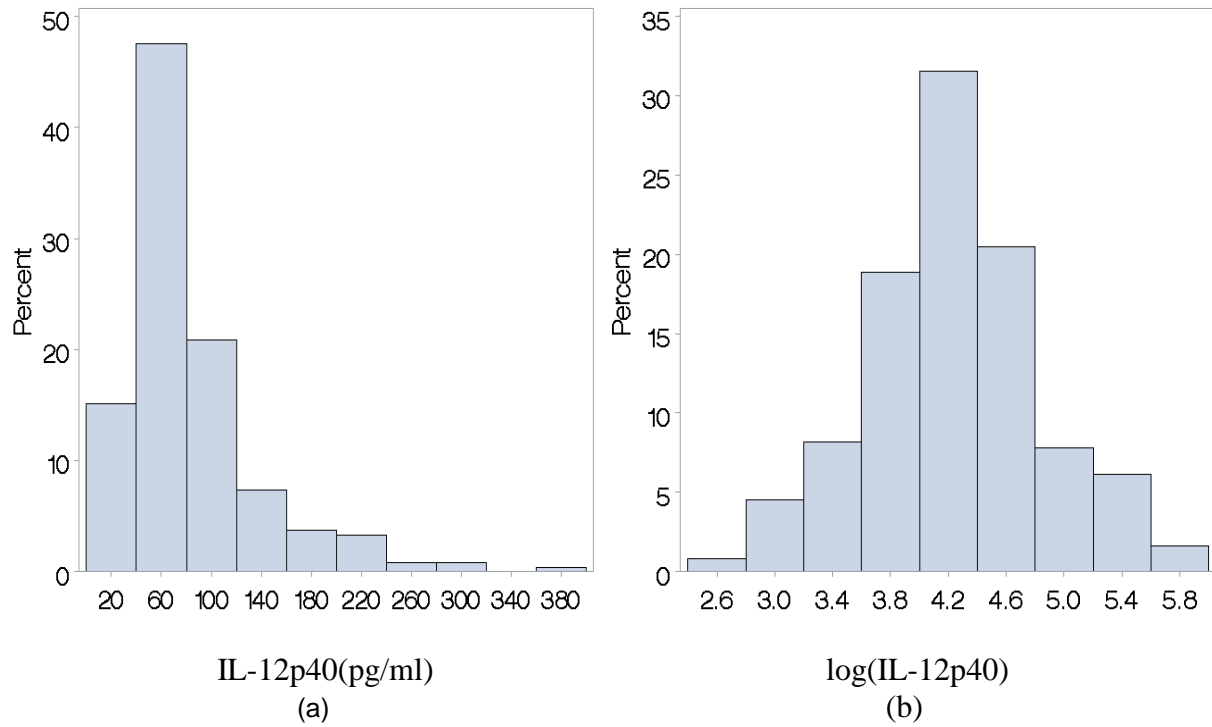
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Supplementary Figure 1. Histogram for raw IL-12p40 data(a, Skewness=3.27, kurtosis=19.00) and log-transformed IL-12p40 values(b, Skewness=0.04, kurtosis=0.47) in 573 melanoma cases (discovery).



Supplementary Figure 2. Histogram for raw IL-12p40 data(a, Skewness=2.20, kurtosis=6.74) and log-transformed IL-12p40 values(b, Skewness=0.10, kurtosis=0.41) in 249 melanoma cases (Case validation set 1).



Supplementary Figure 3. Histogram for raw IL-12p40 data(a, Skewness=1.86, kurtosis=4.64) and log-transformed IL-12p40 values(b, Skewness=-0.06, kurtosis=0.15) in 244 melanoma cases (Case validation set 2).

Supplementary Table 1. Serial IL-12p40 plasma levels from 66 patients who underwent at least 2 blood draws (median time between blood draws 6.3 months)\*

	N	Average days from 1st blood draw	IL-12p40(pg/ml)			log transformed IL-12p40 <sup>#</sup>	
			Mean	Standard Deviation	Median	Mean	Standard Deviation
Ist blood draw	66	0	121.020	78.778	97.964	4.616	0.606
2nd blood draw	66	596.2	136.650	107.644	108.392	4.682	0.684
3rd blood draw	36	806.5	161.458	140.311	121.026	4.777	0.810
4th blood draw	21	1107.3	96.830	54.895	83.457	4.404	0.627

\*Linear mixed model for the effect of time on log e of IL-12p40,  $P=0.1209$ .

# Correlation between duration and change of log transformed IL-12p40 Pearson  $r^2=0.14$ ,  $P=0.1097$ .

Supplementary Table 2. Comparison of log-transformed IL-12p40 levels between melanoma discovery and two validation sets, and between Control set and Validation set 1

Status	N	Mean	95% CI		Univariate <i>P</i> -value*
Case-discovery set	573	4.306	3.133	5.478	
Case-validation set 1	249	4.569	3.453	5.684	0.438
Case-validation set 2	244	4.247	3.052	5.441	0.200
Control set**	299	4.569	3.623	5.516	0.958

\* CI, confidence interval

\*\* Control set compared with Case-validation set 1 since IL-12p40 levels in those two sets were measured using the same assay kit in 2011; No difference in log-transformed IL-12p40 level between Control set and stage I/II Validation set 1 patients (N=106, mean=4.549, 95% CI= 4.440-4.658) (P=0.784)

Supplementary Table 3. Comparison of IL-12p40 plasma levels(pg/ml) from the same blood draw in 11 individuals tested by two scientists in 2007 and 2011 using different-generation assay kits\*.

	IL-12p40 (pg/ml)	IL-12p40 (pg/ml)
Lab ID	Scientist1_2007	Scientist2_2011
1657	21.818	21.856
1659	114.668	97.408
1696	69.645	81.681
1837	40.780	68.395
1905	144.769	171.354
2008	86.700	72.948
2440	125.080	182.826
2457	47.120	66.705
2496	174.010	189.058
2581	102.840	197.811
2612	147.414	179.625
Log e Mean±Std <sup>#</sup>	4.426±0.644	4.623±0.680

\*Pearson correlation  $r^2=0.87$ ,  $P=0.001$

# Log-transformed mean increase from 2007 to 2011, 4.5%

Supplementary Table 4. Comparison of IL-12p40 plasma levels(pg/ml) from the same blood draw in 11 individuals tested by the same scientist and same-generation assay kit in 2009 and 2011\*.

Lab ID	2009	2011
1657	25.812	21.856
1659	123.256	97.408
1696	71.668	81.681
1837	55.414	68.395
1905	165.227	171.354
2008	74.089	72.948
2440	171.187	182.826
2457	67.085	66.705
2496	179.572	189.058
2581	101.324	197.811
2612	164.227	179.625
Log e		
Mean $\pm$ Std <sup>#</sup>	4.548 $\pm$ 0.607	4.623 $\pm$ 0.680

\*Wilcoxon matched-pairs signed-rank test  $P=0.1307$ .

\*Pearson correlation  $r^2=0.88$ ,  $P<0.001$

# Log-transformed mean increase from 2009 to 2011, 1.6%



Supplementary Table 5. Association of clinical factors with melanoma outcome in the univariate analysis\*

Variable	DFS		MSS		OS	
	HR(95%CI)	<i>P</i> -value	HR(95%CI)	<i>P</i> -value	HR(95%CI)	<i>P</i> -value
All(N=1066)						
Stage#	-	-	2.11(2.59-3.73)	<0.0001	2.47(2.12-2.88)	<0.0001
Stage I/II 1(N=751)						
Tumor thickness	1.11(1.08-1.14)	<0.0001	1.11(1.07-1.16)	<0.0001	1.10(1.06-1.14)	<0.0001
Ulceration(Present VS not identified)	4.67(3.02-7.21)	<0.0001	5.90(3.28-10.60)	<0.0001	3.40(2.11-5.50)	<0.0001
Mitosis(>=1 VS <1)	9.56(4.16-22.00)	<0.0001	9.17(2.84-29.64)	0.0002	4.09(2.09-7.98)	<0.0001
Stage#	4.88(3.33-7.14)	<0.0001	6.23(3.56-10.90)	<0.0001	3.94(2.56-6.06)	<0.0001

\* CI, confidence interval

# Stage coded as a continuous variable(I=1, I/II=1.5, II=2, III=3, IV=4)

Supplementary Table 6. Meta-analysis of the relationship between per 10 unit change of IL12p40 and survival in the merged dataset

Analysis	<i>P</i> -value <sup>a</sup>	<i>P</i> ( <i>R</i> )-value <sup>b</sup>	HR <sup>a</sup>	HR( <i>R</i> ) <sup>b</sup>	Q	I
<b>Overall survival</b>						
Univariate-All	6.44 × 10 <sup>-7</sup>	6.44 × 10 <sup>-7</sup>	1.0354	1.0354	0.7274	0
Multivariate-all	3.01 × 10 <sup>-5</sup>	2.62 × 10 <sup>-3</sup>	1.0336	1.0347	0.1892	39.95
Univariate-stage I/II	8.02 × 10 <sup>-7</sup>	4.21 × 10 <sup>-5</sup>	1.0466	1.0504	0.2854	20.24
Multivariate-stage I/II	8.78 × 10 <sup>-7</sup>	2.85 × 10 <sup>-3</sup>	1.0533	1.072	0.0732	61.75
Univariate-stage III/IV	1.04 × 10 <sup>-1</sup>	1.04 × 10 <sup>-1</sup>	1.0211	1.0211	0.4037	0
Multivariate-stage III/IV	2.00 × 10 <sup>-1</sup>	3.10 × 10 <sup>-1</sup>	1.0167	1.0174	0.2608	25.6
<b>Melanoma specific survival</b>						
Univariate-All	7.59 × 10 <sup>-5</sup>	7.59 × 10 <sup>-5</sup>	1.0329	1.0329	0.8529	0
Multivariate-all	3.97 × 10 <sup>-4</sup>	3.97 × 10 <sup>-4</sup>	1.0336	1.0336	0.4933	0
Univariate-stage I/II	3.04 × 10 <sup>-4</sup>	3.04 × 10 <sup>-4</sup>	1.0454	1.0454	0.4052	0
Multivariate-stage I/II	3.35 × 10 <sup>-5</sup>	5.27 × 10 <sup>-3</sup>	1.0593	1.0757	0.1143	53.9
Univariate-stage III/IV	6.69 × 10 <sup>-2</sup>	6.69 × 10 <sup>-2</sup>	1.0239	1.0239	0.6842	0
Multivariate-stage III/IV	1.25 × 10 <sup>-1</sup>	1.25 × 10 <sup>-1</sup>	1.0203	1.0203	0.4837	0
<b>Disease-free survival</b>						
Univariate	4.21 × 10 <sup>-4</sup>	4.21 × 10 <sup>-4</sup>	1.0342	1.0342	0.963	0
Multivariate	8.48 × 10 <sup>-5</sup>	8.48 × 10 <sup>-5</sup>	1.0413	1.0413	0.9252	0

a. *P*-value and hazard ratio[HR] for the effect of IL-12p40 on outcome in the combined data set using fixed effect model ;

b. *P*-value and hazard ratio(*R*)[HR(*R*)] for the effect of IL-12p40 on outcome in the combined data set using random effect model;