Supplementary data

Supplementary patients and methods

Recruitment of patients and family members

Psoriasis vulgaris (PsV) and psoriasis arthritis (PsA) patients were diagnosed by board certified dermatologists and rheumatologists mainly at university hospitals in Germany as previously described (Lohr et al., 2016); study groups were similar to the ones given in (Lohr et al., 2019). Individuals affected by GPP and PPP were diagnosed considering the ERASPEN consensus criteria as shown in (Haskamp et al., 2020). SAPHO syndrome patients were a subset of 52 patients presented in (Assmann et al., 2020). Family members of eight previously published patients with GPP (n=7) or AGEP (n=1) (Haskamp et al., 2020) were contacted via the index patient and recruited after agreeing to the study; their diagnoses were self-reported by the index patient.

The study was approved by the ethical committee of the Friedrich-Alexander-Universität Erlangen-Nürnberg and the ones of further involved universities. Written informed consent was obtained from each affected and control individual before enrolment. We conducted investigations according to Declaration of Helsinki principles.

Genotyping of patients, family members and control individuals

We used single TaqMan genotyping assays, microarrays and Sanger sequencing to genotype seven variants in the *MPO* gene.

Available family members of eight of the previously published GPP/ AGEP patients with *MPO* variants were sequenced by Sanger as described previously (Haskamp et al., 2020).

We genotyped the four variants c.752T>C/p.(Met251Thr), c.995C>T/p.(Ala332Val), c.1555_1568del/p.(Met519Profs*21), c.2031-2A>C/r.[2030_2031ins[2031-109_2031-1],2030_2031ins[2030+1_2031-1] using predesigned TaqMan assays (n=3) or a self-designed assay (Supplementary Table 7) (Life Technologies, Carlsbad, CA, U.S.A.) as described previously (Huffmeier et al., 2009). DNAs of four GPP patients with rare *MPO* variants were included in the analyses as positive controls. DNAs of all missing genotypes were sequenced by Sanger resulting in genotyping rates of 100%.

Additionally, we selected four *MPO* variants that were directly genotyped on global screening arrays (GSA; gsamd-24v1-0_20011747_a1, Illumina, San Diego, CA, USA) in 2,725 individuals (1,328 PsV, 1,344 PsA, 6 PPP, 47 SAPHO syndrome). Genotype plots were visually assessed; thereby we excluded c.1642C>T/p.(Arg548Trp) due to unreliable clustering of the three genotypes (Supplementary Figure 2B). Plots of the remaining three variants c.752T>C/p.et251Thr), c.1705C>T/p.(Arg569Trp) and c.2031-2A>C are shown in Supplementary Figure 2. We included probands with a microarray-wide genotyping rate of ≥98%. Distributions of SNPs fulfilled Hardy-Weinberg equilibrium using a threshold of 0.001. All genotypes of rare *MPO* variants obtained by one of the two SNP genotyping methods were confirmed by Sanger sequencing. We could compare 7,338 genotypes (3 SNPs in 2,446 DNAs (1,073 PsV, 1,321 PsA, 4 PPP, 48 SAPHO syndrome)) that were genotyped with both TaqMan and microarray and obtained concordance rates of 100% (c.752T>C/p.(Met251Thr) using two primer pairs), 100% (c.1705C>T/p.(Arg569Trp)) and 99.81% (c.2031-2A>C). Genotypes for c.2031-2A>C were discordant in six individuals genotyped by microarray; Sanger sequencing of all mismatches indicated heterozygosity, but also for a neighboring variant (c.2031-6A>G) probably resulting in false homozygous states of c.2031-2A>C. Allele frequencies obtained by Taqman and microarray were comparable (Supplementary Table 8).

Three SNPs - c.1642C>T/p.(Arg548Trp), c.1705C>T/p.(Arg569Trp) and c.1768C>T/p.(Arg590Cys) - located to exon 10 were sequenced by Sanger in all patient groups.

The overall genotyping rate of seven coding variants in *MPO* in 3,663 individuals was 100%. We gained genotypes of less than seven SNPs from additionally 441 patients in order to calculate associations at single SNP level.

Analysis of two *IL36RN* variants in pedigree VI

A PCR product encompassing the two *IL36RN* coding variants was cloned form probands' DNAs into a vector and transformed into competent E. coli as described previously (Mossner et al., 2018). Colonies were sequenced for the two variants by Sanger. In both carriers, one of the two variants per colony was detected indicating compound heterozygosity.

Statistical analysis

For data analysis, we used Plink and the R package vcfR (Knaus and Grunwald, 2017, Purcell et al., 2007) (R-Core-Team, 2013). To assess differences in cumulative allele frequency between cases and controls, we used one Fisher's exact test per psoriatic subtype; the shown p-values were not adjusted for the number of psoriatic subtypes analyzed. The same test was used for the association analysis of psoriatic subtypes with single *MPO* variants and the stratification analysis for family history of psoriasis dependent on carrier-ship of *MPO* variants. To test whether mutations in *MPO* or *MPO* and *IL36RN* predict affection status, we performed a logistic regression analysis and determined the p-value by ANOVA. The same model was used, when we tested the dependency of the MPO genotype (wildtype vs. mutant) and the severity of disease. To test for dependency of age of onset and number of *MPO* variants, we fitted a linear

model and used ANOVA to compute p-values as described previously (Haskamp et al., 2020).

Supplementary Table 1: Numbers and allele frequencies of *MPO* variants in largest possible number of psoriasis patients (four subtypes: PsV: n=1,227-1,488, PsA: n=1,317-1,358, PPP: n=271, SAPHO syndrome: n=52) and control individuals (938-962 German control individuals, 63,565-64,591 Non-Finnish European (NFE) individuals of gnomAD (Karczewski et al., 2020)) and results of association analyses. Frequency distributions of the missense or splice variants in patient and control groups were compared using a Fisher's exact test with p-values not adjusted for no. of tests.

Variant at nucleotide/ protein level (degree of MPO-deficiency by rare variant)	PsV n (%)	p	PsA n (%)	p	PPP n (%)	p	SAPHO n (%)	p	control indiv. n (%)	NFE indiv. n (%)	Σ control indiv. n (%)
c.752T>C/ p.(Met251Thr) (P)	38/2,736 (1.39)	n.s.	40/2,710 (1.48)	n.s.	8/542 (1.48)	n.s.	2/104 (1.92)	n.s.	25/1,908 (1.31)	1,735/127,130 (1.36)	1,760/129,038 (1.36)
c.995C>T/ p.(Ala332Val) (P)	36/2,464 (1.46)	n.s.	39/2,672 (1.46)	n.s.	19/542 (3.51)	0.008	1/104 (0.96)	n.s.	34/1,900 (1.79)	2,314/129,174 (1.79)	2,348/131,074 (1.79)
c.1555_1568del/ p.(Met519Profs*21) (C)	8/2,458 (0.33)	0.032	6/2,634 (0.23)	n.s.	2/542 (0.37)	n.s.	1/104 (0.96)	n.s.	2/1,876 (0.11)	189/129,180 (0.15)	191/131,056 (0.15)
c.1642C>T/ p.(Arg548Trp) (C)	0/2,976 (0)	n.s.	0/2,716 (0)	n.s.	0/542 (0)	n.s.	0/104 (0)	n.s.	1/1,924 (0.05)	67/129,140 (0.05)	68/131,064 (0.05)
c.1705C>T/ p.(Arg569Trp) (C)	10/2,976 (0.34)	n.s.	6/2,716 (0.22)	n.s.	3/542 (0.55)	n.s.	1/104 (0.96)	n.s.	3/1,924 (0.16)	370/129,182 (0.29)	373/131,106 (0.28)

Variant at nucleotide/ protein level (degree of MPO-deficiency by rare variant)	PsV n (%)	p	PsA n (%)	p	PPP n (%)	p	SAPHO n (%)	p	control indiv. n (%)	NFE indiv. n (%)	Σ control indiv. n (%)
c.1768C>T/ p.(Arg590Cys) (C)	0/2,454 (0)	n.s.	0/2,674 (0)	n.s.	0/542 (0)	n.s.	0/104 (0)	n.s.	0/1,924 (0.0)	9/129,050 (0.01)	9/130,974 (0.01)
c.2031-2A>C (C)	18/2,736 (0.66)	n.s.	15/2,710 (0.55)	n.s.	4/542 (0.74)	n.s.*	1/104 (0.96)	n.s.	11/1,892 (0.58)	936/128,900 (0.73)	947/130,792 (0.72)

C = complete, indiv. = individuals, n = number of alleles/ overall number of alleles in respective study group, n.s. = not significant (p-value>0.2), P = partial, p = p-value, *p-value =0.8

Supplementary Table 2: Genotypes and clinical data of eleven patients (PsV: n=6, PsA: n=3, PPP: n=2) and ten control individuals with two *MPO* variants. The degree of MPO-deficiency was estimated based on functional studies of the missense variants described previously (Haskamp et al., 2020, Marchetti et al., 2004, Nauseef et al., 1994, Romano et al., 1997). Severity of PsV was assessed by dermatologists based on PASI score and general dermatological symptoms and classified in the three categories of mild, intermediate and severe disease.

Psoriasis subtype/ control group	Genotype	estimated MPO-deficiency in %	Age of onset	Family history of psoriasis	Severity of disease	Details on manifestations, concomitant psoriatic disease
PsV	c.2031-2A>C + c.752T>C/ p.(Met251Thr)	90%	39	unknown	unknown	plaques at hands
PsV	c.995C>T/ p.(Ala332Val) + c.1705C>T/ p.(Arg569Trp)	83%	unknown	unknown	unknown	unknown
PsV	c.995C>T/ p.(Ala332Val) + c.1555_1568del/ p.(Met519Profs*21)	83%	27	negative	mild	nails affected
PsV	c.995C>T/ p.(Ala332Val) + c.1555_1568del/ p.(Met519Profs*21)	83%	20	positive	unknown	unknown
PsV	c.752T>C/ p.(Met251Thr) + c.995C>T/ p.(Ala332Val)	73%	25	positive	intermediate	nail dystrophy
PsV	homoz. c.995C>T/ p.(Ala332Val)	65%	23	positive	mild	toenails affected
PsA	homoz. c.1555_1568del/ p.(Met519Profs*21)	100%	10	negative	n.a.	polyarthritis; PsV
PsA	c.2031-2A>C + c.752T>C/ p.(Met251Thr)	90%	21	negative	n.a.	oligoarthritis; hands affected; PsV
PsA	homoz. c.752T>C/ p.(Met251Thr)	80%	22	positive	n.a.	enthesitis; polyarthritis; hands affected; PsV

PPP	c.995C>T/ p.(Ala332Val) + c.1555_1568del/ p.(Met519Profs*21)	83%	46	positive	n.a.	number of pustules >60; hands and palms affected; nails affected; PsV capitis
PPP	c.995C>T/ p.(Ala332Val) + c.1555_1568del/ p.(Met519Profs*21)	83%	20	negative	n.a.	number of pustules >60; hands and palms affected; nails affected; PsV capitis
German control	c.2031-2A>C + c.752T>C/ p.(Met251Thr)	90%				
German control	c.2031-2A>C + c.752T>C/ p.(Met251Thr)	90%				
German control	c.752T>C/ p.(Met251Thr) + c.995C>T/ p.(Ala332Val)	73%				
in-house exomes	c.2031-2A>C + c.1907C>T/ p.(Thr636Met)	100%				
in-house exomes	c.2031-2A>C + c.1907C>T/ p.(Thr636Met)	100%				
in-house exomes	c.752T>C/ p.(Met251Thr) + c.1555_1568del/ p.(Met519Profs*21)	90%				
in-house exomes	homoz. c.325delA/ p.(A108fs5)	100%				
1000 Genomes	c.1642C>T/ p.(Arg548Trp) + c.518A>G/ p.(Tyr173Cys)	100%				
1000 Genomes	c.2031-2A>C + c.752T>C/ p.(Met251Thr)	90%				
1000 Genomes	homoz. c.752T>C/ p.(Met251Thr)	80%				

homoz. = homozygous, n.a. = not applicable

Supplementary Table 3: Numbers and genotype frequencies of *MPO* variants in 2,741 psoriasis patients (four subtypes: PsV: n=1,105, PsA: n=1,313, PPP: n=271, SAPHO syndrome: n=52) and 3,759 control individuals (922 German control individuals, 404 individuals of non-Finnish European ancestry from the 1000 Genomes Project (Genomes Project et al., 2015), 2,433 in-house exome sequences probands described previously (Haskamp et al., 2020)). To test whether mutations in MPO predict affection status, we performed a logistic regression analysis and determined the p-value by ANOVA. The p-value was not significant.

No. of *MPO* variants	**PsV n (%)**	**PsA n (%)**	**PPP n (%)**	**SAPHO syndrome n (%)**	**2,741 psoriasis patients n (%)**	**3,759 control indiv. n (%)**
2	6 (0.54)	3 (0.23)	2 (0.74)	0 (0)	11 (0.40)	10 (0.27)
1	75 (6.79)	97 (7.39)	24 (8.86)	6 (11.54)	202 (7.37)	285 (7.58)
0	1,024 (92.67)	1,213 (92.38)	245 (90.41)	46 (88.46)	2,528 (92.23)	3,464 (92.15)
					p-value: 0.94	

n = number of patients, indiv. = individuals

Supplementary Table 4: Numbers and genotype frequencies of *MPO* and *IL36RN* variants in 1,771 psoriasis patients (four subtypes: PsV: n=708, PsA: n=759, PPP: n=253, SAPHO syndrome: n=51) and 3,759 control individuals (922 German control individuals, 404 individuals of Non-Finnish European ancestry of the 1000 Genomes Project, 2,433 in-house exome sequences). To test whether mutations in MPO and IL36RN predict affection status, we performed a logistic regression analysis and determined the p-value by ANOVA. The p-value was not significant.

No. of *MPO* & *IL36RN* variants	**PsV n (%)**	**PsA n (%)**	**PPP n (%)**	**SAPHO syndrome n (%)**	**1,771 psoriasis patients n (%)**	**3,759 control indiv. n (%)**
2	4 (0.56)	1 (0.13)	2 (0.79)	0 (0)	7 (0.4)	15 (0.4)
1	58 (8.2)	62 (8.17)	28 (11.07)	8 (15.69)	156 (8.81)	310 (8.25)
0	646 (91.24)	696 (91.7)	223 (88.14)	43 (84.31)	1,608 (90.8)	3,434 (91.35)
					p-value: 0.53	

n = number of patients, indiv. = individuals

Supplementary Table 5: Severity of PsV depending on the number of *MPO* variants in 580 patients. Severity of disease was assessed by dermatologists based on PASI score and general dermatological symptoms and classified in the three categories of mild, intermediate and severe disease. The p-value in the range of marginally significant association was obtained by regression analysis of *MPO* genotype (wildtype vs. mutant) and severity of PsV.

	Severity of PsV			
No. of *MPO* variants	**mild n (%)**	**intermediate n (%)**	**severe n (%)**	**p-value**
0	200 (90.5)	232 (93.17)	105 (95.45)	0.09
1	20 (9.05)	16 (6.43)	5 (4.55)	
2	1 (0.45)	1 (0.4)	0 (0)	

n = number of patients

Supplementary Table 6: Stratification of 2,488 psoriasis patients with information on family history of psoriasis (PsV: n=1,214 with, PsA: n=1,054, PPP: n=192, SAPHO syndrome: n=28) and association analyses according to presence/ absence of *MPO* variants.

Psoriasis subtype	family history of psoriasis				P-value
	negative		positive		
	Patients without *MPO* variant n (%)	Patients with *MPO* variant n (%)	Patients without *MPO* variant n (%)	Patients with *MPO* variant n (%)	
PsV	621 (93.2)	44 (6.6)	507 (92.3)	42 (7.7)	n.s.
PsA	588 (93.2)	43 (6.8)	388 (91.9)	35 (8.3)	n.s.
PPP	108 (85.7)	18 (14.3)	58 (87.9)	8 (12.1)	n.s.
SAPHO syndrome	20 (87)	3 (13)	4 (80)	1 (20)	n.s.
Σ	1,337 (92.5)	108 (7.5)	957 (91.8)	86 (8.2)	n.s.

n = number of patients, Σ = sum, n.s. = not significant

Supplementary Table 7: Details of predesigned and self-designed TaqMan genotyping assays.

rsID	Variant at nucleotide/ protein level	Taqman assay ID / Probes & Primers
rs56378716	c.752T>C/ p.(Met251Thr)	C__27531740_20
rs28730837	c.995C>T/ p.(Ala332Val)	C__25922741_20
rs963955387	c.1555_1568del/ p.(Met519Profs*21)	Probes: AGCCCATGGAACC for wildtype (VIC-labeled) CAGCCCCCGTGTC for deletion (FAM-labeled) Primers: forward GTCTTCACCAATGCCTTCCG reverse CTGGCCTAGGTCCTGCTTAC
rs35897051	c.2031-2A>C/	C__60526215_10

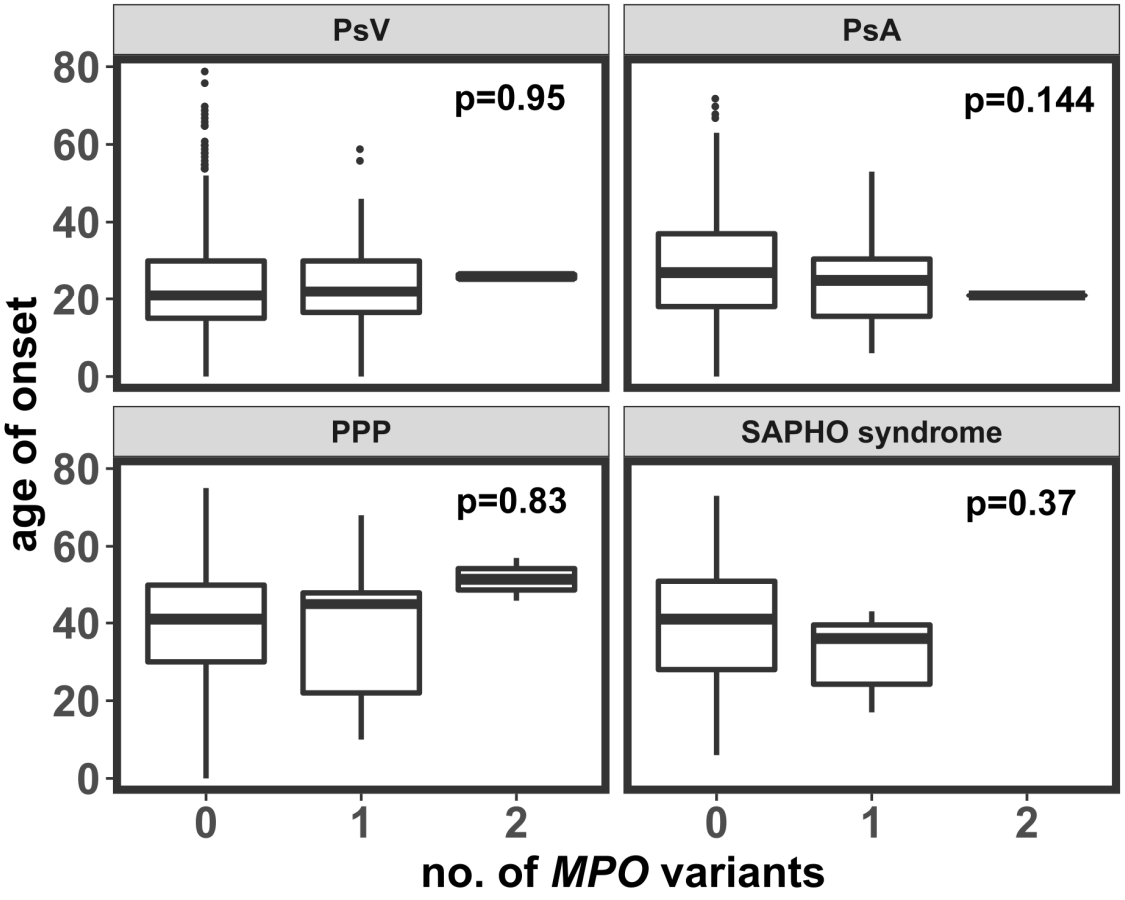
Supplementary Table 8: Comparison of allele frequencies obtained by single SNP genotyping (PsV: number of patients=1,105; PsA: n=1,313) and microarray analysis (PsV: n=1,328; PsA: n=1,344).

Variant at nucleotide/ protein level	single SNP genotyping				microarray			
	PsV		PsA		PsV		PsA	
	rare alleles n (%)	wt alleles n (%)	rare alleles n (%)	wt alleles n (%)	rare alleles n (%)	wt alleles n (%)	rare alleles n (%)	wt alleles n (%)
c.752T>C/ p.(Met251Thr)	27 (1.22)	2,181 (98.78)	38 (1.45)	2,588 (98.55)	37 (1.39)	2,619 (98.61)	40 (1.49)	2,648 (98.51)
c.1705C>T/ p.(Arg569Trp)	5 (0.23)	2,203 (99.77)	6 (0.23)	2,620 (99.77)	7 (0.26)	2,649 (99.74)	7 (0.26)	2,681 (99.74)
c.2031-2A>C	15 (0.68)	2,193 (99.32)	15 (0.57)	2,611 (99.43)	16 (0.6)	2,640 (99.40)	15 (0.56)	2,673 (99.44)

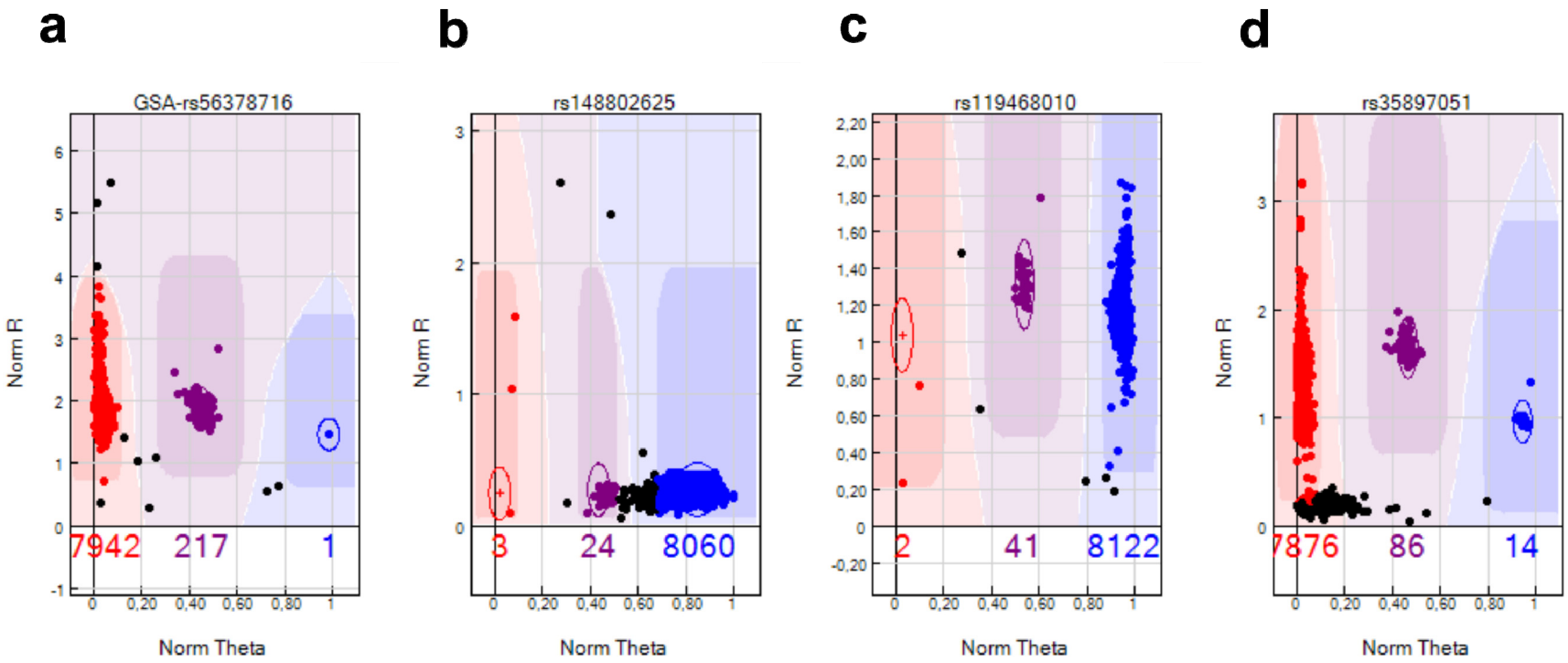
n = number of alleles, wt= wildtype

Supplementary Figures

Supplementary Figure 1: Correlation of age of onset and number of *MPO* variants in 1,647 psoriasis patients (four subtypes: PsV: n=1,003; PsA: n=1,145; PPP: n=201; SAPHO syndrome: n=48). The bold horizontal line of the boxplots corresponds to the median, the upper and lower line of the boxes to the 75th and 25th percentile, respectively. The whisker lines extend the boxes to the most extreme values, but less than 1.5fold of the inter-quartile range. Outliers are indicated as dots. We fitted a linear model (age/ mutations) and calculated p-values by ANOVA. There was no significant correlation in each subgroup, and when summarized (data not shown, p=0.65).



Supplementary Figure 2: Plots of genotypes of four *MPO* variants: a c.752T>C/ p.(Met251Thr) (rs56378716), **b** c.1642C>T/ p.(Arg548Trp) (rs148802625), **c** c.1705C>T/ p.(Arg569Trp) (rs119468010) and **d** c.2031-2A>C (rs35897051) generated by microarray analysis. We used polar coordinate system with the radius on the x-axis and the polar angle in radians on the y-axis. Clusters of genotypes are colored in blue or red (homozygous alleles, respectively) or purple (heterozygous). Circles indicate genotype calls of high confidence. Black genotypes (dots) could not be assigned to a genotype cluster. Genotyping in B for c.1642C>T/ p.(Arg548Trp) (rs148802625) was unreliable and therefore not considered for further analysis.



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