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DIAGNOSTIC AND THERAPEUTIC METHODS FOR THE TREATMENT OF RHEUMATOID ARTHRITIS (RA)

Abstract

The present invention provides prognostic, predictive, and therapeutic methods for the treatment of rheumatoid arthritis (RA). The invention is based, at least in part, on the discovery that the expression level of one or more biomarkers described herein in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) from an individual having RA can be used in methods of determining whether an individual having RA is likely to exhibit disease progression, identifying an individual having RA who is likely to respond to a treatment including a disease modifying anti-rheumatic drug (DMARD), predicting responsiveness of an individual having RA to a treatment including a DMARD, selecting a therapy for an individual having RA, and treating an individual having RA, as well as related kits.

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Background/Summary

SEQUENCE LISTING

[0001] The instant application contains a Sequence Listing, which has been submitted electronically in XML format and is hereby incorporated by reference in its entirety. Said XML copy, created on Apr. 28, 2025, is named 50474-175003_Sequence_Listing_4_28_25 and is 62,383 bytes in size.

FIELD OF THE INVENTION

[0002] The present invention is directed to diagnostic and therapeutic methods for the treatment of rheumatoid arthritis (RA). In particular, the invention provides methods for prognosing disease progression and activity, methods of predicting therapeutic responsiveness, methods of monitoring responsiveness to treatment, methods of selecting a treatment, methods of treatment, and diagnostic kits.

BACKGROUND OF THE INVENTION

[0003] Rheumatoid arthritis (RA) is an autoimmune disease of unknown etiology characterized by symmetrical joint involvement, inflammation, synovial lining hyperplasia, and formation of invasive granulation tissue. If uncontrolled, the disease leads to eventual joint destruction accompanied by significant morbidity and increased mortality. The disease is heterogeneous with respect to its clinical and biological presentation, as well as in its response to treatment.

[0004] It is generally thought that RA is initiated by a breach of immune tolerance, in particular, to citrullinated self-antigens that can occur years before the onset of clinical symptoms. Multiple cellular players have been identified to contribute to the pathophysiology of RA, including autoreactive T cell and B cell accumulation in the synovium, accompanied by production of autoantibodies directed at a variety of joint antigens, infiltration of inflammatory macrophages into the synovial lining and sublining, and elevated production of cytokines and chemokines that serve to recruit, activate, and sustain synovitis.

[0005] Multiple therapeutic approaches have been shown to be highly effective at managing RA symptoms and improving prognosis, including disease modifying anti-rheumatic drugs (DMARDs), such as methotrexate and leflunomide, and biologic therapeutic agents targeting TNFα, IL-6, T cell co-stimulation, B cells, and Janus kinases. However, predicting which individuals would benefit optimally from different therapeutic modalities has been challenging due to heterogeneity of RA.

[0006] Thus, there exists an unmet need for improved diagnostic and therapeutic methods for the treatment of individuals having RA.

SUMMARY OF THE INVENTION

[0007] The present invention provides diagnostic methods, therapeutic methods, and kits for treating an individual having rheumatoid arthritis (RA).

[0008] In a first aspect, the invention features a method of predicting disease progression in an individual having RA, the method comprising determining an expression level of one or more genes set forth in Table 1 in a sample from the individual, wherein a change in the expression level of the one or more genes relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression. In certain embodiments, the change is an increase, and the one or more genes set forth in Table 1 are selected from one or more genes set forth in Table 2. In certain embodiments, the one or more genes set forth in Table 2 comprise one or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise two or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise three or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise four or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise five or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise six or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 comprise the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the one or more genes set forth in Table 2 consist of the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In certain embodiments, the expression level of the one or more genes set forth in Table 2 is increased in the sample relative to the reference expression level, and the method further comprises administering to the individual a therapeutic agent other than, or in addition to a disease modifying anti-rheumatic drug (DMARD).
[0009] In other embodiments, the change is a decrease, and the one or more genes set for in Table 1 are selected from one or more genes set forth in Table 3. In certain embodiments, the expression level of the one or more genes set forth in Table 3 is decreased in the sample relative to the reference expression level, and the method further comprises administering to the individual a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD).
[0010] In certain embodiments, the disease progression is radiographic progression. In certain embodiments, the radiographic progression is characterized by an increase in ShSS values over a defined time period.
[0011] In a second aspect, the invention features a method of treating an individual having RA, the method comprising administering a therapeutic agent other than, or in addition to, a DMARD to the individual, wherein the individual has been identified as one who is more likely to exhibit disease progression by an embodiment of the first aspect.
[0012] In a third aspect, the invention features a method of treating an individual having RA, the method comprising: (a) obtaining a sample from the individual; (b) performing a gene expression assay on the sample and detecting (i) an increased expression level of one or more genes set forth in Table 2 in the sample and/or (ii) a decreased level of one or more genes set forth in Table 3 relative to a reference expression level; (c) identifying the individual as having an increased likelihood of benefitting from a therapeutic agent other than, or in addition to, a DMARD; and (d) administering to the individual a therapeutic agent other than, or in addition to, a DMARD.
[0013] In a fourth aspect, the invention features a method of treating an individual having RA, the individual being identified as having (i) an increased expression level of one or more genes set forth in Table 2 in a sample from the individual and/or (ii) a decreased expression level of one or more genes set forth in Table 3 in a sample from the individual relative to a reference expression level, the method comprising administering to the individual a therapeutic agent other than, or in addition to, a DMARD.

[0014] In a fifth aspect, the invention features a method of treating an individual having RA, the method comprising: (a) determining an expression level of one or more genes set forth in Table 2 or Table 3 in a sample from the individual, wherein (i) the expression level of one or more genes set forth in Table 2 in the sample is determined to be increased and/or (ii) the expression level of the one or more genes set forth in Table 3 is determined to be decreased relative to a reference expression level, and (b) administering to the individual a therapeutic agent other than, or in addition to, a DMARD based on the expression level of the one or more genes set forth in Table 2 or Table 3 determined in step (a).
[0015] In certain embodiments of any one of the third, fourth, and fifth aspects, the one or more genes set forth in Table 2 comprise one or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBAS3A.
[0016] In certain embodiments of any of the preceding aspects, the expression level of the one or more genes set forth in Table 1 is an average of the expression level of the one or more genes set forth in Table 1. In certain embodiments, the average of the expression level of the one or more genes set forth in Table 1 is an average of a normalized expression level of the one or more genes set forth in Table 1.
[0017] In certain embodiments of any of the preceding aspects, the expression level of the one or more genes set forth in Table 1 is a median of the expression level of the one or more genes set forth in Table 1. In certain embodiments, the median of the expression level of the one or more genes set forth in Table 1 is a median of a normalized expression level of the one or more genes set forth in Table 1.
[0018] In certain embodiments of any of the preceding aspects, the normalized expression level of the one or more genes set forth in Table 1 is the expression level of the one or more genes set forth in Table 1 normalized to a reference gene. In certain embodiments, the reference gene is ACTB, GAPDH, GUSB, HPRT1, PGK1, RPL19, TUBB, TMEM55B, or a combination thereof.
[0019] In certain embodiments of any of the preceding aspects, the reference expression level is a pre-assigned expression level of the one or more genes set forth in Table 1. In certain embodiments, the reference expression level is the expression level of the one or more genes set forth in Table 1 in a reference population of individuals having RA who have not been previously treated with a DMARD, the reference population of individuals consisting of a first subset of individuals who exhibited disease progression and a second subset of individuals who did not exhibit disease progression, wherein the reference expression level significantly separates the first and second subsets of individuals based on a significant difference between the expression level of the one or more genes set forth in Table 1 in the first subset of individuals compared to that of the second subset of individuals. In certain embodiments, the first subset of individuals exhibited disease progression and the second subset of individuals did not exhibit disease progression after about 12 months.
[0020] In certain embodiments of the first or fifth aspect, the method further comprises determining one or more clinical covariates of the individual.
[0021] In certain embodiments of the second or fifth aspect, one or more clinical covariates has been determined for the individual.
[0022] In certain embodiments of any of the preceding aspects, the one or more clinical covariates are one or more of: disease activity score 28-erythrocyte sedimentation rate (DAS28-ESR), disease activity score 28-C reactive protein (DAS28-CRP), rheumatoid factor (RF) titer, disease duration, baseline pathotype, and 12 max ultrasound synovial thickening (USST) and ultrasound power Doppler (USPD) scores. In certain embodiments, the clinical covariate is DAS28-ESR. In certain embodiments, the clinical covariate is a RF titer.
[0023] In certain embodiments of any of the preceding aspects, the expression level is a nucleic acid expression level. In certain embodiments, the nucleic acid expression level is an mRNA expression level. In certain embodiments, the mRNA expression level is determined by direct

digital counting of nucleic acids, RNA-seq, RT-qPCR, qPCR, multiplex qPCR or RT-qPCR, microarray analysis, or a combination thereof. In certain embodiments, the direct digital counting of nucleic acids is by NANOSTRING® NCOUNTER® analysis.

[0024] In certain embodiments of any of the preceding aspects, the expression level is a protein expression level. In certain embodiments, the protein expression level is determined by an immunoassay, liquid chromatography-mass spectrometry (LC-MS) technology, nephelometry, aptamer technology, or a combination thereof.

[0025] In a sixth aspect, the invention features a method of identifying an individual having RA who may benefit from a treatment comprising a DMARD, the method comprising determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.

[0026] In a seventh aspect, the invention features a method for selecting a therapy for an individual having RA, the method comprising determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.

[0027] In certain embodiments of the sixth or seventh aspect, the method further comprises determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score that is at or above a reference lymphoid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.

[0028] In certain embodiments of the sixth or seventh aspect, the myeloid eigengene score from a sample is at or above a reference myeloid eigengene score, and the method further comprises administering to the individual a therapeutically effective amount of a DMARD.

[0029] In an eighth aspect, the invention features a method of treating an individual having RA, the method comprising administering a DMARD to the individual, wherein the individual has been identified as one who is more likely to benefit from a treatment comprising a DMARD by an embodiment of the sixth or seventh aspect.

[0030] In a ninth aspect, the invention features a method of treating RA in an individual identified as having a myeloid eigengene score from a sample from the individual that is at or above a reference myeloid eigengene score, the method comprising administering to the individual a DMARD. In certain embodiments, prior to the administering, a lymphoid eigengene score from a sample from the individual has been determined to be at or above a reference lymphoid eigengene score.

[0031] In a tenth aspect, the invention features a method of treating an individual having RA, the method comprising: (a) obtaining a sample form the individual; (b) performing a gene expression assay on the sample and determining a myeloid eigengene score equal to or increased relative to a reference myeloid eigengene score; (c) identifying the individual as having an increased likelihood of benefitting from a DMARD; and (d) administering to the individual a DMARD. In certain embodiments, prior to the identifying the individual as having an increased likelihood of benefitting from a DMARD, the method further comprises performing a gene expression assay on the sample and determining a lymphoid eigengene score equal to or increased relative to a reference lymphoid eigengene score.

[0032] In an eleventh aspect, the invention features a method of treating an individual having RA, the method comprising: (a) determining a myeloid eigengene score from a sample from the individual, wherein the myeloid eigengene score from the sample is determined to be at or above a reference myeloid eigengene score, and (b) administering to the individual a DMARD based on the myeloid eigengene score determined in step (a). In certain embodiments, prior to the administering, the method further comprises determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score in the sample is determined to be at or above a

reference lymphoid eigengene score.
[0033] In certain embodiments of any of the sixth, seventh, eighth, ninth, tenth, and eleventh aspects, the reference myeloid eigengene score is from a reference population of individuals having RA who have been treated with a DMARD therapy, the population of individuals consisting of a first subset of individuals who responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy, wherein the reference myeloid eigengene score significantly separates the first and second subsets of individuals, based on a significant difference between the myeloid eigengene score in the first subset of individuals compared to that of the second subset of individuals. In certain embodiments, the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.
[0034] In certain embodiments of any of the sixth, seventh, eighth, ninth, tenth, and eleventh aspects, the reference lymphoid eigengene score is from a reference population of individuals having RA, the population of individuals consisting of a first subset of individuals who responded to DMARD therapy and a second subset of individuals who did not respond to DMARD therapy, wherein the reference lymphoid eigengene score significantly separates the first and second subsets of individuals, based on a significant difference between the lymphoid eigengene score in the first subset of individuals compared to that of the second subset of individuals.
[0035] In certain embodiments of any of the sixth, seventh, eighth, ninth, tenth, and eleventh aspects, the individual has not been previously treated with a DMARD.
[0036] In certain embodiments of any of the sixth, seventh, eighth, ninth, tenth, and eleventh aspects, the individual has been previously treated with a DMARD.
[0037] In a twelfth aspect, the invention features a method for monitoring the response of an individual having RA to treatment with a DMARD, the method comprising: (a) determining a first myeloid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD, (b) determining a second myeloid eigengene score from a sample from the individual at second time point, and (c) comparing the first myeloid eigengene score with the second myeloid eigengene score, wherein a decrease in the second myeloid eigengene score relative to the first myeloid eigengene score is predictive of an individual who is likely to respond treatment with a DMARD. In certain embodiments, the method further comprises (a) determining a first lymphoid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD, (b) determining a second lymphoid eigengene score from a sample from the individual at second time point, and (c) comparing the first lymphoid eigengene score with the second lymphoid eigengene score, wherein a decrease in the second lymphoid eigengene score relative to the first lymphoid eigengene score is predictive of an individual who is likely to respond treatment with a DMARD. In certain embodiments, the second myeloid eigengene score is decreased relative to the first myeloid eigengene score, and the method further comprises administering an additional dose of a DMARD to the individual. In certain embodiments, the second lymphoid eigengene score is decreased relative to the first lymphoid eigengene score.
[0038] In certain embodiments of the twelfth aspect, the individual has been previously treated with a DMARD.
[0039] In certain embodiments of the twelfth aspect, the decrease is between about 1.25-fold to about 5-fold. In certain embodiments, the decrease is between about 1.25-fold to about 2-fold. In certain embodiments, the decrease is between about 1.25-fold to about 1.5-fold. In certain embodiments, the decrease is at least about 1.25-fold.
[0040] In certain embodiments of any of the preceding aspects, the sample is a synovial sample.
[0041] In certain embodiments, the synovial sample is a synovial tissue sample or a synovial fluid sample.
[0042] In certain embodiments of any of the preceding aspects, the DMARD is methotrexate,

hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, mycophenolate mofetil, or a combination thereof.
[0043] In certain embodiments of any of the first, second, third, fourth, and fifth aspects, the therapeutic agent other than a DMARD is a B cell antagonist, a Janus kinase (JAK) antagonist, a tumor necrosis factor (TNF) antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor antagonist, an IL-6 receptor antagonist, or a combination thereof. In certain embodiments, the JAK antagonist is tofacitinib. In certain embodiments, the IL-6 receptor antagonist is tocilizumab. In certain embodiments, the B cell antagonist is rituximab.

Description

BRIEF DESCRIPTION OF THE DRAWINGS
[0044] The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawing(s) will be provided by the Office upon request and payment of the necessary fee.
[0045] FIG. **1**A is a table showing baseline demographics of the subset of 144 individuals in the PEAC cohort that were enrolled in the study.
[0046] FIG. **1**B is a representative image of an ultrasound (US)-guided wrist biopsy. The inset grayscale transverse US image of the wrist joint demonstrates the biopsy needle entering the joint space under the extensor tendon complex.
[0047] FIG. **1**C is a graph indicating the numbers and types of joints sampled.
[0048] FIG. **2**A is a series of representative images of histological samples acquired from individuals recruited to the Pathobiology of Early Arthritis Cohort (PEAC) showing immune cell infiltration (CD3+, T cells, CD68+ macrophages, CD20+ B cells, and CD138+ plasma cells) and H&E staining per pathotype. All images are at 10× magnification.
[0049] FIG. **2**B is a table showing mean levels of immune cell infiltration (CD3+, T cells, CD68+ macrophages, CD20+ B cells, and CD138+ plasma cells) per pathotype; asterisk denotes significant differences between groups.
[0050] FIG. **2**C is a plot of concordance between the NANOSTRING® and RNA-sequencing gene expression in the PEAC cohort data. The overall Spearman rho=0.85 between the two measurements. Individual patients are colored based on gene pathotype assignments (red=lymphoid, purple=myeloid, green=pauciimmune-fibroid, black=additional biology).
[0051] FIG. **2**D is a series of plots showing lymphoid (left panel), myeloid (center panel), and pauciimmune-fibroid (right panel) eigengene scores against synovial aggregation scores (G1, G2, and G3=grade 1, 2, and 3 aggregates, respectively). Asterisks denote statistical significance as determined by linear regression across groups: *=$p<0.05$; **=$p<0.01$; ***=$p<0.001$; and n.s.=not significant.
[0052] FIG. **3**A is a heatmap of NANOSTRING® data. Raw log 2 NANOSTRING® counts for 212 genes and 111 samples from individuals were normalized per probe to give a mean of 0, and standard deviation of 1. Normalized data were clustered by row and column using Euclidean distance and Ward's linkage. Samples are colored according to IHC-determined pathotype, with ungraded samples colored grey. Rows are colored according to the pathotype with which the gene was originally associated, with RA biology-associated genes colored black.
[0053] FIG. **3**B is a series of plots showing eigengene scores versus IHC-determined pathotypes. Individual eigengene scores are plotted for each sample, grouped and colored by the pathotype as determined by IHC. Asterisks represent statistical significance as determined by linear regression across groups: *=$p<0.05$; **=$p<0.01$; and ***=$p<0.001$.
[0054] FIG. **3**C is a radar plot of standardized eigengene scores. Eigengene values were normalized to give a mean of 0 and standard deviation of 1. Samples were grouped by pathotype, and the mean

(solid lines) and standard error of the mean (shaded region) was calculated for the normalized eigengenes. Spokes of the radar plot represent the distance along each normalized eigengene for each sample group.

[0055] FIG. **3**D is a volcano plot of gene expression differences across pathotypes. For each gene, a 1-way ANOVA was performed comparing expression across the three pathotypes. The −log.sub.10 p-value from the 1-way ANOVA is plotted against the root mean square of the log.sub.2-fold changes between each pair of eigengenes. Genes are colored according to the pathotype in which it was initially identified, with RA biology-associated genes colored black.

[0056] FIG. **4**A is a series of plots comparing eigengene scores. Eigengene values for baseline samples are plotted against each other, with samples colored according to IHC-determined pathotype.

[0057] FIG. **4**B is a series of volcano plots showing comparisons between the three pathotypes. Each pathotype was compared to the other two using linear regression. The log.sub.2-fold change and −log.sub.10 p-values for these comparisons are shown, with genes colored according to the pathotype to which they were initially assigned and RA biology-associated genes colored black. Genes that were significant at a Benjamini-Hochberg adjusted p-value<0.01 are shown as filled dots, while those not meeting this cutoff are shown as open circles.

[0058] FIG. **5**A is a table showing 12-month radiographic outcome of individuals stratified according to pauciimmune-fibroid/myeloid versus lymphoid pathotypes.

[0059] FIG. **5**B is a volcano plot of pre-treatment genes differentially expressed in individuals who progress radiographically after one year (ShSS≥1). P-values were from the two-sample t-test comparing the progressors and non-progressors without adjustment. 46 genes with a p-value<0.05 are highlighted in red and located above the dashed line.

[0060] FIG. **5**C is a series of plots showing lymphoid (left panel), myeloid (center panel), and pauciimmune-fibroid (right panel) baseline scores against radiographic progression status (ΔShSS≥1) at one year. **=p<0.01 by t-test.

[0061] FIG. **5**D is a gene set enrichment plot showing that osteoclast-specific genes are more highly expressed in the lymphoid pathotype versus fibroid/myeloid pathotypes. The x-axis shows the log 2 fold change between lymphoid and fibroid/myeloid samples. The density shown in grey shows the distribution of fold changes across all genes assayed; osteoclast-specific genes are shown as black points, with the overall distribution of fold changes shown as a black line.

[0062] FIG. **5**E is a ROC graph showing the identification of clinical and gene expression features predictive of radiographic progression at one year. The 46 genes as listed in Table 1 and selected covariates (eight clinical covariates: baseline RF titer, disease duration, VAS, swollen joint number, DAS28-ESR, baseline pathotype, and 12 max ultrasound synovial thickening (USST) and ultrasound power Doppler (USPD) scores) were entered simultaneously into a logistic model with an L1 regularization penalty (LASSO) in order to determine the optimal sparse prediction model. The model exhibited improved predictive performance with penalized clinical covariates (blue dashed line) compared to without penalizing clinical covariates (red dotted line), and both these models outperform just the clinical measures alone (black line).

[0063] FIG. **6**A is a lambda training curve from the final glmnet fitted model. The red dots represent mean binomial deviance using 10-fold cross-validation. The error bars represent standard error of binomial deviance. The vertical dotted lines indicate minimum binomial deviance (λ.sub.min) and a more regularized model for which the binomial deviance error is within one standard error of the minimum binomial deviance (λ.sub.1se). λ.sub.min was selected, corresponding to nine non-zero coefficients in the final model.

[0064] FIG. **6**B is a table showing the non-zero coefficients associated with the final variables selected by the LASSO regression.

[0065] FIG. **7**A is a table showing baseline clinical and histological parameters stratified according to the three pathological subtypes (n=129). Asterisks indicates significant differences.

[0066] FIG. **7**B is a chart showing the correlation analysis of each eigengene score with metrics of clinical disease activity, autoantibodies, acute phase reactants, and ultrasonography. Values represent Spearman correlation coefficients between the clinical variables and the individual eigengene scores. Asterisks represent the significance of the correlation coefficient: *=p<0.05; **=p<0.01; and ***=p<0.001.
[0067] FIG. **8**A is a table showing clinical changes in disease activity per pathotype (lymphoid, myeloid, and pauciimmune-fibroid) and treatment regimens according to pathotype.
[0068] FIG. **8**B is a volcano plot showing changes in gene expression between baseline and six months in individuals with a EULAR response. Individual points are colored by the pathotype in which the gene was originally identified, with RA biology-associated genes colored black.
[0069] FIG. **8**C is a volcano plot showing changes in gene expression between baseline and six months in individuals with a EULAR non-response. Individual points are colored by the pathotype in which the gene was originally identified, with RA biology-associated genes colored black.
[0070] FIG. **8**D is a series of plots showing correlation of pre-treatment lymphoid (left panel), myeloid (center panel), and pauciimmune-fibroid (right panel) eigengene scores with change in DAS28-ESR after six months of DMARD treatment. Spearman's correlation coefficient is shown, along with the significance of this value.
[0071] FIG. **8**E is a series of paired plots for baseline and six-month lymphoid (left panels), myeloid (center panels), and pauciimmune-fibroid (right panels) eigengene scores in individuals who achieved good or poor clinical responses to DMARD treatment at six months by the EULAR response criteria. Individuals who achieved a good response, or failed to achieve a moderate response, according to EULAR criteria are shown. For each individual, the pre-treatment eigengene scores are connected to the post-treatment eigengene score, for each of the three eigengenes. Asterisks represent significance of the difference between pre- and post-treatment samples using a linear mixed effects model with sample date as a fixed effect and individual as a random effect: *=p<0.05; **=p<0.01; and ***=p<0.001.
[0072] FIG. **9**A is a chart showing the correlation of pre-treatment serum CXCL13, sICAM1, MMP3, and IL-8 with clinical disease metrics and ultrasonography scores. Values represent Spearman correlation coefficients between the clinical variables and the individual eigengene scores. Asterisks represent the significance of the correlation coefficient: *=p<0.05; **=p<0.01; and ***=p<0.001.
[0073] FIG. **9**B is a chart showing the correlation of pre-treatment serum CXCL13, sICAM1, MMP3, and IL-8 with synovial histology scores. Values represent Spearman correlation coefficients between the clinical variables and the individual eigengene scores. Asterisks represent the significance of the correlation coefficient: *=p<0.05; **=p<0.01; and ***=p<0.001.
[0074] FIG. **9**C is a plot of the concentration of serum CXCL13 versus synovial pathotype status, showing elevated levels in lymphoid pathotypes versus myeloid or pauciimmune-fibroid. P-values were calculated using a t-test.
[0075] FIG. **9**D is a plot of the concentration of serum MMP3 versus synovial pathotype status, showing elevated levels in lymphoid pathotypes versus myeloid or pauciimmune-fibroid. P-values were calculated using a t-test.

DETAILED DESCRIPTION

I. Definitions

[0076] It is to be understood that aspects and embodiments of the invention described herein include “comprising,” “consisting,” and “consisting essentially of” aspects and embodiments. As used herein, the singular form “a,” “an,” and “the” includes plural references unless indicated otherwise.
[0077] The term “about” as used herein refers to the usual error range for the respective value readily known to the skilled person in this technical field. Reference to “about” a value or parameter herein includes (and describes) embodiments that are directed to that value or parameter

per se. For example, description referring to “about X” includes description of “X.”
[0078] The term “pathotype” as used herein refers to a subtype of RA characterized by certain pathological, histological, and/or clinical features of RA. The pathological, histological, and/or clinical features of RA may be further associated with the expression of one or more particular genes or one or more particular proteins, or a particular pattern of expression of a combination of genes or a combination of proteins. Such pathotypes include, but are not limited to, the lymphoid pathotype (e.g., characterized by B cell-rich aggregates), myeloid pathotype (e.g., characterized by a predominant macrophage infiltrate), and pauciimmune-fibroid pathotype (e.g., characterized by and few infiltrating immune cells, but still expansion of fibroblast lineage cells in the sublining and lining layers). In some embodiments, the pathotype is a lymphoid pathotype and is further characterized by the expression of one or more genes set forth in Table 9. In some embodiments, the pathotype is a myeloid pathotype and is further characterized by the expression of one or more genes set forth in Table 10. In some embodiments, the pathotype is a pauciimmune-fibroid pathotype and is further characterized by the expression of one or more genes set forth in Table 11.
[0079] The term “biomarker” as used herein refers to an indicator, e.g., a predictive and/or prognostic indicator, which can be detected in a sample (e.g., a gene) or derived from one or more indicators detected from a sample (e.g., an eigengene score). The biomarker may serve as an indicator of a particular subtype of a disease or disorder (e.g., rheumatoid arthritis) characterized by certain molecular, pathological, histological, and/or clinical features. In some embodiments, a biomarker is a gene. In other embodiments, a biomarker is a collection of genes. Biomarkers include, but are not limited to, polynucleotides (e.g., DNA, and/or RNA), polynucleotide copy number alterations (e.g., DNA copy numbers), polypeptides, polypeptide and polynucleotide modifications (e.g. posttranslational modifications), carbohydrates, and/or glycolipid-based molecular markers.
[0080] Such biomarkers include, but are not limited to, CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, UBASH3A, BLK, BTK, BTLA, CCL19, CD19, CD38, CD40LG, CILP, DENND2D, DKK3, FCRL5, FGF2, FGF9, HLA-DOB, ICOS, JCHAIN, IL36B, IRF4, LOC100505746, LY9, MAP4K1, MMP1, NOG, PIM2, POU2AF1, RHOH, SEL1L3, SIRPG, SLAMF6, SLC31A1, SPIB, TIGIT, TLR10, TMC6, TNF, TNFRSF11B, TNFRSF17, TRAF3PIP3, and XBP1, or a combination thereof. In some embodiments, the biomarker is one or more biomarkers selected from the group consisting of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In other embodiments, the biomarker is one or more biomarkers selected from the group consisting of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, UBASH3A, BLK, BTK, BTLA, CCL19, CD19, CD38, CD40LG, DENND2D, FCRL5, HLA-DOB, ICOS, JCHAIN, IL36B, IRF4, LOC100505746, LY9, MAP4K1, MMP1, PIM2, POU2AF1, RHOH, SEL1L3, SIRPG, SLAMF6, SLC31A1, SPIB, TIGIT, TLR10, TMC6, TNF, TNFRSF17, TRAF3PIP3, and XBP1. In yet other embodiments, the biomarker is one or more biomarkers selected from the group consisting of CILP, DKK3, FGF2, FGF9, NOG, and TNFRSF11B. Expression of such a biomarker may be determined to be higher or lower in a sample obtained from a patient sensitive or responsive to a treatment (e.g., treatment with a rheumatoid arthritis therapy that includes a DMARD therapy) than a reference level including, e.g., the median expression level of the biomarker in a sample from a group/population of patients, e.g., patients having RA, and being tested for responsiveness to a treatment; the median expression level of the biomarker in a sample from a group/population of patients, e.g., patients having RA, and identified as not responding to a treatment; the level in a sample previously obtained from the individual at a prior time; or the level in a sample from a patient who received prior treatment (e.g., treatment with a DMARD).
[0081] The term “CD180” as used herein, refers to any native CD180 antigen from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed CD180 as well as any form

of CD180 that results from processing in the cell. The term also encompasses naturally occurring variants of CD180, e.g., splice variants or allelic variants. CD180 is also referred to in the art as lymphocyte antigen 64 (LY64), radioprotective 105 kDa protein (RP105), and Ly78. The nucleic acid sequence of an exemplary human CD180 is shown under NCBI Reference Sequence: NM_005582.2 or in SEQ ID NO: 1. The amino acid sequence of an exemplary protein encoded by human CD180 is shown under UniProt Accession No. Q99467 or in SEQ ID NO: 2.

[0082] The term “CSF2” as used herein, refers to any native granulocyte-macrophage colony-stimulating factor from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed CSF2 as well as any form of CSF2 that results from processing in the cell. The term also encompasses naturally occurring variants of CSF2, e.g., splice variants or allelic variants. CSF2 is also referred to in the art as GM-CSF, colony-stimulating factor, CSF, molgramostin, and sargramostim. The nucleic acid sequence of an exemplary human CSF2 is shown under NCBI Reference Sequence: NM_000758.3 or in SEQ ID NO: 3. The amino acid sequence of an exemplary protein encoded by human CSF2 is shown under UniProt Accession No. P04141 or in SEQ ID NO: 4.

[0083] The term “CXCL1” as used herein, refers to any native C—X—C motif chemokine 1 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed CXCL1 as well as any form of CXCL1 that results from processing in the cell. The term also encompasses naturally occurring variants of CXCL1, e.g., splice variants or allelic variants. CXCL1 is also referred to in the art as chemokine ligand 1, growth-regulated alpha protein, GRO-alpha (1-73), melanoma growth stimulatory activity, MGSA, neutrophil-activating protein 3, and NAP-3. The nucleic acid sequence of an exemplary human CXCL1 is shown under NCBI Reference Sequence: NM_001511.3 or in SEQ ID NO: 5. The amino acid sequence of an exemplary protein encoded by human CXCL1 is shown under UniProt Accession No. P09341 or in SEQ ID NO: 6.

[0084] The term “DENND1C” as used herein, refers to any native DENN domain-containing protein 1C from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed DENND1C as well as any form of DENND1C that results from processing in the cell. The term also encompasses naturally occurring variants of DENND1C, e.g., splice variants or allelic variants. DENND1C is also referred to in the art as connecdenn 3 and protein FAM31C. The nucleic acid sequence of an exemplary human DENND1C is shown under NCBI Reference Sequence: NM_001290331.1 or in SEQ ID NO: 7. The amino acid sequence of an exemplary protein encoded by human DENND1C is shown under UniProt Accession No. Q81V53 or in SEQ ID NO: 8.

[0085] The term “MMP10” as used herein, refers to any native matrix metallopeptidase 10 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed MMP10 as well as any form of MMP10 that results from processing in the cell. The term also encompasses naturally occurring variants of MMP10, e.g., splice variants or allelic variants. MMP10 is also referred to in the art as stromelysin-2, SL-2, and transin-2. The nucleic acid sequence of an exemplary human MMP10 is shown under NCBI Reference Sequence: NM_002425.2 or in SEQ ID NO: 9. The amino acid sequence of an exemplary protein encoded by human MMP10 is shown under UniProt Accession No. P09238 or in SEQ ID NO: 10.

[0086] The term “SDC1” as used herein, refers to any native syndecan 1 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed SDC1 as well as any form of SDC1 results from processing in the cell. The term also encompasses naturally occurring variants of SDC1, e.g., splice variants or allelic variants. SDC1 is also referred to in the art as

SYND1 and CD138. The nucleic acid sequence of an exemplary human SDC1 is shown under NCBI Reference Sequence: NM_001006946.1 or in SEQ ID NO: 11. The amino acid sequence of an exemplary protein encoded by human SDC1 is shown under UniProt Accession No. P18827 or in SEQ ID NO: 12.

[0087] The term “UBASH3A” as used herein, refers to any native ubiquitin associated and SH3 domain containing protein A from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed UBASH3A as any form of UBASH3A from processing in the cell. The term also encompasses naturally occurring variants of UBASH3A, e.g., splice variants or allelic variants. UBASH3A is also referred to in the art as Cbl-interacting protein 4, CLIP4, suppressor of T cell receptor signaling 2, STS-2, T cell ubiquitin ligand 1, and TULA-1. The nucleic acid sequence of an exemplary human UBASH3A is shown under NCBI Reference Sequence: NM_001001895.2 or in SEQ ID NO: 13. The amino acid sequence of an exemplary protein encoded by human UBASH3A is shown under UniProt Accession No. P57075 or in SEQ ID NO: 14.

[0088] As used herein, the terms “myeloid eigengene score” and “myeloid eigengene,” each of which may be used interchangeably, refer to a numerical value that reflects the degree or amount of pathological, histological, and/or clinical features of an RA of the myeloid pathotype, and which is correlated with the expression level of one or more genes set forth in Table 10 detected in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) obtained from an individual (e.g., an individual having RA). The myeloid eigengene score may serve as a biomarker (e.g., a predictive and/or prognostic biomaker) of RA of the myeloid pathotype.

[0089] As used herein, the terms “lymphoid eigengene score” and “lymphoid eigengene” each of which may be used interchangeably, refer to a numerical value that reflects the degree or amount of pathological, histological, and/or clinical features of an RA of the lymphoid phenotype, and which is correlated with the expression level of one or more genes set forth in Table 9 detected in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) obtained from an individual (e.g., an individual having RA). The lymphoid eigengene score may serve as a biomarker (e.g., a predictive and/or prognostic biomarker) of RA of the lymphoid phenotype.

[0090] As used herein, the terms “pauciimmune-fibroid eigengene score” and “pauciimmune-fibroid eigengene” each of which may be used interchangeably, refer to a numerical value that reflects the degree or amount of pathological, histological, and/or clinical features of an RA of the pauciimmune-fibroid pathotype, and which is correlated with the expression level of one or more genes set forth in Table 11 detected in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) obtained from an individual (e.g., an individual having RA). The pauciimmune-fibroid eigengene score may serve as a biomarker (e.g., a predictive and/or prognostic biomaker) of RA of the pauciimmune-fibroid pathotype.

[0091] The term “detecting” is used herein in the broadest sense to include both qualitative and quantitative measurements of a target molecule. Detecting includes identifying the mere presence of the target molecule in a sample as well as determining whether the target molecule is present in the sample at detectable levels. Detecting may be direct or indirect.

[0092] The terms “level of expression” or “expression level” in general are used interchangeably and generally refer to the amount of a biomarker in a biological sample. “Expression” generally refers to the process by which information (e.g., gene-encoded and/or epigenetic information) is converted into the structures present and operating in the cell. Therefore, as used herein, “expression” may refer to transcription into a polynucleotide, translation into a polypeptide, or even polynucleotide and/or polypeptide modifications (e.g., posttranslational modification of a polypeptide). Fragments of the transcribed polynucleotide, the translated polypeptide, or polynucleotide and/or polypeptide modifications (e.g., posttranslational modification of a polypeptide) shall also be regarded as expressed whether they originate from a transcript generated

by alternative splicing or a degraded transcript, or from a posttranslational processing of the polypeptide, e.g., by proteolysis. “Expressed genes” include those that are transcribed into a polynucleotide as mRNA and then translated into a polypeptide, and also those that are transcribed into RNA but not translated into a polypeptide (for example, transfer and ribosomal RNAs). An expression level for more than one gene of interest may be determined by aggregation methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the median or mean of all the expression levels of the genes of interest. Before aggregation, the expression level of each gene of interest may be normalized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, normalized to the expression level of one or more housekeeping genes, or normalized to a total library size, or normalized to the median or mean expression level value across all genes measured. In some instances, before aggregation across multiple genes of interest, the normalized expression level of each gene of interest may be standardized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the Z-score of the normalized expression level of each gene of interest.

[0093] The term “sample,” as used herein, refers to a composition that is obtained or derived from a patient and/or individual of interest that contains a cellular and/or other molecular entity that is to be characterized and/or identified, for example, based on physical, biochemical, chemical, and/or physiological characteristics. Samples include, but are not limited to, synovial tissue samples, synovial fluid samples, tissue samples, primary or cultured cells or cell lines, cell supernatants, cell lysates, platelets, serum, plasma, vitreous fluid, lymph fluid, follicular fluid, seminal fluid, amniotic fluid, milk, whole blood, blood-derived cells, urine, cerebro-spinal fluid, saliva, sputum, tears, perspiration, mucus, tumor lysates, and tissue culture medium, tissue extracts such as homogenized tissue, tumor tissue, cellular extracts, and combinations thereof.

[0094] A sample or cell that “expresses” a protein of interest is one in which mRNA encoding the protein, or the protein, including fragments thereof, is determined to be present in the sample or cell.

[0095] As used herein, the terms “reference expression level” and “reference level” are used interchangeably to refer to an expression level against which another expression level, e.g., the expression level of one or more genes described herein (e.g., any gene or combination of genes set forth in any one of Tables 1-3) in a sample from an individual is compared, e.g., to make a diagnostic (e.g., predictive and/or prognostic) and/or therapeutic determination. For example, the reference expression level may be derived from expression levels in a reference population (e.g., the median expression level in a reference population, e.g., a population of patients having RA who have not been treated with an RA therapy (e.g., a DMARD (e.g., methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, or mycophenolate mofetil)), a reference sample, and/or a pre-assigned value (e.g., a cut-off value which was previously determined to significantly (e.g., statistically significantly)) separate a first subset of individuals who exhibited disease progression and a second subset of individuals who did not exhibit disease progression, wherein the reference expression level significantly separates the first and second subsets of individuals based on a significant difference between the expression level in the first subset of individuals compared to that of the second subset of individuals. In some embodiments, the cut-off value may be the median or mean expression level in the reference population. In other embodiments, the reference level may be the top 40%, the top 30%, the top 20%, the top 10%, the top 5%, or the top 1% of the expression level in the reference population. In particular embodiments, the cut-off value may be the median expression level in the reference population. It will be appreciated by one skilled in the art that the numerical value for the reference expression level may vary depending on the indication or disorder (e.g., RA), the methodology used to detect expression levels (e.g., RNA-seq, microarray analysis, or RT-qPCR), and/or the specific combinations of genes examined (e.g., any combination of the genes set forth in Tables 1-

3).
[0096] As used herein, the terms “reference myeloid eigengene score” and “reference myeloid eigengene” are used interchangeably to refer to a myeloid eigengene score against which another myeloid eigengene score is compared, e.g., to make a diagnostic (e.g., predictive and/or prognostic) and/or therapeutic determination. For example, the reference myeloid eigengene score may be a myeloid eigengene score in a reference sample, a reference population, and/or a pre-determined value. In some instances, the reference myeloid eigengene score is a cut-off value that significantly separates a first subset of individuals who have been treated with an RA therapy (e.g., a DMARD therapy, e.g., therapy including methotrexate) in a reference population and a second subset of individuals who been treated with an RA therapy (e.g., a DMARD therapy, e.g., therapy including methotrexate) in the same reference population based on a significant difference between an individual's responsiveness to treatment with the RA therapy, wherein the cut-off value significantly separates the first subset of individuals who responded to the RA therapy from the second subset of individuals who did not respond to the RA therapy. In some instances, the cut-off value may be the median or mean myeloid eigengene score in the reference population. In other instances, the reference myeloid eigengene score may be the top 40%, the top 30%, the top 20%, the top 10%, the top 5%, or the top 1% of the myeloid eigengene score value in the reference population. It will be appreciated by one skilled in the art that the value for the reference myeloid eigengene score may vary depending on the methodology used to detect expression levels of genes comprising the score (e.g., direct digital counting of nucleic acids (e.g., NANOSTRING® NCOUNTER®), RNA-seq, microarray analysis, or RT-qPCR) and/or the specific combinations of genes examined to derive the reference myeloid eigengene score (e.g., any combination of the genes set forth in Table 10).
[0097] As used herein, the terms “reference lymphoid eigengene score” and “reference lymphoid eigengene” are used interchangeably to refer to a lymphoid eigengene score against which another lymphoid eigengene score is compared, e.g., to make a diagnostic (e.g., predictive and/or prognostic) and/or therapeutic determination. For example, the reference lymphoid eigengene score may be a lymphoid eigengene score in a reference sample, a reference population, and/or a pre-determined value. In some instances, the reference lymphoid eigengene score is a cut-off value that significantly separates a first subset of individuals who have been treated with an RA therapy (e.g., a DMARD therapy, e.g., therapy including methotrexate) in a reference population and a second subset of individuals who been treated with an RA therapy (e.g., a DMARD therapy, e.g., therapy including methotrexate) in the same reference population based on a significant difference between an individual's responsiveness to treatment with the RA therapy, wherein the cut-off value significantly separates the first subset of individuals who responded to the RA therapy from the second subset of individuals who did not respond to the RA therapy. In some instances, the cut-off value may be the median or mean lymphoid eigengene score in the reference population. In other instances, the reference lymphoid eigengene score may be the top 40%, the top 30%, the top 20%, the top 10%, the top 5%, or the top 1% of the lymphoid eigengene score value in the reference population. It will be appreciated by one skilled in the art that the value for the reference lymphoid eigengene score may vary depending on the methodology used to detect expression levels of genes comprising the score (e.g., direct digital counting of nucleic acids (e.g., NANOSTRING® NCOUNTER®), RNA-seq, microarray analysis, or RT-qPCR), and/or the specific combinations of genes examined to derive the reference lymphoid eigengene score (e.g., any combination of the genes set forth in Table 9).
[0098] Expression “above” a level (e.g., above a reference level), “increased,” “increased expression,” “increased expression level,” “increased levels,” “elevated,” “elevated expression,” “elevated expression levels,” or “elevated levels” refers to an increased expression, increased levels of a biomarker or an increased eigengene score in an individual relative to the expression level of the biomarker or eigengene score in a control (e.g., an individual or individuals who are not

suffering from the disease or disorder (e.g., RA), an internal control (e.g., a housekeeping biomarker), the level of a biomarker or eigengene score in a sample obtained from the individual prior to administration of a therapy (e.g., a DMARD)), or relative to a reference level (e.g., the median expression level of the biomarker or eigengene score in samples from a group/population of patients, e.g., patients having RA who are being tested for responsiveness to an RA therapy that includes a DMARD; the median expression level of the biomarker or eigengene score in samples from a group/population of patients, e.g., patients having RA who have been identified as not responding to a DMARD; or the level in a sample previously obtained from the individual at a prior time).

[0099] Expression "below" a level (e.g., below a reference level), "decreased," "decreased expression," "decreased expression level," "decreased levels," "reduced," "reduced expression," "reduced expression levels," or "reduced levels" refers to decreased expression, decreased levels, of a biomarker or a decreased eigengene score in an individual relative to the expression level of the biomarker or eigengene score in a control (e.g., an individual or individuals who are not suffering from the disease or disorder (e.g., RA), an internal control (e.g., a housekeeping biomarker), or the level of a biomarker or eigengene score in a sample obtained prior to administration of a therapy (e.g., a DMARD), or relative to a reference level (e.g., the median expression level of the biomarker or eigengene score in samples from a group/population of patients, e.g., patients having RA who are being tested for responsiveness to an RA therapy that includes a DMARD (e.g., methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, or mycophenolate mofetil); the median expression level of the biomarker or eigengene score in samples from a group/population of patients, e.g., patients having RA who have been identified as not responding to an RA therapy that includes a DMARD; or the level in a sample previously obtained from the individual at a prior time). In some embodiments, reduced expression is little or no expression.

[0100] A "reference sample," "reference cell," "reference tissue," "control sample," "control cell," or "control tissue," as used herein, refers to a sample, cell, tissue, or standard that is used for comparison purposes. In one embodiment, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is obtained from a healthy and/or non-diseased part of the body (e.g., tissue or cells) of the same patient or individual. For example, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue may be healthy and/or non-diseased cells or tissue adjacent to the diseased cells or tissue (e.g., cells or tissue adjacent to an affected joint). In another embodiment, a reference sample is obtained from an untreated tissue and/or cell of the body of the same patient or individual. In yet another embodiment, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is obtained from a healthy and/or non-diseased part of the body (e.g., tissues or cells) of an individual who is not the patient or individual. In even another embodiment, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is obtained from an untreated tissue and/or cell of the body of an individual who is not the patient or individual. In another embodiment, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is obtained from a patient prior to administration of a therapy (e.g., an RA therapy that includes a DMARD (e.g., methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, or mycophenolate mofetil) and/or a biologic therapeutic agent (e.g., a B cell antagonist (e.g., rituximab), a Janus kinase (JAK) antagonist (e.g., tofacitinib), a tumor necrosis factor (TNF) antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor antagonist, or an IL-6 receptor antagonist (tocilizumab)).

[0101] The term "housekeeping biomarker" refers to a biomarker or group of biomarkers (e.g., polynucleotides and/or polypeptides) which are typically similarly present in all cell types. In some embodiments, the housekeeping biomarker is a "housekeeping gene." A "housekeeping gene" refers herein to a gene or group of genes which encode proteins whose activities are essential for

the maintenance of cell function and which are typically similarly present in all cell types. Exemplary housekeeping genes may include, but are not limited to, ACTB, GAPDH, GUSB, HPRT1, PGK1, RPL19, TUBB, and TMEM55B.

[0102] The term “ACTB” as used herein, refers to any native beta-actin from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed ACTB as well as any form of ACTB that results from processing in the cell. The term also encompasses naturally occurring variants of ACTB, e.g., splice variants of ACTB, e.g., splice variants or allelic variants. ACTB is also referred to in the art as beta (p)-actin, actin beta, PS1TP5-binding protein 1, beta cytoskeletal actin, PS1TP5BP1, BRWS1, and actin, cytoplasmic 1. The nucleic acid sequence of an exemplary human ACTB is shown under NCBI Reference Sequence: NM_002046.6 or in SEQ ID NO: 15. The amino acid sequence of the exemplary protein encoded by human ACTB is shown under UniProt Accession No. P60709-1 or in SEQ ID NO: 16. The term “GAPDH” as used herein, refers to any native glyceraldehyde-3-phosphate dehydrogenase from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed GAPDH as well as any form of GAPDH that results from processing in the cell. The term also encompasses naturally occurring variants of GAPDH, e.g., splice variants of GAPDH, e.g., splice variants or allelic variants. GAPDH is also referred to in the art as GAPD, EC 1.2.1.12, Epididymis Secretory Sperm Binding Protein Li 162eP, Aging-Associated Gene 9 Protein, HEL-S-162eP, HEL-S-162eP, EC 1.2.1, G3PD, G3PDH, and peptidyl-cysteine S-nitrosylase GAPDH. The nucleic acid sequence of an exemplary human GAPDH is shown under NCBI Reference Sequence: NM_002046.6 or in SEQ ID NO: 17. The amino acid sequence of the exemplary protein encoded by human GAPDH is shown under UniProt Accession No. P04406 or in SEQ ID NO: 18.

[0103] The term “GUSB” as used herein, refers to any native glucuronidase beta from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed GUSB as well as any form of GUSB that results from processing in the cell. The term also encompasses naturally occurring variants of GUSB, e.g., splice variants of GUSB, e.g., splice variants or allelic variants. GUSB is also referred to in the art as EC 3.2.1.31, beta-G1, beta-D-glucuronidase, beta-glucuronidase, MPS7, BG, and glucuronidase, beta. The nucleic acid sequence of an exemplary human GUSB is shown under NCBI Reference Sequence: NM_000181.3 or in SEQ ID NO: 19. The amino acid sequence of the exemplary protein encoded by human GUSB is shown under UniProt Accession No. P08236 or in SEQ ID NO: 20.

[0104] The term “HPRT1” as used herein, refers to any native hypoxanthine phosphoribosyltransferase 1 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed HPRT1 as well as any form of HPRT1 that results from processing in the cell. The term also encompasses naturally occurring variants of HPRT1, e.g., splice variants of HPRT1, e.g., splice variants or allelic variants. HPRT1 is also referred to in the art as EC 2.4.2.8, HGPRTase, HGPRT, HPRT, hypoxanthine-guanine phosphoribosyltransferase 1, hypoxanthine-guanine phosphoribosyltransferase, and testicular tissue Protein Li 89. The nucleic acid sequence of an exemplary human HPRT1 is shown under NCBI Reference Sequence: NM_000194.2 or in SEQ ID NO: 21. The amino acid sequence of the exemplary protein encoded by human HPRT1 is shown under UniProt Accession No. P00492 or in SEQ ID NO: 22.

[0105] The term “PGK1” as used herein, refers to any native phosphoglycerate kinase 1 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed PGK1 as well as any form of PGK1 that results from processing in the cell. The term also encompasses naturally occurring variants of PGK1, e.g., splice variants of PGK1, e.g., splice variants or allelic variants.

PGK1 is also referred to in the art as cell migration-inducing gene 10 protein, primer recognition protein 2, EC 2.7.2.3, PRP 2, PGKA, epididymis secretory sperm binding protein Li 68p, HEL-S-68p, and MIG10. The nucleic acid sequence of an exemplary human PGK1 is shown under NCBI Reference Sequence: NM_000291.3 or in SEQ ID NO: 23. The amino acid sequence of the exemplary protein encoded by human PGK1 is shown under UniProt Accession No. P00558 or in SEQ ID NO: 24.

[0106] The term “RPL19” as used herein, refers to any native ribosomal protein L19 from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed RPL19 as well as any form of RPL19 that results from processing in the cell. The term also encompasses naturally occurring variants of RPL19, e.g., splice variants of RPL19, e.g., splice variants or allelic variants. RPL19 is also referred to in the art as large ribosomal subunit protein EL19, large ribosomal subunit protein EL19, ribosomal protein L19, cytosolic, N-terminus truncated, and L19. The nucleic acid sequence of an exemplary human RPL19 is shown under NCBI Reference Sequence: NM_000981.3 or in SEQ ID NO: 25. The amino acid sequence of the exemplary protein encoded by human RPL19 is shown under UniProt Accession No. P84098 or in SEQ ID NO: 26.

[0107] The term “TUBB” as used herein, refers to any native tubulin beta class I from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed TUBB as well as any form of TUBB that results from processing in the cell. The term also encompasses naturally occurring variants of TUBB, e.g., splice variants of TUBB, e.g., splice variants or allelic variants. TUBB is also referred to in the art as tubulin, beta polypeptide, tubulin beta-5 chain, TUBB5, class I beta-tubulin, tubulin beta-1 chain, tubulin beta chain, beta Ib tubulin, beta1-tubulin, tubulin, beta, OK/SW-CI.56, CDCBM6, CSCSC1, TUBB1, and M40. The nucleic acid sequence of an exemplary human TUBB is shown under NCBI Reference Sequence: NM_001293212.1 or in SEQ ID NO: 27. The amino acid sequence of the exemplary protein encoded by human TUBB is shown under UniProt Accession No. P07437 or in SEQ ID NO: 28.

[0108] The term “TMEM55B” as used herein, refers to any native transmembrane protein 55B from any vertebrate source, including mammals such as primates (e.g., humans) and rodents (e.g., mice and rats), unless otherwise indicated. The term encompasses “full-length,” unprocessed TMEM55B as well as any form of TMEM55B that results from processing in the cell. The term also encompasses naturally occurring variants of TMEM55B, e.g., splice variants of TMEM55B, e.g., splice variants or allelic variants. TMEM55B is also referred to in the art as phosphatidylinositol-4,5-bisphosphate 4-phosphatase 1, PIP4P1, type I phosphatidylinositol-4,5-bisphosphate 4-phosphatase, type I Ptdlns-4,5-P(2) 4-phosphatase, type 1 Ptdlns-4,5-P2 4-Ptase, Ptdlns-4,5-P2 4-Ptase I, EC 3.1.3.78, C14orf9, type 1 phosphatidylinositol 4,5-bisphosphate 4-phosphatase, Ptdlns-4,5-P(2) 4-phosphatase type I, and chromosome 14 open reading frame 9. The nucleic acid sequence of an exemplary human TMEM55B is shown under NCBI Reference Sequence: NM_001100814.2 or in SEQ ID NO: 29. The amino acid sequence of the exemplary protein encoded by human TMEM55B is shown under UniProt Accession No. Q86T03 or in SEQ ID NO: 30.

[0109] By “correlate” or “correlating” is meant comparing, in any way, the performance and/or results of a first analysis or protocol with the performance and/or results of a second analysis or protocol. For example, one may use the results of a first analysis or protocol in carrying out a second protocols and/or one may use the results of a first analysis or protocol to determine whether a second analysis or protocol should be performed. With respect to the embodiment of polypeptide analysis or protocol, one may use the results of the polypeptide expression analysis or protocol to determine whether a specific therapeutic regimen should be performed. With respect to the embodiment of polynucleotide analysis or protocol, one may use the results of the polynucleotide expression analysis or protocol to determine whether a specific therapeutic regimen should be

performed.
[0110] “Amplification,” as used herein, generally refers to the process of producing multiple copies of a desired sequence. “Multiple copies” mean at least two copies. A “copy” does not necessarily mean perfect sequence complementarity or identity to the template sequence. For example, copies can include nucleotide analogs such as deoxyinosine, intentional sequence alterations (such as sequence alterations introduced through a primer comprising a sequence that is hybridizable, but not complementary, to the template), and/or sequence errors that occur during amplification.
[0111] The technique of “polymerase chain reaction” or “PCR” as used herein, generally refers to a procedure wherein minute amounts of a specific piece of nucleic acid, RNA and/or DNA, are amplified as described, for example, in U.S. Pat. No. 4,683,195. Generally, sequence information from the ends of the region of interest or beyond needs to be available, such that oligonucleotide primers can be designed; these primers will be identical or similar in sequence to opposite strands of the template to be amplified. The 5′ terminal nucleotides of the two primers may coincide with the ends of the amplified material. PCR can be used to amplify specific RNA sequences, specific DNA sequences from total genomic DNA, and cDNA transcribed from total cellular RNA, bacteriophage, or plasmid sequences, etc. See generally Mullis et al., *Cold Spring Harbor Symp. Quant. Biol.* 51:263 (1987) and Erlich, ed., *PCR Technology*, (Stockton Press, NY, 1989). As used herein, PCR is considered to be one, but not the only, example of a nucleic acid polymerase reaction method for amplifying a nucleic acid test sample, comprising the use of a known nucleic acid (DNA or RNA) as a primer and utilizes a nucleic acid polymerase to amplify or generate a specific piece of nucleic acid or to amplify or generate a specific piece of nucleic acid which is complementary to a particular nucleic acid.
[0112] The term “multiplex-PCR” refers to a single PCR reaction carried out on nucleic acid obtained from a single source (e.g., an individual) using more than one primer set for the purpose of amplifying two or more DNA sequences in a single reaction.
[0113] “Quantitative real-time polymerase chain reaction” or “qRT-PCR” refers to a form of PCR wherein the amount of PCR product is measured at each step in a PCR reaction. This technique has been described in various publications including, for example, Cronin et al., Am. J. Pathol. 164(1):35-42 (2004) and Ma et al., *Cancer Cell* 5:607-616 (2004).
[0114] The term “microarray” refers to an ordered arrangement of hybridizable array elements, preferably polynucleotide probes, on a substrate.
[0115] The term “RNAseq,” also called “RNA-seq” or “Whole Transcriptome Shotgun Sequencing (WTSS),” refers to the use of high-throughput sequencing technologies to sequence and/or quantify cDNA to obtain information about a sample's RNA content. Publications describing RNAseq include: Wang et al. *Nature Reviews Genetics* 10(1):57-63, 2009; Ryan et al. *BioTechniques* 45(1):81-94, 2008; and Maher et al. *Nature* 458(7234):97-101, 2009.
[0116] “Polynucleotide” or “nucleic acid,” as used interchangeably herein, refers to polymers of nucleotides of any length, and include DNA and RNA. The nucleotides can be deoxyribonucleotides, ribonucleotides, modified nucleotides or bases, and/or their analogs, or any substrate that can be incorporated into a polymer by DNA or RNA polymerase, or by a synthetic reaction. Thus, for instance, polynucleotides as defined herein include, without limitation, single- and double-stranded DNA, DNA including single- and double-stranded regions, single- and double-stranded RNA, and RNA including single- and double-stranded regions, hybrid molecules comprising DNA and RNA that may be single-stranded or, more typically, double-stranded or include single- and double-stranded regions. In addition, the term “polynucleotide” as used herein refers to triple-stranded regions comprising RNA or DNA or both RNA and DNA. The strands in such regions may be from the same molecule or from different molecules. The regions may include all of one or more of the molecules, but more typically involve only a region of some of the molecules. One of the molecules of a triple-helical region often is an oligonucleotide. The terms “polynucleotide” and “nucleic acid” specifically includes mRNA and cDNAs.

[0117] A polynucleotide may comprise modified nucleotides, such as methylated nucleotides and their analogs. If present, modification to the nucleotide structure may be imparted before or after assembly of the polymer. The sequence of nucleotides may be interrupted by non-nucleotide components. A polynucleotide may be further modified after synthesis, such as by conjugation with a label. Other types of modifications include, for example, “caps,” substitution of one or more of the naturally-occurring nucleotides with an analog, internucleotide modifications such as, for example, those with uncharged linkages (e.g., methyl phosphonates, phosphotriesters, phosphoamidates, carbamates, and the like) and with charged linkages (e.g., phosphorothioates, phosphorodithioates, and the like), those containing pendant moieties, such as, for example, proteins (e.g., nucleases, toxins, antibodies, signal peptides, poly-L-lysine, and the like), those with intercalators (e.g., acridine, psoralen, and the like), those containing chelators (e.g., metals, radioactive metals, boron, oxidative metals, and the like), those containing alkylators, those with modified linkages (e.g., alpha anomeric nucleic acids), as well as unmodified forms of the polynucleotide(s). Further, any of the hydroxyl groups ordinarily present in the sugars may be replaced, for example, by phosphonate groups, phosphate groups, protected by standard protecting groups, or activated to prepare additional linkages to additional nucleotides, or may be conjugated to solid or semi-solid supports. The 5′ and 3′ terminal OH can be phosphorylated or substituted with amines or organic capping group moieties of from 1 to 20 carbon atoms. Other hydroxyls may also be derivatized to standard protecting groups. Polynucleotides can also contain analogous forms of ribose or deoxyribose sugars that are generally known in the art, including, for example, 2′-O-methyl-, 2′-O-allyl-, 2′-fluoro-, or 2′-azido-ribose, carbocyclic sugar analogs, α-anomeric sugars, epimeric sugars such as arabinose, xyloses or lyxoses, pyranose sugars, furanose sugars, sedoheptuloses, acyclic analogs, and abasic nucleoside analogs such as methyl riboside. One or more phosphodiester linkages may be replaced by alternative linking groups. These alternative linking groups include, but are not limited to, embodiments wherein phosphate is replaced by P(O)S (“thioate”), P(S)S (“dithioate”), “(O)NR.sub.2 (“amidate”), P(O)R, P(O)OR′, CO or CH.sub.2 (“formacetal”), in which each R or R′ is independently H or substituted or unsubstituted alkyl (1-20 C) optionally containing an ether (—O—) linkage, aryl, alkenyl, cycloalkyl, cycloalkenyl or araldyl. Not all linkages in a polynucleotide need be identical. The preceding description applies to all polynucleotides referred to herein, including RNA and DNA.

[0118] “Oligonucleotide,” as used herein, generally refers to short, single-stranded polynucleotides that are, but not necessarily, less than about 250 nucleotides in length. Oligonucleotides may be synthetic. The terms “oligonucleotide” and “polynucleotide” are not mutually exclusive. The description above for polynucleotides is equally and fully applicable to oligonucleotides.

[0119] The term “primer” or “probe” as used interchangeably herein, refers to a single-stranded polynucleotide that is capable of hybridizing to a nucleic acid and allowing polymerization of a complementary nucleic acid, generally by providing a free 3′-OH group.

[0120] An “isolated” nucleic acid molecule is a nucleic acid molecule that is identified and separated from at least one contaminant nucleic acid molecule with which it is ordinarily associated in the natural source of the nucleic acid. An isolated nucleic acid molecule is other than in the form or setting in which it is found in nature. Isolated nucleic acid molecules therefore are distinguished from the nucleic acid molecule as it exists in natural cells.

[0121] The term “diagnosis” is used herein to refer to the identification or classification of a molecular or pathological state, disease or condition (e.g., RA). For example, “diagnosis” may refer to the classification of a particular pathotype of RA, for instance, by histopathological criteria, or by molecular features (e.g., a subtype characterized by expression of one or a combination of biomarkers (e.g., particular genes or proteins encoded by said genes)).

[0122] As used herein, the terms “individual,” “patient,” and “subject” are used interchangeably and refer to any single animal, more preferably a mammal (including such non-human animals as, for example, cats, dogs, horses, rabbits, zoo animals, cows, pigs, sheep, and non-human primates)

for which treatment is desired. In particular embodiments, the patient herein is a human. The patient may be an “RA patient,” i.e., one who is suffering from RA, or at risk for suffering from RA, or suffering from one or more symptoms of RA.

[0123] As used herein, “treatment” (and grammatical variations thereof such as “treat” or “treating”) refers to clinical intervention in an attempt to alter the natural course of the individual being treated, and can be performed either for prophylaxis or during the course of clinical pathology. Desirable effects of treatment include, but are not limited to, preventing occurrence or recurrence of disease, alleviation of symptoms, diminishment of any direct or indirect pathological consequences of the disease, decreasing the rate of disease progression, amelioration or palliation of the disease state, and remission or improved prognosis. In some embodiments, treatment is with an RA therapy that includes a DMARD (e.g., methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, or mycophenolate mofetil) and/or a biologic therapeutic agent (e.g., a B cell antagonist (e.g., rituximab), a Janus kinase (JAK) antagonist (e.g., tofacitinib), tumor necrosis factor (TNF) antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor antagonist, or an IL-6 receptor antagonist (e.g., tocilizumab)) are used to delay development of a disease or to slow the progression of a disease or disorder (e.g., RA).

[0124] As used herein, “administering” is meant a method of giving a dosage of a compound (e.g., a DMARD (e.g., methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, or mycophenolate mofetil) and/or a biologic therapeutic agent (e.g., a B cell antagonist (e.g., rituximab), a Janus kinase (JAK) antagonist (e.g., tofacitinib), tumor necrosis factor (TNF) antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor antagonist, or an IL-6 receptor antagonist (e.g., tocilizumab)) to an individual. The compositions utilized in the methods described herein can be administered, for example, orally, subcutaneously, intramuscularly, intravenously, intradermally, percutaneously, intraarterially, intraperitoneally, intralesionally, intracranially, intraarticularly, intraprostatically, intrapleurally, intratracheally, intrathecally, intranasally, intravaginally, intrarectally, topically, intratumorally, peritoneally, subconjunctivally, intravesicularly, mucosally, intrapericardially, intraumbilically, intraocularly, intraorbitally, intravitreally (e.g., by intravitreal injection), by eye drop, topically, transdermally, parenterally, by inhalation, by injection, by implantation, by infusion, by continuous infusion, by localized perfusion bathing target cells directly, by catheter, by lavage, in creams, or in lipid compositions. In some instances, the compositions utilized in the methods described herein can be administered orally. In some instances, the compositions utilized in the methods described herein can be administered subcutaneously. The compositions utilized in the methods described herein can also be administered systemically or locally. The method of administration can vary depending on various factors (e.g., the compound or composition being administered and the severity of the condition, disease, or disorder (e.g., RA) being treated).

[0125] The term “concurrently” is used herein to refer to administration of two or more therapeutic agents, where at least part of the administration overlaps in time. Accordingly, concurrent administration includes a dosing regimen when the administration of one or more agent(s) continues after discontinuing the administration of one or more other agent(s).

[0126] By “reduce or inhibit” is meant the ability to cause an overall decrease of 20%, 30%, 40%, 50%, 60%, 70%, 75%, 80%, 85%, 90%, 95%, or greater. Reduce or inhibit can refer, for example, to the symptoms of the disorder (e.g., RA) being treated.

[0127] A “therapeutically effective amount” or “effective amount” refers to an amount of a therapeutic agent to treat or prevent a disease or disorder (e.g., RA) in a mammal. In the case of RA, the therapeutically effective amount of the therapeutic agent may reduce the potential for rheumatoid arthritis, reduce the occurrence of the rheumatoid arthritis, and/or a reduce the severity of rheumatoid arthritis, preferably, to an extent that the individual no longer suffers discomfort and/or altered function due to it. For example, a therapeutically effective amount can refer to the

amount necessary of a therapy, when administered to a subject, to prevent rheumatoid arthritis from occurring and/or to cure or to alleviate rheumatoid arthritis symptoms, signs, or causes. A therapeutically effective amount also refers to the amount of a therapeutic necessary to mitigate or decrease at least one disease activity index and/or clinical symptom, and/or inhibit, delay, or reverse the progression of the condition and/or prevent or delay of the onset of a disease or illness. For an RA therapy, efficacy in vivo can, for example, be determined by using the ACR and/or European League Against Rheumatism (EULAR) clinical response parameters in the patients with RA, or by assaying a molecular determinant of the degree of RA in the patient.

[0128] The term “antagonist” is used in the broadest sense, and includes any molecule that partially or fully blocks, inhibits, or neutralizes a biological activity of a native polypeptide. In a similar manner, the term “agonist” is used in the broadest sense and includes any molecule that mimics a biological activity of a native polypeptide. Suitable agonist or antagonist molecules specifically include agonist or antagonist antibodies or antibody fragments, fragments or amino acid sequence variants of native polypeptides, small molecules inhibitors, and the like. Methods for identifying agonists or antagonists of a polypeptide may comprise contacting a polypeptide with a candidate agonist or antagonist molecule and measuring a detectable change in one or more biological activities normally associated with the polypeptide.

[0129] A “B cell antagonist” as used herein is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of a B cell with one or more of its interaction partners. In some embodiments, a B cell antagonist is a molecule that upon binding to a B cell surface marker, destroys or depletes B cells in a mammal and/or interferes with one or more B cell functions, e.g., by reducing or preventing a humoral response elicited by the B cell. The antagonist in certain instances is able to deplete B cells (i.e. reduce circulating B cell levels) in a mammal treated therewith. Such depletion may be achieved via various mechanisms such as ADCC and/or CDC, inhibition of B cell proliferation, and/or induction of B cell death (e.g. via apoptosis). In some embodiments, a “B cell antagonist” is a molecule that inhibits the binding of CD20 to its binding partners. In some embodiments, the B cell antagonist inhibits the activation of CD20. In some embodiments, the B cell antagonist includes an anti-CD20 antibody (e.g., rituximab), antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction of CD20 with one or more of its interaction partners. In some embodiments, the B cell antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the B cell antagonist (e.g., the B cell binding antagonist) inhibits CD20. In a particular embodiment, a B cell antagonist has a binding affinity (dissociation constant) to a B cell, or a molecule expressed on the surface thereof, of about 1,000 nM or less. In another embodiment, a B cell antagonist has a binding affinity to a B cell, or a molecule expressed on the surface thereof, of about 100 nM or less. In another embodiment, a B cell antagonist has a binding affinity to a B cell, or a molecule expressed on the surface thereof, of about 50 nM or less. In another embodiment, a B cell antagonist has a binding affinity to a B cell, or a molecule expressed on the surface thereof, of about 10 nM or less. In another embodiment, a B cell antagonist has a binding affinity to a B cell, or a molecule expressed on the surface thereof, of about 1 nM or less. In a particular embodiment, a B cell antagonist inhibits B cell signaling with an IC50 of 1,000 nM or less. In another embodiment, a B cell antagonist inhibits B cell signaling with an IC50 of 500 nM or less. In another embodiment, a B cell antagonist inhibits B cell signaling with an IC50 of 50 nM or less. In another embodiment, a B cell antagonist inhibits B cell signaling with an IC50 of 10 nM or less. In another embodiment, a B cell antagonist inhibits B cell signaling with an IC50 of 1 nM or less.

[0130] A “Janus kinase antagonist” or “JAK antagonist,” as used interchangeable herein, is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of a Janus kinase with one or more of its interaction partners. In some

embodiments, a Janus kinase antagonist is a molecule that upon binding to a Janus kinase, decreases, blocks, inhibits, abrogates, or interferes with JAK/Signal Transducer and Activator of Transcription (STAT) activation or function in a mammal treated therewith. In some embodiments, the Janus kinase antagonist includes an antibody (e.g., tofacitinib), antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction a Janus kinase with one or more of its interaction partners. In some embodiments, the Janus kinase antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the Janus kinase antagonist inhibits a Janus kinase. In a particular embodiment, a Janus kinase antagonist has a binding affinity (dissociation constant) to a Janus kinase of about 1,000 nM or less. In another embodiment, a Janus kinase antagonist has a binding affinity to a Janus kinase of about 100 nM or less. In another embodiment, a Janus kinase antagonist has a binding affinity to a Janus kinase of about 50 nM or less. In another embodiment, a Janus kinase antagonist has a binding affinity to a Janus kinase of about 10 nM or less. In another embodiment, a Janus kinase antagonist has a binding affinity to a Janus kinase of about 1 nM or less. In a particular embodiment, a Janus kinase antagonist inhibits Janus kinase signaling with an IC50 of 1,000 nM or less. In another embodiment, a Janus kinase antagonist inhibits Janus kinase signaling with an IC50 of 500 nM or less. In another embodiment, a Janus kinase antagonist inhibits Janus kinase signaling with an IC50 of 50 nM or less. In another embodiment, a Janus kinase antagonist Janus kinase signaling with an IC50 of 10 nM or less. In another embodiment, a Janus kinase antagonist inhibits Janus kinase signaling with an IC50 of 1 nM or less.

[0131] A “tumor necrosis factor antagonist” or “TNF antagonist,” as used interchangeable herein, is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of a TNF with one or more of its interaction partners. In some embodiments, a TNF antagonist is a molecule that upon binding to a TNF, decreases, blocks, inhibits, abrogates, or interferes with TNF activation or function in a mammal treated therewith. In some embodiments, the TNF antagonist includes an antibody, antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction a TNF with one or more of its interaction partners. In some embodiments, the TNF antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the TNF antagonist inhibits TNF. In a particular embodiment, a TNF antagonist has a binding affinity (dissociation constant) to a TNF of about 1,000 nM or less. In another embodiment, a TNF antagonist has a binding affinity to a TNF of about 100 nM or less. In another embodiment, a TNF antagonist has a binding affinity to a TNF of about 50 nM or less. In another embodiment, a TNF has a binding affinity to a TNF of about 10 nM or less. In another embodiment, a TNF antagonist has a binding affinity to a TNF of about 1 nM or less. In a particular embodiment, a TNF antagonist inhibits TNF signaling with an IC50 of 1,000 nM or less. In another embodiment, a TNF antagonist inhibits TNF signaling with an IC50 of 500 nM or less. In another embodiment, a TNF antagonist TNF signaling with an IC50 of 50 nM or less. In another embodiment, a TNF antagonist TNF signaling with an IC50 of 10 nM or less. In another embodiment, a TNF antagonist inhibits TNF signaling with an IC50 of 1 nM or less.

[0132] A “decoy tumor necrosis factor receptor” or “decoy TNF receptor,” as used interchangeable herein, is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of a TNF with one or more of its interaction partners. In some embodiments, a decoy TNF receptor is a molecule that upon binding to a TNF, decreases, blocks, inhibits, abrogates, or interferes with TNF activation or function in a mammal treated therewith. In some embodiments, the decoy TNF receptor includes an antibody, antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction a TNF with one or more of its interaction partners. In some embodiments, the decoy

TNF receptor is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the decoy TNF receptor inhibits TNF. In a particular embodiment, a decoy TNF receptor has a binding affinity (dissociation constant) to a TNF of about 1,000 nM or less. In another embodiment, a decoy TNF receptor has a binding affinity to a TNF of about 100 nM or less. In another embodiment, a decoy TNF receptor has a binding affinity to a TNF of about 50 nM or less. In another embodiment, a decoy TNF receptor has a binding affinity to a TNF of about 10 nM or less. In another embodiment, a decoy TNF receptor has a binding affinity to a TNF of about 1 nM or less. In a particular embodiment, a decoy TNF receptor inhibits TNF signaling with an IC50 of 1,000 nM or less. In another embodiment, a decoy TNF receptor inhibits TNF signaling with an IC50 of 500 nM or less. In another embodiment, a decoy TNF receptor TNF signaling with an IC50 of 50 nM or less. In another embodiment, a decoy TNF receptor TNF signaling with an IC50 of 10 nM or less. In another embodiment, a decoy TNF receptor inhibits TNF signaling with an IC50 of 1 nM or less.

[0133] A “T cell costimulatory signal antagonist” as used herein is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of a T cell with one or more of its interaction partners. In some embodiments, a T cell costimulatory signal antagonist is a molecule that upon binding to a T cell, decreases, blocks, inhibits, abrogates, or interferes with the T cell. In some embodiments, a “T cell costimulatory signal antagonist” is a molecule that inhibits the binding of CD80 and CD86 to its binding partners. In some embodiments, the T cell costimulatory signal antagonist inhibits the activation of a T cell. In some embodiments, the T cell costimulatory signal antagonist includes an antibody, antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction of a T cell with one or more of its interaction partners. In some embodiments, the T cell costimulatory signal antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the T cell costimulatory signal antagonist inhibits a T cell. In a particular embodiment, a T cell costimulatory signal antagonist has a binding affinity (dissociation constant) to a T cell, or a molecule expressed on the surface thereof, of about 1,000 nM or less. In another embodiment, a T cell costimulatory signal antagonist has a binding affinity to a T cell, or a molecule expressed on the surface thereof, of about 100 nM or less. In another embodiment, a T cell costimulatory signal antagonist has a binding affinity to a T cell of about 50 nM or less. In another embodiment, a T cell costimulatory signal antagonist has a binding affinity to a T cell, or a molecule expressed on the surface thereof, of about 10 nM or less. In another embodiment, a T cell costimulatory signal antagonist has a binding affinity to a T cell, or a molecule expressed on the surface thereof, of about 1 nM or less. In a particular embodiment, a T cell costimulatory signal antagonist inhibits T cell signaling with an IC50 of 1,000 nM or less. In another embodiment, a T cell costimulatory signal antagonist inhibits T cell signaling with an IC50 of 500 nM or less. In another embodiment, a T cell costimulatory signal antagonist inhibits T cell signaling with an IC50 of 50 nM or less. In another embodiment, a T cell costimulatory signal antagonist inhibits T cell signaling with an IC50 of 10 nM or less. In another embodiment, a T cell costimulatory signal antagonist inhibits T cell signaling with an IC50 of 1 nM or less.

[0134] An “IL-1 receptor antagonist,” “interleukin 1 receptor antagonist,” or “IL-1R antagonist” as used interchangeably herein is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of IL-1 receptor with one or more of its interaction partners. In some embodiments, an IL-1 receptor antagonist is a molecule that upon binding to an IL-1 receptor, decreases, blocks, inhibits, abrogates, or interferes with IL-1R activation or function in a mammal treated therewith. In some embodiments, the IL-1 receptor antagonist includes an antibody, antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction of IL-1 receptor with one or more of its

interaction partners. In some embodiments, the IL-1 receptor antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the IL-1 receptor antagonist (e.g., the IL-1 receptor binding antagonist) inhibits IL-1. In a particular embodiment, an IL-1R inhibitor has a binding affinity (dissociation constant) to IL-1R of about 1,000 nM or less. In another embodiment, an IL-1R antagonist has a binding affinity to IL-1R of about 100 nM or less. In another embodiment, an IL-1R antagonist has a binding affinity to IL-1R of about 50 nM or less. In another embodiment, an IL-1R antagonist has a binding affinity to IL-1R of about 10 nM or less. In another embodiment, an IL-1R antagonist has a binding affinity to IL-1R of about 1 nM or less. In a particular embodiment, an IL-1R antagonist inhibits IL-1R signaling with an IC50 of 1,000 nM or less. In another embodiment, an IL-1R antagonist inhibits IL-1R signaling with an IC50 of 500 nM or less. In another embodiment, an IL-1R antagonist inhibits IL-1R signaling with an IC50 of 50 nM or less. In another embodiment, an IL-1R antagonist inhibits IL-1R signaling with an IC50 of 10 nM or less. In another embodiment, an IL-1R antagonist inhibits IL-1R signaling with an IC50 of 1 nM or less.

[0135] An “IL-6 receptor antagonist,” “interleukin 6 receptor antagonist,” or “IL-6R antagonist,” as used interchangeably herein, is any molecule that decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of IL-6 receptor with one or more of its interaction partners. In some embodiments, an IL-6 receptor antagonist is a molecule that upon binding to an IL-6 receptor, decreases, blocks, inhibits, abrogates, or interferes with IL-6R activation or function in a mammal treated therewith. In some embodiments, the IL-6 receptor antagonist includes an antibody (e.g., tocilizumab), antigen-binding fragments thereof, an immunoadhesin, a fusion protein, an oligopeptide, and other molecules that decrease, block, inhibit, abrogate, or interfere with signal transduction resulting from the interaction of IL-6 receptor with one or more of its interaction partners. In some embodiments, the IL-6 receptor antagonist is a polypeptide, a small molecule, or a nucleic acid. In some embodiments, the IL-6 receptor antagonist (e.g., the IL-6 receptor binding antagonist) inhibits IL-6. In a particular embodiment, an IL-6R antagonist has a binding affinity (dissociation constant) to IL-6R of about 1,000 nM or less. In another embodiment, an IL-6R antagonist has a binding affinity to IL-6R of about 100 nM or less. In another embodiment, an IL-6R antagonist has a binding affinity to IL-6R of about 50 nM or less. In another embodiment, an IL-6R antagonist has a binding affinity to IL-6R of about 10 nM or less. In another embodiment, an IL-6R antagonist has a binding affinity to IL-6R of about 1 nM or less. In a particular embodiment, an IL-6R antagonist inhibits IL-6R signaling with an IC50 of 1,000 nM or less. In another embodiment, an IL-6R antagonist inhibits IL-6R signaling with an IC50 of 500 nM or less. In another embodiment, an IL-6R antagonist inhibits IL-6R signaling with an IC50 of 50 nM or less. In another embodiment, an IL-6R antagonist inhibits IL-6R signaling with an IC50 of 10 nM or less. In another embodiment, an IL-6R antagonist inhibits IL-6R signaling with an IC50 of 1 nM or less.

[0136] The term “antibody” herein is used in the broadest sense and specifically covers monoclonal antibodies, polyclonal antibodies, half-antibodies, multispecific antibodies (e.g., bispecific antibodies) formed from at least two intact antibodies, and antibody fragments so long as they exhibit the desired biological activity. The term “immunoglobulin” (Ig) is used interchangeably with antibody herein.

[0137] A “blocking” antibody or an antibody “antagonist” is one which inhibits or reduces biological activity of the antigen it binds. For example, an IL-6 receptor antagonist antibody binds IL-6 receptor and decreases, blocks, inhibits, abrogates, or interferes with signal transduction resulting from the interaction of IL-6 receptor with IL-6. Preferred blocking antibodies or antagonist antibodies completely inhibit the biological activity of the antigen.

[0138] An “article of manufacture” is any manufacture (e.g., a package or container) or kit comprising at least one reagent, e.g., a medicament for treatment of a disease or disorder (e.g., RA), or a probe for specifically detecting a biomarker (e.g., one or more of CD180, CSF2, CXCL1,

DENND1C, MMP10, SDC1, and UBASH3A) described herein. In certain embodiments, the manufacture or kit is promoted, distributed, or sold as a unit for performing the methods described herein.

[0139] The term “package insert” is used to refer to instructions customarily included in commercial packages of therapeutic products, that contain information about the indications, usage, dosage, administration, combination therapy, contraindications, and/or warnings concerning the use of such therapeutic products.

[0140] A “sterile” formulation is aseptic or free from all living microorganisms and their spores. The phrase “based on” when used herein means that the information about one or more biomarkers is used to inform a treatment decision, information provided on a package insert, or marketing/promotional guidance.

[0141] A “pharmaceutically acceptable carrier” refers to an ingredient in a pharmaceutical formulation, other than an active ingredient, which is nontoxic to a subject. A pharmaceutically acceptable carrier includes, but is not limited to, a buffer, excipient, stabilizer, or preservative.

[0142] “Disease progression” or “progression” as used interchangeably herein, refers to radiographic progression and/or increased disease activity. Radiographic progression can be defined, for example, by an increase in ShSS (e.g., an increase in ShSS over a defined time period). For example, the time period may be, at a minimum, one year after initial examination.

[0143] “Disease activity” or “rheumatoid arthritis disease activity” as used herein, refer to the severity or intensity of rheumatoid arthritis and can be determined by, for example, ACR and/or EULAR clinical indices including, but not limited to, DAS28-ESR, DAS28-CRP, levels of ESR, CRP, ACPA titer, RF titer, swollen joint counts, VAS, assessment of joint damage (e.g., by x-ray, Ultrasound Synovial Thickening (USST) assessment, or Ultrasound Power Doppler scores (USPD)), or a combination thereof. A DAS28-ESR of <3.2 is indicative of low disease activity, while a DAS28-ESR of 3.2-5.1 is indicative of moderate disease activity, and a DAS28-ESR of >5.1 is indicative of the most severe disease activity. RF titer seropositivity indicates high disease activity, while RF seronegativity indicates low disease activity. An ESR level of greater than 28 mm/Hr, a CRP level of greater than 1 mg/dL, a positive ACPA titer, swollen joint counts of at least one, and/or the presence of erosion or joint space narrowing by X-ray are abnormal and indicative of disease activity.

[0144] “Responsiveness” or “effective response” can be assessed using any endpoint indicating a benefit to the individual and includes, without limitation, (i) inhibition, to some extent, of disease progression, including slowing down and complete arrest; (ii) reduction in the number of disease episodes and/or symptoms; (iii) reduction in lesional size; (iv) inhibition (i.e., reduction, slowing down or complete stopping) of disease cell infiltration into adjacent peripheral organs and/or tissues; (v) inhibition (i.e. reduction, slowing down or complete stopping) of disease spread; (vi) decrease of autoimmune response, which may, but does not have to, result in the regression or ablation of the disease lesion; (vii) relief, to some extent, of one or more symptoms associated with the disorder; (viii) increase in the length of disease-free presentation following treatment; and/or (ix) decreased mortality at a given point of time following treatment.

II. Methods

[0145] Provided herein are methods and assays for diagnosing a patient having rheumatoid arthritis (RA); identifying an individual having RA who is likely to exhibit disease progression; identifying an individual having RA who may benefit from a treatment including a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD); identifying an individual having RA who may benefit from a treatment including a DMARD; determining whether an individual having RA is likely to respond to treatment with an RA therapy that includes a DMARD; selecting a therapy for an individual having RA; treating an individual having RA based on a diagnostic method of the invention; optimizing therapeutic efficacy of an RA therapy; and monitoring therapeutic efficacy of an RA therapy. The methods and assays described herein are

based on the finding that the expression level of at least one or more biomarkers described herein in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) from an individual having RA can be used to predict the therapeutic efficacy of an RA therapy, for example, a DMARD (i.e., a DMARD), or a therapy including a biologic therapeutic agent (i.e., a biologic therapeutic agent), alone or in combination with a DMARD. Any of the methods and assays may further include determining a myeloid, lymphoid, and/or pauciimmune-fibroid eigengene score. Any of the methods and assays provided herein may further include administering a DMARD (e.g., a DMARD described in Section II-B below) to the individual. Accordingly, provided herein are also methods and assays of evaluating the expression of one or more biomarkers in a sample from an individual. Any of the methods provided herein may include administering an RA therapy other than, or in additional to, a DMARD (e.g., an RA therapy other than, or in additional to, a DMARD described in Section II-B, below) to the individual. Any of the methods may further include administering an effective amount of an additional therapeutic agent, as described herein, to the individual.

A. Diagnostic Methods and Assays

(i) Prognostic Diagnostic Methods and Assays

[0146] The present invention provides methods that may be used to identify an individual having RA who is likely to exhibit disease progression, the methods and assays including determining an expression level of one or more genes set forth in Table 1 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, or 46 genes set forth in Table 1) in a sample from the individual, wherein a change in the expression level of the one or more genes relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression.

TABLE-US-00001 TABLE 1 NCBI Gene ID Gene Name NCBI Gene ID Gene Name 4064 CD180 27177 IL36B 1437 CSF2 3662 IRF4 2919 CXCL1 100505746 ITGB2-AS1 79958 DENND1C 4063 LY9 4319 MMP10 11184 MAP4K1 6382 SDC1 4312 MMP1 53347 UBASH3A 9241 NOG 640 BLK 11040 PIM2 695 BTK 5450 POU2AF1 151888 BTLA 399 RHOH 6363 CCL19 23231 SEL1L3 930 CD19 55423 SIRPG 952 CD38 114836 SLAMF6 959 CD40LG 1317 SLC31A1 8483 CILP 6689 SPIB 79961 DENND2D 201633 TIGIT 27122 DKK3 81793 TLR10 83416 FCRL5 11322 TMC6 2247 FGF2 7124 TNF 2254 FGF9 4982 TNFRSF11B 3112 HLA-DOB 608 TNFRSF17 29851 ICOS 80342 TRAF3PIP3 3512 JCHAIN 7494 XBP1

[0147] In some instances, the methods and assays provided herein may involve determining an expression level of one or more genes set forth Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) or Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) in a sample from an individual, wherein a change in the expression level of the one or more genes set forth in Table 2 or Table 3 relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression.

TABLE-US-00002 TABLE 2 NCBI Gene ID Gene Name NCBI Gene ID Gene Name 640 BLK 11184 MAP4K1 695 BTK 4312 MMP1 151888 BTLA 4319 MMP10 6363 CCL19 11040 PIM2 4064 CD180 5450 POU2AF1 930 CD19 399 RHOH 952 CD38 6382 SDC1 959 CD40LG 23231 SEL1L3 1437 CSF2 55423 SIRPG 2919 CXCL1 114836 SLAMF6 79958 DENND1C 1317 SLC31A1 79961 DENND2D 6689 SPIB 83416 FCRL5 201633 TIGIT 3112 HLA-DOB 81793 TLR10 29851 ICOS 11322 TMC6 3512 JCHAIN 7124 TNF 27177 IL36B 608 TNFRSF17 3662 IRF4 80342 TRAF3PIP3 100505746 ITGB2-AS1 53347 UBASH3A 4063 LY9 7494 XBP1

TABLE-US-00003 TABLE 3 NCBI Gene ID Gene Name 8483 CILP 27122 DKK3 2247 FGF2 2254 FGF9 9241 NOG 4982 TNFRSF11B

[0148] In some instances, the methods and assays provided herein may involve determining an expression level of one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37,

38, 39, or 40 genes set forth in Table 2). For example, the method may include determining the expression level of one or more genes set forth in Table 2, wherein an increase in the expression level (e.g., an increase in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of the one or more genes set forth in Table 2 relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression. In certain instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0149] In particular instances, the methods and assays may include determining an expression level of one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) in a sample from an individual. For example, the method or assay may include determining the expression level of one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, wherein an increase in the expression level (e.g., an increase in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of the one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression. In certain instances, the increased expression level of the one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the increased expression level of one or more of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0150] In particular instances, the method and assays may include determining an expression level of at least two of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample from an individual that is increased (e.g., an increased expression level of about 10%, 20%, 30%,

40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of the at least two genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of at least two of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the at least two genes. In some instances, the increased expression level of at least two of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least two genes. In some instances of the diagnostic method, the expression levels of a combination of two genes set forth in Table 1, such as any of the exemplary combinations shown in Table 4, may be determined.

TABLE-US-00004 TABLE 4 Two-Gene Combinations of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CD180 and CSF2 CD180 and CXCL1 CD180 and DENND1C CD180 and MMP10 CD180 and SDC1 CD180 and UBASH3A CSF2 and CXCL1 CSF2 and DENND1C CSF2 and MMP10 CSF2 and SDC1 CSF2 and UBASH3A CXCL1 and DENND1C CXCL1 and MMP10 CXCL1 and SDC1 CXCL1 and UBASH3A DENND1C and MMP10 DENND1C and SDC1 DENND1C and UBASH3A MMP10 and SDC1 MMP10 and UBASH3A SDC1 and UBASH3A

[0151] In particular instances, the method and assays may include determining an expression level of at least three of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample from an individual that is increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of the at least three genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of at least three of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the at least three genes. In some instances, the increased expression level of at least three of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least three genes. In some instances of the diagnostic method, the expression levels of a combination of three genes set forth in Table 1, such as any of the exemplary combinations shown in Table 5, may be determined.

TABLE-US-00005 TABLE 5 Three-Gene Combinations of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CD180, CSF2, and CXCL1 CD180, CSF2, and DENND1C CD180, CSF2, and MMP10 CD180, CSF2, and SDC1 CD180, CSF2, and UBASH3A CD180, CXCL1, and DENND1C CD180, CXCL1, and MMP10 CD180, CXCL1, and SDC1 CD180,

CXCL1, and UBASH3A CD180, DENND1C, and MMP10 CD180, DENND1C, and SDC1 CD180, DENND1C, and UBASH3A CD180, MMP10, and SDC1 CD180, MMP10, and UBASH3A CD180, SDC1, and UBASH3A CSF2, CXCL1 and DENND1C CSF2, CXCL1, and MMP10 CSF2, CXCL1, and SDC1 CSF2, CXCL1, and UBASH3A CSF2, DENND1C, and MMP10 CSF2, DENND1C, and SDC1 CSF2, DENND1C, and UBASH3A CSF2, MMP10, and SDC1 CSF2, MMP10, and UBASH3A CSF2, SDC1, and UBASH3A CXCL1, DENND1C, and MMP10 CXCL1, DENND1C and SDC1 CXCL1, DENND1C, and UBASH3A CXCL1, MMP10, and SDC1 CXCL1, MMP10, and UBASH3A CXCL1, SDC1, and UBASH3A DENND1C, MMP10, and SDC1 DENND1C, MMP10, and UBASH3A DENND1C, SDC1, and UBASH3A MMP10, SDC1, and UBASH3A

[0152] In particular instances, the method and assays may include determining an expression level of at least four of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample from an individual that is increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of the at least four genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of at least four of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the at least four genes. In some instances, the increased expression level of at least four of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least four genes. In some instances of the diagnostic methods, the expression levels of a combination of four genes set forth in Table 1, such as any of the exemplary combinations shown in Table 6, may be determined.

TABLE-US-00006 TABLE 6 Four-Gene Combinations of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CD180, CSF2, CXCL1, and DENND1C CD180, CSF2, CXCL1, and MMP10 CD180, CSF2, CXCL1, and SDC1 CD180, CSF2, CXCL1, and UBASH3A CD180, CSF2, DENND1C, and MMP10 CD180, CSF2, DENND1C, and SDC1 CD180, CSF2, DENND1C, and UBASH3A CD180, CSF2, MMP10, and SDC1 CD180, CSF2, MMP10, and UBASH3A CD180, CSF2, SDC1, and UBASH3A CD180, CXCL1, DENND1C, and MMP10 CD180, CXCL1, DENND1C, and SDC1 CD180, CXCL1, DENND1C, and UBASH3A CD180, CXCL1, MMP10, and SDC1 CD180, CXCL1, MMP10, and UBASH3A CD180, CXCL1, SDC1, and UBASH3A CD180, DENND1C, MMP10, and SDC1 CD180, DENND1C, MMP10, and UBASH3A CD180, DENND1C, SDC1, and UBASH3A CD180, MMP10, SDC1, and UBASH3A CSF2, CXCL1, DENND1C, and MMP10 CSF2, CXCL1, DENND1C, and SDC1 CSF2, CXCL1, DENND1C, and UBASH3A CSF2, CXCL1, MMP10, and SDC1 CSF2, CXCL1, MMP10, and UBASH3A CSF2, CXCL1, SDC1, and UBASH3A CSF2, DENND1C, MMP10, and SDC1 CSF2, DENND1C, MMP10, and UBASH3A CSF2, DENND1C, SDC1, and UBASH3A CSF2, MMP10, SDC1, and UBASH3A CXCL1, DENND1C, MMP10, and SDC1 CXCL1, DENND1C, MMP10, and UBASH3A CXCL1, DENND1C, SDC1, and UBASH3A CXCL1, MMP10, SDC1, and UBASH3A DENND1C, MMP10, SDC1, and UBASH3A

[0153] In particular instances, the method and assays may include determining an expression level of at least five of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in the

sample from an individual that is increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of at the least five genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of at least five of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the at least five genes. In some instances, the increased expression level of at least five of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least five genes. In some instances of the diagnostic methods, the expression levels of a combination of five genes set forth in Table 1, such as any of the exemplary combinations shown in Table 7, may be determined.

TABLE-US-00007 TABLE 7 Five-Gene Combinations of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CD180, CSF2, CXCL1, DENND1C, and MMP10 CD180, CSF2, CXCL1, DENND1C, and SDC1 CD180, CSF2, CXCL1, DENND1C, and UBASH3A CD180, CSF2, CXCL1, MMP10, and SDC1 CD180, CSF2, CXCL1, MMP10, and UBASH3A CD180, CSF2, CXCL1, SDC1, and UBASH3A CD180, CSF2, DENND1C, MMP10, and SDC1 CD180, CSF2, DENND1C, MMP10, and UBASH3A CD180, CSF2, DENND1C, SDC1, and UBASH3A CD180, CSF2, MMP10, SDC1, and UBASH3A CD180, CXCL1, DENND1C, MMP10, and SDC1 CD180, CXCL1, DENND1C, MMP10, and UBASH3A CD180, CXCL1, DENND1C, SDC1, and UBASH3A CD180, CXCL1, MMP10, SDC1, and UBASH3A CD180, DENND1C, MMP10, SDC1, and UBASH3A CSF2, CXCL1, DENND1C, MMP10, and SDC1 CSF2, CXCL1, DENND1C, MMP10, and UBASH3A CSF2, CXCL1, DENND1C, SDC1, and UBASH3A CSF2, CXCL1, MMP10, SDC1, and UBASH3A CSF2, DENND1C, MMP10, SDC1, and UBASH3A CXCL1, DENND1C, MMP10, SDC1, and UBASH3A

[0154] In particular instances, the method and assays may include determining an expression level of at least six of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample that is increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of the at least six genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of at least six of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the at least six genes. In some instances, the increased expression level of at least six of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least six

genes. In some instances of the diagnostic methods, the expression levels of a combination of six genes set forth in Table 1, such as any of the exemplary combinations shown in Table 8, may be determined.

TABLE-US-00008 TABLE 8 Six-Gene Combinations of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CD180, CSF2, CXCL1, DENND1C, MMP10, and SDC1 CD180, CSF2, CXCL1, DENND1C, MMP10, and UBASH3A CD180, CSF2, CXCL1, DENND1C, SDC1, and UBASH3A CD180, CSF2, CXCL1, MMP10, SDC1, and UBASH3A CD180, CSF2, DENND1C, MMP10, SDC1, and UBASH3A CD180, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A

[0155] In particular instances, the method and assays may include determining an expression level of the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample that is increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level of the seven genes identifies the individual as one who is likely to exhibit disease progression. In certain instances, the increased expression level of the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the seven genes. In some instances, the increased expression level of the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the seven genes.

[0156] In some instances, the methods and assays provided herein may involve determining an expression level of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) in a sample from an individual. For example, the method or assay may include determining the expression level of one or more genes set forth in Table 3, wherein a decrease in expression level (e.g., a decrease in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of the one or more genes set forth in Table 3 relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression. In certain instances, the decreased expression level of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the decreased expression level of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0157] In any of the prognostic methods and assays described above, the reference expression level may be a reference expression level in a reference population of individuals having RA who have

not been previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who exhibited disease progression and a second subset of individuals who did not exhibit disease progression. In some instances, the reference expression level significantly separates each of the first and second subsets of individuals based on a significant difference in the expression level of the one or more genes set forth in Table 1, Table 2, or Table 3 in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals exhibited disease progression, and the second subset of individuals did not exhibit disease progression after about 12 months.

[0158] In any of the prognostic methods and assays described above in which the expression level of one or more biomarkers, selected from the biomarkers set forth in Table 1, Table 2, or Table 3, are determined in a sample from an individual and compared to a reference expression level (e.g., a pre-assigned expression level of the one or more genes set forth in Table 1, Table 2, or Table 3), it is to be understood that, in some instances, the expression level of the one or more biomarkers may be an average of the expression level of the one or more biomarkers. In some instances, the expression level of the one or more biomarkers may be a median of the expression level of the one or more biomarkers. In some instances, the expression level the one or more biomarkers may be normalized, e.g., to a reference gene, e.g., a housekeeping gene. In some instances, the reference gene is ACTB, GAPDH, GUSB, HPRT1, PGK1, RPL19, TUBB, or TMEM55B. In some instances, the expression level of the one or more biomarkers may be an average of a normalized expression level of the one or more biomarkers. In some instances, the expression level of the one or more biomarkers may be a median of a normalized expression level of the one or more biomarkers.

[0159] In any of the prognostic methods and assays described above in which the expression level of more than one biomarker, selected from the genes set forth in Table 1, Table 2, or Table 3, is determined in a sample from an individual and compared to a reference expression level (e.g., a pre-assigned expression level of the one or more genes set forth in Table 1, Table 2, or Table 3), it is to be understood that, in some instances, the expression level of each individual biomarker in the sample is compared to a reference expression level for each individual biomarker. For example, if the expression level of CD180 and CXCL1 are determined in a sample from an individual and compared to reference expression levels for CD180 and CXCL1, in some instances, the expression level of CD180 in the sample from the individual is compared to the reference expression level for CD180, and the expression level of CXCL1 in the sample from the individual is compared to the reference expression level for CXCL1. In other instances, an expression level for more than one gene of interest may be determined by aggregation methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the median or mean of all the expression levels of the genes of interest. Before aggregation, the expression level of each gene of interest may be normalized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, normalized to the expression level of one or more housekeeping genes, or normalized to a total library size, or normalized to the median or mean expression level value across all genes measured. In some instances, before aggregation across multiple genes of interest, the normalized expression level of each gene of interest may be standardized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the Z-score of the normalized expression level of each gene of interest.

[0160] In any of the prognostic methods and assays described above, the disease progression may be radiographic progression. The radiographic progression may be characterized by an increase in ShSS (e.g., an increase in ShSS over a defined time period).

[0161] The present invention also provides methods and assays that may be used to identify an individual having RA who may be more likely to exhibit disease activity that involves determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score identifies the individual as one who may be more likely to exhibit disease activity. In some instances, the methods and assays

provided herein may further involve determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score from the sample that is at or above a reference lymphoid eigengene score identifies the individual as one who may be more likely to exhibit disease activity.

[0162] The present invention provides methods and assays that may be used to identify an individual having RA who may be less likely to exhibit disease activity that involves determining a pauciimmune-fibroid eigengene score from a sample from the individual, wherein a pauciimmune-fibroid eigengene score from the sample that is at or above a reference pauciimmune-fibroid eigengene score identifies the individual as one who may be less likely to exhibit disease activity.

[0163] In any of the prognostic methods and assays described above, the reference myeloid eigengene score may be a reference myeloid eigengene score in a reference population of individuals having RA, the population of individuals consisting of a first subset of individuals who exhibited disease activity and a second subset of individuals who did not exhibit disease activity. In some instances, the reference myeloid eigengene score level significantly separates each of the first and second subsets of individuals based on a significant difference in the myeloid eigengene score in the first subset of individuals compared to that of the second subset of individuals.

[0164] In any of the prognostic methods and assays described above, the reference lymphoid eigengene score may be a reference lymphoid eigengene score in a reference population of individuals having RA, the population of individuals consisting of a first subset of individuals who exhibited disease activity and a second subset of individuals who did not exhibit disease activity. In some instances, the reference lymphoid eigengene score level significantly separates each of the first and second subsets of individuals based on a significant difference in the lymphoid eigengene score in the first subset of individuals compared to that of the second subset of individuals.

[0165] In any of the prognostic methods and assays described above, the reference pauciimmune-fibroid eigengene score may be a reference pauciimmune-fibroid eigengene score in a reference population of individuals having RA, the population of individuals consisting of a first subset of individuals who exhibited disease activity and a second subset of individuals who did not exhibit disease activity. In some instances, the reference pauciimmune-fibroid eigengene score level significantly separates each of the first and second subsets of individuals based on a significant difference in the pauciimmune-fibroid eigengene score in the first subset of pauciimmune-fibroid compared to that of the second subset of individuals.

[0166] In any of the prognostic methods and assays described above, the severity of disease activity may be assessed by evaluating, for example, ACR and/or EULAR clinical indices including, but not limited to, DAS28-ESR, DAS28-CRP, levels of ESR, CRP, ACPA titer, assessment of joint damage (e.g., by x-ray, Ultrasound Synovial Thickening (USST) assessment, or Ultrasound Power Doppler (USPD) scores), or a combination thereof.

[0167] In any of the prognostic methods and assays described above, the methods and assays may further include determining one or more clinical covariates (e.g., baseline RF titer, disease duration, DAS28-ESR, DAS28-CRP, baseline pathotype, and 12 max USST and USPD scores) of the individual.

[0168] In any of the prognostic methods and assays described above, the methods and assays may further include administering to the individual a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD) (e.g., as described in Section II-B, below). In particular instances, when the change in expression level of the one or more genes set forth in Table 1 or Table 2 relative to a reference expression level is an increase, the method further includes administering to the individual a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD). In particular instances, when the change in expression level of the one or more genes set forth in Table 3 relative to a reference expression level is a decrease, the method further includes administering to the individual a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD).

[0169] In some instances of any of the preceding methods and assays described above, the RA therapeutic agent may be a DMARD (e.g., as described in Section II-B below). In some instances of any of the preceding methods described above, the RA therapeutic agent may be a therapeutic agent other than a DMARD (e.g., as described in Section II-B below).

[0170] In some instances, the individual has not been previously treated with a DMARD. In other instances, the individual has been previously treated with a DMARD.

(ii) Predictive Diagnostic Methods and Assays

[0171] The present invention provides methods and assays of identifying an individual having RA who may benefit from a treatment with an RA therapy that includes a DMARD (e.g., methotrexate) that involves determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score (e.g., above or an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) identifies the individual as one who may benefit from a treatment with a DMARD. In some instances, the methods and assays provided herein may further involve determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score from the sample that is at or above a reference lymphoid eigengene score (e.g., above or an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) identifies the individual as one who may benefit from a treatment with a DMARD.

[0172] The present invention also provides methods and assays for selecting a therapy for an individual having RA, the method or assay including determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score (e.g., above or an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) identifies the individual as one who may benefit from a treatment comprising a DMARD (e.g., methotrexate). In some instances a myeloid eigengene score that is above a reference myeloid eigengene score, or an elevated or increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an elevated myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, the methods and assays may further involve determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score from the sample that is at or above a reference lymphoid eigengene score (e.g., above or an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) identifies the individual as one who may benefit from a treatment comprising a DMARD. In some instances a lymphoid eigengene score that is above a reference lymphoid eigengene score or an elevated or increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an elevated lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-

fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0173] In any of the predictive methods and assays described above, the reference myeloid eigengene score may be a reference myeloid eigengene score in a reference population of individuals having RA who have been previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy. In some instances, the reference myeloid eigengene score significantly separates each of the first and second subsets of individuals based on a significant difference in the myeloid eigengene score in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.

[0174] In any of the predictive methods and assays described above, the reference lymphoid eigengene score may be a reference lymphoid eigengene score in a reference population of individuals having RA who have been previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy. In some instances, the reference lymphoid eigengene score significantly separates each of the first and second subsets of individuals based on a significant difference in the lymphoid eigengene score in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.

[0175] The present invention provides methods and assays that may be used to monitor the response of an individual having RA to treatment with a DMARD, the method or assay including (i) determining a myeloid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (ii) determining a second myeloid eigengene score from a sample from the individual at a second time point; and (iii) comparing the first myeloid eigengene score with a the second myeloid eigengene score, wherein a decrease in the second myeloid eigengene score (e.g., a decrease in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to the first myeloid eigengene score is predictive of an individual who is likely to respond to treatment with a DMARD. The methods and assays may further involve (i) determining a lymphoid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (ii) determining a second lymphoid eigengene score from a sample from the individual at a second time point; and (iii) comparing the first lymphoid eigengene score with a the second lymphoid eigengene score, wherein a decrease in the second lymphoid eigengene score (e.g., a decrease in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to the first lymphoid eigengene score is further predictive of an individual who is likely to respond to treatment with a DMARD. In some instances, a reduced or decreased myeloid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, a reduced myeloid eigengene score refers to an overall decrease of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold,

about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, a reduced or decreased lymphoid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, a reduced lymphoid eigengene score refers to an overall decrease of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0176] In some instances, the second myeloid eigengene score is decreased relative to the first myeloid eigengene score, and the method or assay further involves administering and additional dose of a DMARD to the individual. In some instances, the second lymphoid eigengene score is decreased relative to the first lymphoid eigengene score, and the method or assay further involves administering and additional dose of a DMARD to the individual.

[0177] In some instances, the second myeloid eigengene score is decreased relative to the first myeloid eigengene score, the second lymphoid eigengene score is decreased relative to the first lymphoid eigengene score, and the method or assay further involves administering and additional dose of a DMARD to the individual.

[0178] In some instances of any of the preceding methods or assays involving determining a myeloid eigengene score and/or lymphoid eigengene score, the individual has been previously treated with a DMARD. In other instances, the individual has not been previously treated with a DMARD.

iii. Exemplary Approaches for Determination of Biomarker Expression Levels

[0179] The methods and assays provided herein may include determining an expression level of one or more genes in a sample (e.g., a synovial tissue sample, a synovial fluid sample, or a combination thereof) from an individual. The sample from the individual may be an archival sample, a fresh sample, or a frozen sample. The expression level of the one or more genes can be determined qualitatively and/or quantitatively based on any suitable criterion known in the art, including, but not limited to, the measurement of DNA, mRNA, cDNA, proteins, protein fragments, and/or gene copy number levels in an individual. Methodologies for measuring such biomarkers are known in the art and understood by the skilled artisan, including, but not limited to, whole genome sequencing, polymerase chain reaction (PCR) including quantitative real time PCR (qRT-PCR) and other amplification type detection methods, such as, for example, branched DNA, SISBA, TMA and the like, RNASeq, microarray analysis, gene expression profiling, whole-genome sequencing (WGS), and/or serial analysis of gene expression ("SAGE"), direct digital counting of nucleic acids (e.g., Nanostring nCounter), immunohistochemistry ("IHC"), Western blot analysis, immunoprecipitation, molecular binding assays, ELISA, ELIFA, fluorescence activated cell sorting ("FACS"), MassARRAY, proteomics, biochemical enzymatic activity assays, in situ hybridization (ISH), fluorescence in situ hybridization (FISH), Southern analysis, Northern analysis, as well as any one of the wide variety of assays that can be performed by protein, gene, and/or tissue array analysis. Typical protocols for evaluating the status of genes and gene products are found, for example, in Ausubel et al. eds. (Current Protocols In Molecular Biology, 1995), Units 2 (Northern Blotting), 4 (Southern Blotting), 15 (Immunoblotting) and 18 (PCR Analysis).

Multiplexed immunoassays such as those available from Rules Based Medicine or Meso Scale Discovery ("MSD") may also be used.

[0180] In some instances of any of the preceding methods and assays, the expression level of a biomarker may be a nucleic acid expression level (e.g., a DNA expression level or an RNA expression level (e.g., an mRNA expression level)). Any suitable method of determining a nucleic acid expression level may be used. In some instances, the nucleic acid expression level is determined using direct digital counting of nucleic acids (e.g., Nanostring nCounter), RNAseq, RT-qPCR, qPCR, multiplex qPCR or RT-qPCR, microarray analysis, SAGE, MassARRAY technique, ISH, or a combination thereof.

[0181] Methods for the evaluation of mRNAs in cells are well known and include, for example, serial analysis of gene expression (SAGE), whole genome sequencing (WGS), hybridization assays using complementary DNA probes (such as in situ hybridization using labeled riboprobes specific for the one or more genes, Northern blot and related techniques) and various nucleic acid amplification assays (such as RT-PCR (e.g., qRT-PCR) using complementary primers specific for one or more of the genes, and other amplification type detection methods, such as, for example, branched DNA, SISBA, TMA and the like). In addition, such methods can include one or more steps that allow one to determine the levels of target mRNA in a biological sample (e.g., by simultaneously examining the levels a comparative control mRNA sequence of a "housekeeping" gene such as an actin family member). Optionally, the sequence of the amplified target cDNA can be determined. Optional methods include protocols which examine or detect mRNAs, such as target mRNAs, in a tissue or cell sample by microarray technologies. Using nucleic acid microarrays, test and control mRNA samples from test and control tissue samples are reverse transcribed and labeled to generate cDNA probes. The probes are then hybridized to an array of nucleic acids immobilized on a solid support. The array is configured such that the sequence and position of each member of the array is known. For example, a selection of genes whose expression correlates with increased or reduced clinical benefit of treatment comprising an immunotherapy and a suppressive stromal antagonist may be arrayed on a solid support. Hybridization of a labeled probe with a particular array member indicates that the sample from which the probe was derived expresses that gene.

[0182] In other instances of any of the preceding methods, the expression level of a biomarker may be a protein expression level. In certain instances, the method comprises contacting the sample with antibodies that specifically bind to a biomarker described herein under conditions permissive for binding of the biomarker, and detecting whether a complex is formed between the antibodies and biomarker.

[0183] Any method of measuring protein expression levels known in the art or provided herein may be used. For example, in some instances, a protein expression level of a biomarker is determined using a method selected from, but not limited to western blot, enzyme-linked immunosorbent assay (ELISA), immunoprecipitation, immunohistochemistry (IHC), flow cytometry (e.g., fluorescence-activated cell sorting (FACS™)), immunofluorescence, radioimmunoassay, dot blotting, immunodetection methods, HPLC, surface plasmon resonance, optical spectroscopy, mass spectrometry, liquid chromatography-mass spectrometry (LC-MS), nephelometry, aptamer technology, and HPLC.

[0184] In certain instances, the presence and/or expression level/amount of a biomarker protein in a sample is examined using IHC and staining protocols. IHC staining of tissue sections has been shown to be a reliable method of determining or detecting the presence of proteins in a sample. In some instances of any of the methods, assays and/or kits, the biomarker is one or more of the protein expression products of the genes set forth in Table 1. In one instance, an expression level of biomarker is determined using a method comprising: (a) performing IHC analysis of a sample (such as a synovial tissue sample obtained from an individual) with an antibody; and (b) determining expression level of a biomarker in the sample. In some instances, IHC staining

intensity is determined relative to a reference. In some instances, the reference is a reference value. In some instances, the reference is a reference sample (e.g., a control cell line staining sample, a tissue sample from non-RA affected individual, or a synovial tissue sample that is determined to be negative for the biomarker of interest).

[0185] IHC may be performed in combination with additional techniques such as morphological staining and/or in situ hybridization (e.g., ISH). Two general methods of IHC are available; direct and indirect assays. According to the first assay, binding of antibody to the target antigen is determined directly. This direct assay uses a labeled reagent, such as a fluorescent tag or an enzyme-labeled primary antibody, which can be visualized without further antibody interaction. In a typical indirect assay, unconjugated primary antibody binds to the antigen and then a labeled secondary antibody binds to the primary antibody. Where the secondary antibody is conjugated to an enzymatic label, a chromogenic or fluorogenic substrate is added to provide visualization of the antigen. Signal amplification occurs because several secondary antibodies may react with different epitopes on the primary antibody.

[0186] The primary and/or secondary antibody used for IHC typically will be labeled with a detectable moiety. Numerous labels are available which can be generally grouped into the following categories: (a) radioisotopes, such as .sup.35S, .sup.14C, .sup.125I, .sup.3H, and .sup.131I; (b) colloidal gold particles; (c) fluorescent labels including, but are not limited to, rare earth chelates (europium chelates), Texas Red, rhodamine, fluorescein, dansyl, lissamine, umbelliferone, phycocrytherin, phycocyanin, or commercially-available fluorophores such as SPECTRUM ORANGE7 and SPECTRUM GREEN7 and/or derivatives of any one or more of the above; (d) various enzyme-substrate labels are available and U.S. Pat. No. 4,275,149 provides a review of some of these. Examples of enzymatic labels include luciferases (e.g., firefly luciferase and bacterial luciferase; see, e.g., U.S. Pat. No. 4,737,456), luciferin, 2,3-dihydrophthalazinediones, malate dehydrogenase, urease, peroxidase such as horseradish peroxidase (HRPO), alkaline phosphatase, β-galactosidase, glucoamylase, lysozyme, saccharide oxidases (e.g., glucose oxidase, galactose oxidase, and glucose-6-phosphate dehydrogenase), heterocyclic oxidases (such as uricase and xanthine oxidase), lactoperoxidase, microperoxidase, and the like.

[0187] Examples of enzyme-substrate combinations include, for example, horseradish peroxidase (HRPO) with hydrogen peroxidase as a substrate; alkaline phosphatase (AP) with para-Nitrophenyl phosphate as chromogenic substrate; and β-D-galactosidase (β-D-Gal) with a chromogenic substrate (e.g., p-nitrophenyl-β-D-galactosidase) or fluorogenic substrate (e.g., 4-methylumbelliferyl-β-D-galactosidase). For a general review of these, see, for example, U.S. Pat. Nos. 4,275,149 and 4,318,980.

[0188] Specimens may be prepared, for example, manually, or using an automated staining instrument (e.g., a Ventana BenchMark XT or Benchmark ULTRA instrument). Specimens thus prepared may be mounted and coverslipped. Slide evaluation is then determined, for example, using a microscope, and staining intensity criteria, routinely used in the art, may be employed. In some instances, the presence of a biomarker is detected by IHC in >0% of the sample, in at least 1% of the sample, in at least 5% of the sample, in at least 10% of the sample, in at least 15% of the sample, in at least 15% of the sample, in at least 20% of the sample, in at least 25% of the sample, in at least 30% of the sample, in at least 35% of the sample, in at least 40% of the sample, in at least 45% of the sample, in at least 50% of the sample, in at least 55% of the sample, in at least 60% of the sample, in at least 65% of the sample, in at least 70% of the sample, in at least 75% of the sample, in at least 80% of the sample, in at least 85% of the sample, in at least 90% of the sample, in at least 95% of the sample, or more. Samples may be scored using any method known in the art, for example, by a pathologist or automated image analysis.

[0189] In some instances of any of the methods and assays, the biomarker is detected by immunohistochemistry using a diagnostic antibody (e.g., a primary diagnostic antibody). In some instances, the diagnostic antibody specifically binds human antigen. In some instances, the

diagnostic antibody is a non-human antibody. In some instances, the diagnostic antibody is a rat, mouse, or rabbit antibody. In some instances, the diagnostic antibody is a rabbit antibody. In some instances, the diagnostic antibody is a monoclonal antibody. In some instances, the diagnostic antibody is directly labeled. In other instances, the diagnostic antibody is indirectly labeled (e.g., by a secondary antibody).

[0190] In some instances of any of the preceding methods and assays the sample is obtained from the individual prior to (e.g., minutes, hours, days, weeks (e.g., 1, 2, 3, 4, 5, 6, or 7 weeks), months, or years prior to) administration of an RA therapy. In some instances of any of the preceding methods, the sample from the individual is obtained about 2 to about 10 weeks (e.g., 2, 3, 4, 5, 6, 7, 8, 9, or 10 weeks) following administration of an RA therapy. In some instances, the sample from the individual is obtained about 4 to about 6 weeks following administration of an RA therapy.

[0191] In some instances of any of the preceding methods and assays, the expression level or number of a biomarker is detected in a synovial tissue sample, a synovial fluid sample, a primary or cultured cells or cell line, a cell supernatant, a cell lysate, platelets, serum, plasma, vitreous fluid, lymph fluid, follicular fluid, seminal fluid, amniotic fluid, milk, whole blood, blood-derived cells, urine, cerebro-spinal fluid, saliva, sputum, tears, perspiration, mucus, tissue culture medium, tissue extracts such as homogenized tissue, cellular extracts, or any combination thereof. In some instances, the sample is a tissue sample (e.g., a synovial tissue sample), a cell sample, a whole blood sample, a plasma sample, a serum sample, or a combination thereof. In some instances, the sample is a synovial tissue sample wherein, the synovial tissue sample includes resident cells, synovium-infiltrating immune cells, or a combination thereof. In some instances, the synovial tissue sample is a formalin-fixed and paraffin-embedded (FFPE) sample, an archival sample, a fresh sample, or a frozen sample.

[0192] For example, in some instances for any of the preceding methods and assays, the expression level of a biomarker in a sample (e.g., a synovial tissue sample) is detected in synovium-infiltrating immune cells, synovial cells, PBMCs, or combinations thereof using known techniques (e.g., flow cytometry or IHC). Synovium-infiltrating immune cells include, but are not limited to T lymphocytes, B lymphocytes (including plasma cells), or other bone marrow-lineage cells including granulocytes (e.g., mast cells), monocytes, macrophages, dendritic cells, and natural killer (NK) cells. In some instances, the staining for a biomarker is detected as membrane staining, cytoplasmic staining, or combinations thereof. In other instances, the absence of a biomarker is detected as absent or no staining in the sample, relative to a reference sample.

[0193] In certain instances, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is a single sample or combined multiple samples from the same patient or individual that are obtained at one or more different time points than when the test sample is obtained. For example, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is obtained at an earlier time point from the same patient or individual than when the test sample is obtained. Such reference sample, reference cell, reference tissue, control sample, control cell, or control tissue may be useful if the reference sample is obtained during initial diagnosis of RA and the test sample is later obtained when the disease has progressed.

[0194] In certain instances, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is a combined multiple samples from one or more healthy individuals who are not the patient. In certain instances, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is a combined multiple samples from one or more individuals with a disease or disorder (e.g., RA) who are not the patient or individual. In certain instances, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is pooled RNA samples from normal tissues or pooled plasma or serum samples from one or more individuals who are not the patient. In certain instances, a reference sample, reference cell, reference tissue, control sample, control cell, or control tissue is pooled RNA samples from

synovial tissues, synovial fluids, or pooled plasma or serum samples from one or more individuals with a disease or disorder (e.g., RA) who are not the patient.

B. Therapeutic Methods

[0195] The present invention also provides methods for treating an individual having RA. Accordingly, in some instances, the methods of the invention include administering to the individual an RA therapeutic agent. Any of the RA therapeutic agents described herein, or known in the art may be used in connection with the methods.

[0196] Provided herein is a method of treating an individual having RA that includes (i) determining the expression level of one or more of the genes set forth in Table 1 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, or 46 genes set forth in Table 1) in a sample from the individual, wherein the expression level of the one or more of the genes set forth in Table 1 is determined to be changed relative to a reference expression level, and (ii) administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual based on the expression level of the one or more genes determined in step (i). In some instances, the change is an increase. In other instances, the change is a decrease.

[0197] Provided herein is a method of treating an individual having RA that includes (i) determining the expression level of one or more of the genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) in a sample from the individual, wherein the expression level of the one or more of the genes set forth in Table 2 is determined to be increased (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level, and (ii) administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual based on the expression level of the one or more genes determined in step (i). In certain instances, the increased expression level of one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0198] Provided herein is a method of treating an individual having RA that includes (i) determining the expression level of one or more of the genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) in a sample from the individual, wherein the expression level of the one or more of the genes set forth in Table 3 is determined to be decreased (e.g., a decreased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to a reference expression level, and (ii) administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual based on the expression level of the one or more genes determined in step (i). In certain instances, the decreased expression level of the one or more genes set forth in Table 3 (e.g., 1, 2, 3,

4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the decreased expression level of the one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0199] Also provided is a method of treating an individual having RA that includes administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual, wherein the individual has been identified as one who is more likely to exhibit disease progression by one or more of the predictive diagnostic methods described in Section II-A, above.

[0200] The invention also provides a method of treating an individual having RA that includes (i) obtaining a sample from the individual, (ii) performing a gene expression assay on the sample and detecting (a) an increased expression level (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) in the sample and/or (b) a decreased level (e.g., a decreased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) in the sample relative to a reference expression level, (iii) identifying the individual as having an increased likelihood of benefitting from a therapeutic agent other than, or in addition to, a DMARD, and (iv) administering to the individual a therapeutic agent other than, or in addition to, a DMARD. In some instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes. In certain instances, the decreased expression level of the one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the decreased expression level of the one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-

fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0201] The invention also provides a method of treating RA in an individual identified as having an increased expression level (e.g., an increased expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) relative to a reference expression level of the one or more genes, wherein the method includes administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual. In certain instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes, and the method includes administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual. In some instances, the increased expression level of the one or more genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 genes set forth in Table 2) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes, and the method includes administering an effective amount of an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD) to the individual.

[0202] In another instance, the invention provides a method of treating RA in an individual having been identified as having an expression level in a sample from the individual of one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) that is at or above a reference expression level of the one or more genes (e.g., above or an increase in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater), the method including administering to the individual an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD). In certain instances, the increased expression level of one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the increased expression level of one or more genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-

fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0203] In another instance, the invention provides a method of treating RA in an individual having been identified as having an expression level in a sample from the individual of at least two, at least three, at least four, at least five, at least six, or all seven of the following genes in a sample from the individual: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, that is at or above a reference expression level (e.g., above or an increase in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of the at least two, at least three, at least four, at least five, at least six, or all seven genes, the method including administering to the individual an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD). In certain instances, the increased expression level of at least two, at least three, at least four, at least five, at least six, or all seven genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A is an increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of at least two, at least three, at least four, at least five, at least six, or all seven genes. In some instances, the increased expression level of at least two, at least three, at least four, at least five, at least six, or all seven genes selected from CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A is an increase of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the at least two, at least three, at least four, at least five, at least six, or all seven genes.

[0204] In another instance, the invention provides a method of treating RA in an individual having been identified as having an expression level in a sample from the individual of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6) that is at or below a reference expression level (e.g., below or a decrease in the expression level of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) of the one or more genes, the method including administering to the individual an RA therapy (e.g., a therapeutic agent other than, or in addition to, a DMARD). In certain instances, the decreased expression level of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference expression level of the one or more genes. In some instances, the decreased expression level of one or more genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6 genes set forth in Table 3) is a decrease of at least about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, or about 1,000-fold or greater relative to a reference expression level of the one or more genes.

[0205] In any of the therapeutic methods described above, the methods may further include determining one or more clinical covariates (e.g., baseline RF titer, disease duration, DAS28-ESR, DAS28-CRP, baseline pathotype, and 12 max USST and USPD scores) of the individual.

[0206] In any of the therapeutic methods described above, the reference expression level may be a reference expression level in a reference population of individuals having RA who have not been

previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who exhibited disease progression and a second subset of individuals who did not exhibit disease progression. In some instances, the reference expression level significantly separates each of the first and second subsets of individuals based on a significant difference in the expression level of the one or more genes set forth in Table 1, Table 2, or Table 3 in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals exhibited disease progression and the second subset of individuals did not exhibit disease progression after about 12 months.

[0207] In any of the therapeutic methods described above in which the expression level of more than one biomarker selected from the genes set forth in Table 1, Table 2, or Table 3 is determined in a sample from an individual and compared to a reference expression level (e.g. a pre-assigned expression level of the one or more genes set forth in Table 1, Table 2, or Table 3), it is to be understood that, in some instances, the expression level of each individual biomarker in the sample is compared to a reference expression level for each individual biomarker. For example, if the expression level of CD180 and CXCL1 are determined in a sample from an individual and compared to reference expression levels for CD180 and CXCL1, in some instances, the expression level of CD180 in the sample from the individual is compared to the reference expression level for CD180, and the expression level of CXCL1 in the sample from the individual is compared to the reference expression level for CXCL1. In other instances, an expression level for more than one gene of interest may be determined by aggregation methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the median or mean of all the expression levels of the genes of interest. Before aggregation, the expression level of each gene of interest may be normalized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, normalized to the expression level of one or more housekeeping genes, or normalized to a total library size, or normalized to the median or mean expression level value across all genes measured. In some instances, before aggregation across multiple genes of interest, the normalized expression level of each gene of interest may be standardized by using statistical methods known to one skilled in the art and also disclosed herein, including, for example, by calculating the Z-score of the normalized expression level of each gene of interest.

[0208] In any of the therapeutic methods described above in which the expression level of one or more biomarkers, selected from the biomarkers set forth in Table 1, Table 2, or Table 3, is determined in a sample from an individual and compared to a reference expression level (e.g., a pre-assigned expression level of the one or more genes set forth in Table 1, Table 2, or Table 3), it is to be understood that, in some instances, the expression level of one or more genes may be an average of the expression level of the one or more genes. In some instances, the expression level of the one or more genes may be a median of the expression level of the one or more genes. In some instances, the expression level the one or more genes may be normalized, e.g., to a reference gene, e.g., a housekeeping gene. In some instances, the reference gene is ACTB, GAPDH, GUSB, HPRT1, PGK1, RPL19, TUBB, or TMEM55B. In some instances, the expression level of the one or more genes may be an average of a normalized expression level of the one or more genes. In some instances, the expression level of the one or more genes may be a median of a normalized expression level of the one or more genes.

[0209] The present invention provides methods that may be used to treat an individual having RA with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) that involves (i) obtaining a sample form the individual, (ii) performing a gene expression assay on the sample and determining a myeloid eigengene score equal to or increased relative to a reference myeloid eigengene score (e.g., an increased myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater), (iii) identifying the individual as having an increased likelihood of benefitting from an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate), and (iv) administering to the individual an RA therapy (e.g., RA

therapy that includes a DMARD, such as methotrexate). In some instances, an increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an increased myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, the methods may further include, prior to the identifying the individual as having an increased likelihood of benefitting from an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate), performing a gene expression assay on the sample and determining a lymphoid eigengene score equal to or increased relative to a reference lymphoid eigengene score (e.g., an increased lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an increased lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0210] The present invention provides methods that may be used to treat an individual having RA with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) that involves (i) determining the myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample is determined to be at or above a reference myeloid eigengene score (e.g., above or an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater), and (ii) administering an effective amount of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) to the individual based on the myeloid eigengene score of step (i). In some instances, an elevated or increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an elevated myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, the methods may further involve, prior to the administering of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate),

determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score from the sample is determined to be at or above a reference lymphoid eigengene score (e.g., above or an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an elevated or increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an elevated lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0211] Also provided is a method of treating an individual having RA that includes administering an effective amount of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) to the individual, wherein the individual has been identified as one who is more likely to benefit from an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) by one or more of the predictive diagnostic methods described in Section II-A, above. Accordingly, in some instances, the individual has been identified as one who is more likely to benefit from an RA treatment (e.g., RA therapy that includes a DMARD, such as methotrexate) based on a myeloid eigengene score from a sample from the individual that has been determined to be at or above a reference myeloid eigengene score (e.g., above or an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an elevated or increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an elevated myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, prior to administering an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) to the individual, the individual has been identified as one who is more likely to benefit from an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) based further on a lymphoid eigengene score from a sample from the individual that has been determined to be at or above a reference lymphoid eigengene score (e.g., above or an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an elevated or increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an elevated lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold,

about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0212] Also provided is a method of treating an individual having RA that includes administering an effective amount of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) to the individual, wherein the individual has been identified as having a myeloid eigengene score from a sample from the individual that is at or above a reference myeloid eigengene score by one or more of the predictive diagnostic methods described in Section II-A, above. Accordingly, in some instances, the individual has been identified as having a myeloid eigengene score from a sample from the individual that is at or above a reference myeloid eigengene score (e.g., above or an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an elevated or increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an elevated myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, prior to administering an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) to the individual, the individual has been identified as having a lymphoid eigengene score from a sample from the individual that is at or above a reference lymphoid eigengene score (e.g., above or an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater). In some instances, an elevated or increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an elevated lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0213] In particular instances, the methods provided herein may be used to optimize therapeutic efficacy of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate), the method including monitoring a myeloid eigengene score from a sample from the individual during treatment (e.g., over a treatment period) with the RA therapy. Monitoring may include, for example, obtaining and comparing myeloid eigengene scores from samples from the individual collected at time intervals before and/or after administration of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, a myeloid eigengene score may

be obtained from a sample from the individual that was collected at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, or 24 hours; about 1, 2, 3, 4, 5, 6, 7 days; about 1, 2, 3, or 4 weeks; or about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 months before administration of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, a myeloid eigengene score may be obtained from a sample from the individual that was collected at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, or 24 hours; about 1, 2, 3, 4, 5, 6, 7 days; about 1, 2, 3, or 4 weeks; or about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 months after administration of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). The myeloid eigengene scores from samples from the individual collected before and/or after the administration of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) may be compared, wherein an increase in a myeloid eigengene score (e.g., an increase in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) from a sample from the individual collected after treatment relative to a myeloid eigengene score from a sample collected before treatment may indicate a low level of therapeutic efficacy of the RA therapy that was administered, and wherein a decrease in a myeloid eigengene score (e.g., a decrease in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) from a sample from the individual collected after treatment relative to a myeloid eigengene score from a sample collected before treatment may indicate therapeutic efficacy of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) that was administered. In some instances, an elevated or increased myeloid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, an elevated myeloid eigengene score refers to an overall increase of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score. In some instances, a reduced or decreased myeloid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, a reduced myeloid eigengene score refers to an overall decrease of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score.

[0214] In some instances, the reference myeloid eigengene score may be obtained from the individual prior to treatment with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, the method includes monitoring the myeloid eigengene score from a sample from the individual relative to a pre-treatment myeloid eigengene score during treatment (e.g., over a treatment period) with the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate).

[0215] In particular instances, the methods provided herein may further include monitoring a

lymphoid eigengene score from a sample from the individual during treatment (e.g., over a treatment period) with the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). Monitoring may include, for example, obtaining and comparing lymphoid eigengene scores from samples from the individual collected at time intervals before and/or after administration of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, a lymphoid eigengene score may be obtained from a sample from the individual that was collected at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, or 24 hours; about 1, 2, 3, 4, 5, 6, 7 days; about 1, 2, 3, or 4 weeks; or about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 months before administration of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, a lymphoid eigengene score may be obtained from a sample from the individual that was collected at least about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, or 24 hours; about 1, 2, 3, 4, 5, 6, 7 days; about 1, 2, 3, or 4 weeks; or about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 months after administration of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). The lymphoid eigengene scores from samples from the individual collected before and/or after the administration of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) may be compared, wherein an increase in lymphoid eigengene score (e.g., an increase in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) from a sample from the individual collected after treatment relative to a lymphoid eigengene score from a sample collected before treatment may indicate a low level of therapeutic efficacy of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) that was administered, and wherein a decrease (e.g., a decrease in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) in lymphoid eigengene score from a sample from the individual collected after treatment relative to a lymphoid eigengene score from a sample collected before treatment may indicate therapeutic efficacy of the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) that was administered. In some instances, the reference lymphoid eigengene score may be obtained from the individual prior to treatment with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, the method includes monitoring the lymphoid eigengene score from a sample form the individual relative to a pre-treatment lymphoid eigengene score during treatment (e.g., over a treatment period) with the RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, an elevated or increased lymphoid eigengene score, refers to an overall increase of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, an elevated lymphoid eigengene score refers to an overall increase of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score. In some instances, a reduced or decreased lymphoid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, a reduced lymphoid eigengene score refers to an overall decrease of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold,

about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0216] In particular instances, the methods may include (i) determining a first myeloid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (ii) determining a second myeloid eigengene score from a sample from the individual at a second time point; and (iii) comparing the first myeloid eigengene score with the second myeloid eigengene score, wherein a decrease in the second myeloid eigengene score (e.g., a decrease in the myeloid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) relative to the first myeloid eigengene score below a reference eigengene score identifies the individual as one who is likely to respond to treatment with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, the method may further include administering and additional dose of an RA therapeutic agent to the individual. In some instances, a reduced or decreased myeloid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference myeloid eigengene score. In some instances, a reduced myeloid eigengene score refers to an overall decrease of the myeloid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference myeloid eigengene score.

[0217] In some instances, the method may further include prior to the administering of an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate) (i) determining a first lymphoid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (ii) determining a second lymphoid eigengene score from a sample from the individual at a second time point; and (iii) comparing the first lymphoid eigengene score with the second lymphoid eigengene score, wherein a decrease (e.g., a decrease in the lymphoid eigengene score of about 10%, 20%, 30%, 40%, 50%, 60%, 70%, 80%, 90%, 95%, 96%, 97%, 98%, or 99% or greater) in the second lymphoid eigengene score relative to the first lymphoid eigengene score identifies the individual as one who is likely to respond to treatment with an RA therapy (e.g., RA therapy that includes a DMARD, such as methotrexate). In some instances, a reduced or decreased lymphoid eigengene score, refers to an overall decrease of at least about 1.1×, 1.2×, 1.3×, 1.4×, 1.5×, 1.6×, 1.7×, 1.8×, 1.9×, 2×, 2.1×, 2.2×, 2.3×, 2.4×, 2.5×, 2.6×, 2.7×, 2.8×, 2.9×, 3×, 3.5×, 4×, 4.5×, 5×, 6×, 7×, 8×, 9×, 10×, 15×, 20×, 30×, 40×, 50×, 100×, 500×, or 1000× relative to a reference lymphoid eigengene score. In some instances, a reduced lymphoid eigengene score refers to an overall decrease of the lymphoid eigengene score of greater than about 1.1-fold, about 1.2-fold, about 1.3-fold, about 1.4-fold, about 1.5-fold, about 1.6-fold, about 1.7-fold, about 1.8-fold, about 1.9-fold, about 2-fold, about 2.1-fold, about 2.2-fold, about 2.3-fold, about 2.4-fold, about 2.5-fold, about 2.6-fold, about 2.7-fold, about 2.8-fold, about 2.9-fold, about 3-fold, about 3.5-fold, about 4-fold, about 4.5-fold, about 5-fold, about 6-fold, about 7-fold, about 8-fold, about 9-fold, about 10-fold, about 15-fold, about 20-fold, about 30-fold, about 40-fold, about 50-fold, about 100-fold, about 500-fold, about 1,000-fold or greater relative to a reference lymphoid eigengene score.

[0218] In some instances of any of the therapeutic methods described above, the decrease in the

second myeloid eigengene score relative to the first myeloid eigengene score is between about 1.25-fold to about 10-fold. In some instances, the decrease is between about 1.25-fold to about 7.5-fold. In some instances, the decrease is between about 1.25-fold to about 5-fold. In some instances, the decrease is between about 1.25-fold to about 2-fold. In some instances, the decrease is between about 1.25-fold to about 1.5 fold. In some instances, the decrease is at least about 1.25-fold.

[0219] In some instances of any of the therapeutic methods described above, the decrease in the second lymphoid eigengene score relative to the first lymphoid eigengene score is between about 1.25-fold to about 10-fold. In some instances, the decrease is between about 1.25-fold to about 7.5-fold. In some instances, the decrease is between about 1.25-fold to about 5-fold. In some instances, the decrease is between about 1.25-fold to about 2-fold. In some instances, the decrease is between about 1.25-fold to about 1.5 fold. In some instances, the decrease is at least about 1.25-fold.

[0220] In some instances of any of the therapeutic methods described above, the reference myeloid eigengene score may be a reference myeloid eigengene score in a reference population of individuals having RA who have been previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who exhibited responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy. In some instances, the reference myeloid eigengene score level significantly separates each of the first and second subsets of individuals based on a significant difference in the myeloid eigengene score in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.

[0221] In some instances of any of the therapeutic methods described above, the reference lymphoid eigengene score may be a reference lymphoid eigengene score in a reference population of individuals having RA who have been previously treated with a DMARD, the population of individuals consisting of a first subset of individuals who exhibited responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy. In some instances, the reference lymphoid eigengene score level significantly separates each of the first and second subsets of individuals based on a significant difference in the lymphoid eigengene score in the first subset of individuals compared to that of the second subset of individuals. In some instances, the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.

[0222] In some instances of any of the preceding methods, the RA therapeutic agent may be a DMARD.

[0223] In some instances of any of the preceding methods, an RA therapeutic agent other than a DMARD may be administered.

[0224] In some instances of any of the preceding methods, a DMARD may be administered in conjunction with a therapeutic agent other than a DMARD.

[0225] In some instances of any of the preceding methods, a DMARD may include, but is not limited to, methotrexate, hydroxychloroquine, sulfasalazine, lefunomide, azathioprine, cyclophosphamide, cyclosporine, and mycophenolate mofetil.

[0226] In some instances of any of the preceding methods, the therapeutic agent other than a DMARD may include, but is not limited to, a B cell antagonist, a JAK antagonist, a TNF antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor antagonist, an IL-6 receptor antagonist, or a combination thereof.

[0227] In some instances of any of the preceding methods, the B cell antagonist may be, but is not limited to, rituximab (e.g., RITUXAN®).

[0228] In some instances of any of the preceding methods, the JAK antagonist may be, but is not limited to, tofacitinib (e.g. XELJANZ®).

[0229] In some instances of any of the preceding methods, the TNF antagonist may be, but is not limited to, adalimumab, golimumab, infliximab, certolizumab pegol, or a combination thereof.
[0230] In some instances of any of the preceding methods, the TNF decoy receptor may be, but is not limited to, etanercept.
[0231] In some instances of any of the preceding methods, the T cell costimulatory signal antagonist may be, but is not limited to, abatacept.
[0232] In some instances of any of the preceding methods, the IL-1 receptor antagonist may be, but is not limited to, anakinra.
[0233] In some instances of any of the preceding methods, the IL-6 receptor antagonist may be, but is not limited to, tocilizumab (e.g., ACTEMRA®/RoACTEMRA®).
[0234] In some instances of any of the preceding methods, the RA therapeutic agents utilized in the methods described herein can be administered, for example, orally, subcutaneously, intramuscularly, intravenously, intradermally, percutaneously, intraarterially, intraperitoneally, intralesionally, intracranially, intraarticularly, intraprostatically, intrapleurally, intratracheally, intrathecally, intranasally, intravaginally, intrarectally, topically, intratumorally, peritoneally, subconjunctivally, intravesicularly, mucosally, intrapericardially, intraumbilically, intraocularly, intraorbitally, intravitreally (e.g., by intravitreal injection), by eye drop, topically, transdermally, parenterally, by inhalation, by injection, by implantation, by infusion, by continuous infusion, by localized perfusion bathing target cells directly, by catheter, by lavage, in creams, or in lipid compositions. In some instances of any of the preceding methods, the RA therapeutic agents utilized in the methods described herein can be administered orally. In some instances of any of the preceding methods, the RA therapeutic agents utilized in the methods described herein can be administered subcutaneously. The RA therapeutic agents utilized in the methods described herein can also be administered systemically or locally. The method of administration can vary depending on various factors (e.g., the RA therapeutic agents being administered and the severity of the condition, disease, or disorder (e.g., RA) being treated).

III. Compositions and Pharmaceutical Formulations

[0235] In one aspect, the invention is based, in part, on the discovery that biomarkers of the invention can be used to identify individuals having RA who may benefit from an RA therapy, for example, a DMARD (e.g., a DMARD), or a therapy including a biologic therapeutic agent (e.g., a biologic therapeutic agent), alone or in combination with a DMARD.
[0236] In some instances, the individual is less likely to respond to the DMARD alone. In another instance, the invention is based, in part, on the discovery that biomarkers of the invention can be used to monitor and/or assess treatment response for individuals having RA who are treated with RA therapies that include a DMARD. These agents, and combinations thereof, are useful for the treatment of RA, e.g., as part of any of the methods and uses described herein, for example, in Section II above. Any suitable DMARD can be used in the methods and uses described herein.

A. Pharmaceutical Formulations

[0237] Therapeutic formulations of the therapeutic agents, e.g., a DMARD and/or a biologic therapeutic agent, used in accordance with the present invention are prepared for storage by mixing the therapeutic agent having the desired degree of purity with optional pharmaceutically acceptable carriers, excipients, or stabilizers in the form of lyophilized formulations or aqueous solutions. For general information concerning formulations, see, e.g., Gilman et al. (eds.) *The Pharmacological Bases of Therapeutics,* 8th Ed., Pergamon Press, 1990; A. Gennaro (ed.), *Remington's Pharmaceutical Sciences,* 18th Edition, Mack Publishing Co., Pennsylvania, 1990; Avis et al. (eds.) *Pharmaceutical Dosage Forms: Parenteral Medications Dekker, New York,* 1993; Lieberman et al. (eds.) *Pharmaceutical Dosage Forms: Tablets Dekker, New York,* 1990; Lieberman et al. (eds.), *Pharmaceutical Dosage Forms: Disperse Systems Dekker, New York,* 1990; and Walters (ed.) *Dermatological and Transdermal Formulations* (Drugs and the Pharmaceutical Sciences), Vol 119, Marcel Dekker, 2002.

[0238] Acceptable carriers, excipients, or stabilizers are non-toxic to recipients at the dosages and concentrations employed, and include buffers such as phosphate, citrate, and other organic acids; antioxidants including ascorbic acid and methionine; preservatives (such as octadecyldimethylbenzyl ammonium chloride; hexamethonium chloride; benzalkonium chloride, benzethonium chloride; phenol, butyl or benzyl alcohol; alkyl parabens such as methyl or propyl paraben; catechol; resorcinol; cyclohexanol; 3-pentanol; and m-cresol); low molecular weight (less than about 10 residues) polypeptides; proteins, such as serum albumin, gelatin, or immunoglobulins; hydrophilic polymers such as polyvinylpyrrolidone; amino acids such as glycine, glutamine, asparagine, histidine, arginine, or lysine; monosaccharides, disaccharides, and other carbohydrates including glucose, mannose, or dextrins; chelating agents such as EDTA; sugars such as sucrose, mannitol, trehalose or sorbitol; salt-forming counter-ions such as sodium; metal complexes (e.g., Zn-protein complexes); and/or non-ionic surfactants such as TWEEN™, PLURONICS™, or polyethylene glycol (PEG).

[0239] The formulation herein may also contain more than one active compound, preferably those with complementary activities that do not adversely affect each other. The type and effective amounts of such medicaments depend, for example, on the amount and type of therapeutic agent (e.g., a DMARD and/or a biologic therapeutic agent) present in the formulation, and clinical parameters of the patients.

[0240] The active ingredients may also be entrapped in microcapsules prepared, for example, by coacervation techniques or by interfacial polymerization, for example, hydroxymethylcellulose or gelatin-microcapsules and poly-(methylmethacylate) microcapsules, respectively, in colloidal drug delivery systems (for example, liposomes, albumin microspheres, microemulsions, nanoparticles and nanocapsules) or in macroemulsions. Such techniques are disclosed in Remington's Pharmaceutical Sciences 16th edition, Osol, A. Ed., 1980.

[0241] Sustained-release preparations may be prepared. Suitable examples of sustained-release preparations include semi-permeable matrices of solid hydrophobic polymers containing the therapeutic agent, which matrices are in the form of shaped articles, e.g., films, or microcapsules. Examples of sustained-release matrices include polyesters, hydrogels (for example, poly(2-hydroxyethyl-methacrylate), or poly(vinylalcohol)), polylactides (U.S. Pat. No. 3,773,919), copolymers of L-glutamic acid and γ ethyl-L-glutamate, non-degradable ethylene-vinyl acetate, degradable lactic acid-glycolic acid copolymers such as the LUPRON DEPOT™ (injectable microspheres composed of lactic acid-glycolic acid copolymer and leuprolide acetate), and poly-D-(−)-3-hydroxybutyric acid.

[0242] The formulations to be used for in vivo administration must be sterile. This is readily accomplished by filtration through sterile filtration membranes.

IV. Articles of Manufacture and Kits

[0243] In another aspect of the invention, a kit or an article of manufacture containing materials useful for the treatment, prevention, diagnosis, and/or monitoring of individuals is provided.

[0244] In some instances, such kits or articles of manufacture can be used to identify an individual having RA who may benefit from an RA therapy that includes a therapeutic agent other than or in addition to a DMARD (including, but not limited to, a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD). Such articles of manufacture or kits may include (i) reagent(s) for determining the expression level of one or more genes set forth in Table 1, or any combination thereof (e.g., any combination set forth in any one of Tables 2 and 3) in a sample from the individual, and, optionally, (ii) instruction(s) for using the reagent(s) to identify an individual having RA who may benefit from treatment with an RA therapy that includes a therapeutic agent other than, or in addition to, a DMARD. In additional instances, the articles of manufacture or kits may include (i) reagent(s) for determining the expression level of one or more genes set forth in Table 1, or any combination thereof (e.g., any combination set forth in any one of Tables 2 and 3) in a sample from the individual, and, optionally, (ii) instruction(s) for using the reagents to monitor

and/or assess the response of an individual having RA to treatment with an RA therapy that includes a therapeutic agent other than, or in addition to, a DMARD (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD).

[0245] In some instances, such kits or articles of manufacture can be used to identify an individual having RA who may benefit from an RA therapy that includes a therapeutic agent other than, or in addition to, a DMARD (including, but not limited to, a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD). Such articles of manufacture or kits may include (i) reagent(s) for determining the expression level of one or more genes set forth in Table 1, or any combination thereof (e.g., any combination set forth in any one of Tables 2 and 3) in a sample from the individual and (ii) instruction(s) for using the reagent(s) to identify an individual having RA who may benefit from treatment with an RA therapy that includes a therapeutic agent other than, or in addition to, a DMARD. In additional instances, the articles of manufacture or kits may include (i) reagent(s) for determining the expression level of one or more genes set forth in Table 1, or any combination thereof (e.g., any combination set forth in any one of Tables 2 and 3) in a sample from the individual and (ii) instruction(s) for using the reagents to monitor and/or assess the response of an individual having RA to treatment with an RA therapy that includes a therapeutic agent other than, or in addition to, a DMARD (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD).

[0246] Provided herein is a kit or an article of manufacture for identifying an individual having RA who may benefit from treatment with a therapeutic agent other than, or in addition to, a DMARD (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD), the kit or article of manufacture including: (i) reagents for determining the expression level of one or more of the genes set forth in Table 1 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, or 46 of the genes set forth in Table 1) in a sample from the individual, and, optionally, (ii) instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0247] For example, provided herein is a kit or an article of manufacture for identifying an individual having RA who may benefit from treatment with an RA therapy (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD), the kit or article of manufacture including: (i) reagents for determining the expression level of one or more of the following genes in a sample from the individual: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A), and, optionally, (ii) instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0248] In any of the preceding instances, the kit may include reagents for determining the expression level of one or more (e.g., 1, 2, 3, 4, 5, 6, or 7) of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In some embodiments, the kit includes reagents for determining the expression level of at least two, at least three, at least four, at least five, at least six, or all seven of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In some embodiments, the kit includes reagents for includes determining the expression level of two of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 4. In some embodiments, the kit includes reagents for includes determining the expression level of three of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 5. In some embodiments, the kit includes reagents for includes determining the expression level of four of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 6. In some embodiments, the kit includes reagents for

includes determining the expression level of five of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 7. In some embodiments, the kit includes reagents for includes determining the expression level of six of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 8. In some embodiments, the kit includes reagents for involves determining the expression level of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, and, optionally, (ii) instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0249] In some instances, such kits or articles of manufacture can be used to identify an individual having RA who may benefit from an RA therapy that includes a DMARD (including, but not limited to, a DMARD and/or a biologic therapeutic agent). Such articles of manufacture or kits may include (i) reagents for determining the myeloid eigengene, or myeloid eigengene and lymphoid eigengene score(s) in a sample from the individual, and, optionally, (ii) instructions for using the reagents to identify an individual having RA who may benefit from treatment with an RA therapy that includes a DMARD.

[0250] In additional instances, the articles of manufacture or kits may include (i) reagents for determining the myeloid eigengene, or myeloid eigengene and lymphoid eigengene score(s) in a sample from the individual, and, optionally, (ii) instructions for using the reagents to monitor and/or assess the response of an individual having RA to treatment with an RA therapy that includes a DMARD (e.g., a DMARD and/or a biologic therapeutic agent).

[0251] Provided herein is a kit or an article of manufacture for identifying an individual having RA who may benefit from treatment with an RA therapy (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD), the kit or article of manufacture including: (i) reagents for determining the expression level of one or more of the genes set forth in Table 1 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, or 46 of the genes set forth in Table 1) in a sample from the individual, and (ii) instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0252] For example, provided herein is a kit or an article of manufacture for identifying an individual having RA who may benefit from treatment with an RA therapy (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD), the kit or article of manufacture including: (i) reagents for determining the expression level of one or more (e.g., 1, 2, 3, 4, 5, 6, or 7) of the following genes in a sample from the individual: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, and (ii) instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0253] In any of the preceding instances, the kit may include reagents for determining the expression level of one or more (e.g., 1, 2, 3, 4, 5, 6, or 7) of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In some embodiments, the kit includes reagents for determining the expression level of at least two, at least three, at least four, at least five, at least six, or all seven of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A. In some embodiments, the kit includes reagents for includes determining the expression level of two of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 4. In some embodiments, the kit includes reagents for includes determining the expression level of three of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 5. In some embodiments, the kit includes reagents for includes determining the expression level of four of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the

exemplary combinations shown in Table 6. In some embodiments, the kit includes reagents for includes determining the expression level of five of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 7. In some embodiments, the kit includes reagents for includes determining the expression level of six of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 8. In some embodiments, the kit includes reagents for involves determining the expression level of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, and instructions for using the reagents to identify an individual having RA who may benefit from a treatment with an RA therapy comprising a therapeutic agent other than, or in addition to, a DMARD.

[0254] In some instances, such kits or articles of manufacture can be used to identify an individual having RA who may benefit from an RA therapy that includes a DMARD (including, but not limited to, a DMARD and/or a biologic therapeutic agent). Such articles of manufacture or kits may include (i) reagents for determining the myeloid eigengene, or myeloid eigengene and lymphoid eigengene score(s) in a sample from the individual, and (ii) instructions for using the reagents to identify an individual having RA who may benefit from treatment with an RA therapy that includes a DMARD.

[0255] In additional instances, the articles of manufacture or kits may include (i) reagents for determining the myeloid eigengene, or myeloid eigengene and lymphoid eigengene score(s) in a sample from the individual and, (ii) instructions for using the reagents to monitor and/or assess the response of an individual having RA to treatment with an RA therapy that includes a DMARD (e.g., a DMARD and/or a biologic therapeutic agent).

[0256] Provided herein is a kit or an article of manufacture for treating an individual with RA, the kit or article of manufacture including (i) a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD, and (ii) instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of one or more of the genes set forth in Table 2 (e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, or 40 of the genes set forth in Table 2) in a sample from the individual relative to a reference expression level.

[0257] Provided herein is a kit or an article of manufacture for treating an individual with RA, the kit or article of manufacture including (i) a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD, and (ii) instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of one or more of the following genes in a sample from the individual: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A (e.g., 1, 2, 3, 4, 5, 6, or 7 of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) in a sample from the individual relative to a reference expression level.

[0258] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of at least two, at least three, at least four, at least five, at least six, or all seven of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A in a sample from the individual relative to a reference expression level.

[0259] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of two of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and

UBASH3A, for example, any of the exemplary combinations shown in Table 4 in a sample from the individual relative to a reference expression level.

[0260] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of three of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 5 in a sample from the individual relative to a reference expression level.

[0261] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of four of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 6 in a sample from the individual relative to a reference expression level.

[0262] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of five of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 7 in a sample from the individual relative to a reference expression level.

[0263] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of six of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, for example, any of the exemplary combinations shown in Table 8 in a sample from the individual relative to a reference expression level.

[0264] In some embodiments, the kit includes instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and the DMARD to treat or delay the progression of RA in the individual, based on the determination of an increased expression level of CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A, in a sample from the individual relative to a reference expression level.

[0265] Provided herein is a kit or an article of manufacture for treating an individual with RA, the kit or article of manufacture including (i) an RA therapeutic agent (e.g., a biologic therapeutic agent alone, or a biologic therapeutic agent and a DMARD), and (ii) instructions for administering to an individual an effective amount of the biologic therapeutic agent alone, or the biologic therapeutic agent and a DMARD to treat or delay the progression of RA in the individual, based on the determination of a decreased expression level of one or more of the genes set forth in Table 3 (e.g., 1, 2, 3, 4, 5, or 6, of the genes set forth in Table 3) in a sample from the individual relative to a reference expression level.

[0266] Provided herein is a kit or an article of manufacture for treating an individual with RA, the kit or article of manufacture including (i) an RA therapeutic agent (e.g., a DMARD and/or a biologic therapeutic agent), and (ii) instructions for administering to an individual an effective amount of the RA therapeutic agent to treat or delay the progression of RA in the individual, based on the determination of an increased myeloid eigengene score in a sample from the individual relative to a reference myeloid eigengene score.

[0267] Provided herein is a kit or an article of manufacture for treating an individual with RA, the kit or article of manufacture including (i) an RA therapeutic agent (e.g., a DMARD and/or a biologic therapeutic agent), and (ii) instructions for administering to an individual an effective amount of the RA therapeutic agent to treat or delay the progression of RA in the individual, based on the determination of an increased myeloid eigengene score and an increased lymphoid

eigengene score in a sample from the individual relative to a reference myeloid eigengene score and a reference lymphoid eigengene score.

[0268] Any of the kits or articles of manufacture described may include a carrier means being compartmentalized to receive in close confinement one or more container means such as vials, tubes, and the like, each of the container means comprising one of the separate elements to be used in the method. Where the article of manufacture or kit utilizes nucleic acid hybridization to detect the target nucleic acid, the kit may also have containers containing nucleotide(s) for amplification of the target nucleic acid sequence and/or a container comprising a reporter-means, such as an enzymatic, florescent, or radioisotope label.

[0269] In some instances, the article of manufacture or kit includes the container described above and one or more other containers including materials desirable from a commercial and user standpoint, including buffers, diluents, filters, needles, syringes, and package inserts with instructions for use. A label may be present on the container to indicate that the composition is used for a specific application, and may also indicate directions for either in vivo or in vitro use, such as those described above. For example, the article of manufacture or kit may further include a container including a pharmaceutically-acceptable buffer, such as bacteriostatic water for injection (BWFI), phosphate-buffered saline, Ringer's solution, and dextrose solution.

[0270] The kits or articles of manufacture described herein may have a number of instances. In one instance, the kits or articles of manufacture includes a container, a label on said container, and a composition contained within said container, wherein the composition includes one or more polynucleotides that hybridize to a complement of a gene listed herein (e.g., a gene set forth in Table 1, or any combination of genes set forth in Tables 2 and 3) under stringent conditions, and the label on said container indicates that the composition can be used to evaluate the presence of a gene listed herein (e.g., a gene set forth in Table 1, or any combination of genes set forth in Tables 2 and 3) in a sample, and wherein the kit includes instructions for using the polynucleotide(s) for evaluating the presence of the gene RNA or DNA in a particular sample type.

[0271] For oligonucleotide-based articles of manufacture or kits, the article of manufacture or kit can include, for example: (1) an oligonucleotide, e.g., a detectably labeled oligonucleotide, which hybridizes to a nucleic acid sequence encoding a protein or (2) a pair of primers useful for amplifying a nucleic acid molecule. The article of manufacture or kit can also include, e.g., a buffering agent, a preservative, or a protein stabilizing agent. The article of manufacture or kit can further include components necessary for detecting the detectable label (e.g., an enzyme or a substrate). The article of manufacture or kit can also contain a control sample or a series of control samples that can be assayed and compared to the test sample. Each component of the article of manufacture or kit can be enclosed within an individual container and all of the various containers can be within a single package, along with instructions for interpreting the results of the assays performed using the kit.

V. Examples

[0272] The following are examples of the methods of the invention. It is understood that various other embodiments may be practiced, given the general description provided above.

Example 1. Association Between Immunohistochemistry (IHC) Pathotypes and Gene Expression Levels of Individuals Having Rheumatoid Arthritis (RA)

[0273] Ultrasound-guided synovial biopsies were collected from a cohort of 129 individuals enrolled in the Pathobiology of Early Arthritis Cohort (PEAC), a treatment-naïve, early rheumatoid arthritis (RA) patient population with less than 12 months of symptoms duration. Histological analyses and gene expression microarray experiments were performed on isolated synovial tissues to identify pathotype-specific cellular and gene expression markers, evaluate the association between gene expression patterns and the progression of disease, and identify potential biomarkers for prognostic and predictive purposes.

Study Design

[0274] 144 RA individuals fulfilling 2010 American College of Rheumatology (ACR)/European League Against Rheumatism (EULAR) RA Classification Criteria were enrolled at Barts Health National Health Service (NHS) trust as part of the Medical Research Council (MRC) funded multi-center Pathobiology of Early Arthritis Cohort (PEAC). The study received local ethical approval, and all individuals gave written informed consent. Individuals presented with clinically defined synovitis, but with a duration of symptoms of less than 12 months. Study subject characteristics are summarized in FIG. **1**A. Briefly, the average DAS28-ESR score was 5.6 (SD 1.5), approximately 65% were positive for rheumatoid factor and/or anti-citrullinated peptide antibodies (ACPA), and 20% had at least one radiographic erosion. All individuals were naïve to disease-modifying antirheumatic drug (DMARD) and steroid therapy. Upon enrollment and acquisition of demographic and clinical disease parameters, individuals underwent minimally invasive ultrasound (US)-guided synovial biopsy of a clinically active joint (FIG. **1**B). The majority of joints biopsied in this cohort were from the wrist (approximately 65%), with additional representation from MCP/PIP/MTP joints (approximately 15%), while the knee and the elbow together comprised approximately 20% (FIG. **1**C). Individuals were subsequently commenced on standard DMARD therapy (methotrexate (MTX), sulphasalasine (SSZ), and/or hydroxychloroquine (HCQ)) and/or low-dose steroid (intramuscular or oral). A treat-to-target approach to treatment escalation was followed with an aim of low disease activity: 28 joints-disease activity score (DAS28)<3.2. Individuals failing DMARD therapy were commenced on biologic therapy according to the UK National Institute for Clinical Excellence (NICE) prescribing algorithm for RA individuals if they continued to have a DAS28>5.1 at 6 months. After 6 months of treatment, a second synovial biopsy was performed on the same joint (unless clinically contraindicated), together with assessment of disease activity. Ultrasonography scores were collected at the time of biopsy for both the individual biopsied joint as well as a global joint score. Immediately prior to baseline, US-guided synovial biopsy standard longitudinal images of the 1 .sup.st-5.sup.th metacarpo-phalangeal (MCP) joints and midline, radial, and ulnar views of both wrist joints were acquired in addition to standard images of the joint undergoing US-guided synovial biopsy. Images subsequently underwent semi-quantitative (SQ) assessment by a blinded assessor (IL) for both synovial thickening (ST) and power doppler activity (PD) according to standard EULAR outcome measures in rheumatology (OMERACT) US synovitis scores (grade 0-3). For each individual, baseline total mean (12 max) ST (STUS) and PD (PDUS) scores were calculated by deriving the mean of the total scores for ST and PD for all 12 joints, including maximal score in the wrist. STUS and PDUS were also recorded of the biopsied joint. Anonymized plain radiographs of the hands and feet performed at baseline and at 12-month follow-up were scored in time-sequential order according to the van der Heijde modified Sharp score (ShSS) by a trained reader.

Identification of Synovial Pathotypes Based Upon Histology

[0275] To classify individuals into disease pathotypes, synovial biopsy tissue samples were evaluated by immunohistochemical methods. A minimum of six biopsies were paraffin embedded, and 3-μM sections underwent routine hematoxylin and eosin (H&E) staining and were assessed for morphology and sample integrity. Synovial tissue samples collected from 129 of the 144 recruited individuals met sample integrity criteria and were processed for further analyses. In order to determine the degree of immune cell infiltration, sequentially cut sections underwent staining for B cells (CD20), T cells (CD3), macrophages (CD68), and plasma cells (CD138). Sections then underwent SQ scoring (0-4) for CD3, CD20, CD68 lining (CD681) and sublining (CD68sl), and CD138 number. The presence of CD20+ aggregates within synovial tissue was noted, and aggregates were graded (1-3) according to a previously published scoring system (Humby et al. *PLos Medicine* 6(1):e1, 2009).

[0276] Biopsies were then stratified into 1 of 3 synovial pathotypes according to the following criteria: (i) Lymphoid: presence of grade 2-3 CD20+ aggregates, (CD20≥2), and/or CD138>2; (ii) Myeloid: CD68 SL≥2, CD20≤1, and/or CD3≥1, CD138≥2; and (iii) Pauciimmune-fibroid: CD68

SL<2 and CD3, CD20, and CD138<1. Representative microphotographs are shown in FIG. **2**A. Overall, 39% of individuals had a lymphoid pathotype (n=51) rich in immune cells (T, B, and plasma cells); 34% had a myeloid pathotype (n=44) characterized by prevalence of macrophages and T cells, but few B cells and plasma cells; and 27% had a pauciimmune-fibroid pathotype (n=34) characterized by expansion of fibroblasts, but very few immune cells (FIG. **2**B).

Identification of Pathotype-Specific Gene Expression Markers

[0277] To identify genes that were differentially expressed among the three major pathotype clusters, total RNA was extracted from the synovial tissue collected from the subset of PEAC individuals. Synovial samples were homogenized in TRizol Reagent (ThermoFisher Scientific, Life Technologies, Invitrogen Division, UK) using a rotor-stator homogenizer following shearing with a 26-gauge needle. Total RNA was isolated according to the manufacturer's protocol and stored at −80° C. All RNA samples were quantified using spectrophotometric analysis performed on the NanoDrop-ND2000C system (Lab Tech, UK). RNA integrity was determined by RNA Nanochip electrophoresis on the Agilent 2100 Bioanalyzer system (Agilent Technologies, UK). Following confirmation of RNA integrity, 1 μg of total RNA, where available, was used for library preparation using TruSeq RNA Sample Preparation Kit v2 (Illumina). Generated libraries were first amplified with 10 cycles of PCR, the size of the libraries was confirmed using 2200 TapeStation and High Sensitivity D1K screen tape (Agilent Technologies), and their concentration was determined by a qPCR-based method using a library quantification kit (KAPA). The libraries were first multiplexed (five per lane), and then sequenced on Illumina HiSeq2500 (Illumina) to generate 50 million paired-end 75-bp reads.

[0278] Processing and analysis of the RNA-sequencing data was performed using the R programming language, along with packages from the Bioconductor Project. Raw RNA-sequencing reads were processed using the HTSeqGenie Bioconductor package (v. 4.0.1). Briefly, reads were aligned to the reference human genome sequence (build 38) using the GSNAP algorithm and the following parameters: -M 2-n 10-B 2-i 1-N 1-w 200000-E 1-pairmax-rna=200000-clip-overlap. Uniquely aligned read pairs that fell within exons were counted to give an estimate of expression levels for individual genes. Data were further normalized using the variance stabilizing transformation implemented in the DESeq2 Bioconductor package.

[0279] To identify pathotype specific genes, differential expression analysis was performed on the pathotypes classified in a previous microarray study (GEO accession number GSE48780; Dennis et al. *Arthritis Research and Therapy.* 16(2): R90, 2014). Pairwise comparisons were performed between samples in the pauciimmune-fibroid, myeloid, and lymphoid pathotypes. Genes were selected for a pathotype if they showed differential expression with each of the other two pathotypes at a Benjamini-Hochberg adjusted p-value<0.01. These genes were then queried in the baseline RNA-seq data from a subset of 90 individuals from the PEAC cohort.

[0280] For each set of pathotype specific genes, 50 genes were identified that best correlated with the first principal component of the z-score transformed expression data for that gene set. An additional set of 87 genes previously implicated in RA pathobiology were also included. Expression levels determined by NANOSTRING® were concordant with those measured by RNA-sequencing, in samples where both measurements were available (FIG. **2**C). Analyses of NANOSTRING® expression data were performed using R version 3.3.2. For differential expression analyses, the limma Bioconductor package was used with default settings. The Benjamini-Hochberg method was used to adjust for multiple testing, and genes were considered to be differentially expressed if they had an adjusted p-value<0.01. Closer examination of the myeloid pathotype markers identified two clusters of genes, with high correlation within each cluster, but only modest correlation between them. The larger cluster was selected, which contained known myeloid cell-expressed markers, including CD86, PILRA, and C5AR1, as a marker gene set for the myeloid pathotype, reducing the myeloid gene set to 26 genes. The final NANOSTRING® dataset included 212 probes: 49 lymphoid-specific genes (Table 9), 26 myeloid-specific genes (Table 10),

50 pauciimmune-fibroid-specific genes (Table 11), and 87 RA biology-associated genes (Table 12).
TABLE-US-00009 TABLE 9 Lymphoid-Specific Genes NCBI Gene ID Gene Name 973 CD79A 80342 TRAF3IP3 50615 IL21R 9840 TESPA1 11262 SP140 50619 DEF6 23231 SEL1L3 83416 FCRL5 3561 IL2RG 931 MS4A1 64098 PARVG 221188 ADGRG5 3932 LCK 80008 TMEM156 114836 SLAMF6 3738 KCNA3 11040 PIM2 923 CD6 9938 ARHGAP25 4063 LY9 916 CD3E 914 CD2 26279 PLA2G2D 399 RHOH 147138 TMC8 79961 DENND2D 151888 BTLA 79958 DENND1C 81793 TLR10 100505746 ITGB2-AS1 91523 PCED1B 57823 SLAMF7 55423 SIRPG 4064 CD180 374403 TBC1D10C 54440 SASH3 84433 CARD11 11322 TMC6 53347 UBASH3A 5133 PDCD1 84941 HSH2D 201633 TIGIT 9834 FAM30A 11184 MAP4K1 3112 HLA-DOB 6693 SPN 81030 ZBP1 6689 SPIB 5450 POU2AF1
TABLE-US-00010 TABLE 10 Myeloid-Specific Genes NCBI Gene ID Gene Name 54443 ANLN 3687 ITGAX 114548 NLRP3 126014 OSCAR 84034 EMILIN2 3576 CXCL8 4688 NCF2 5329 PLAUR 8843 HCAR3 1230 CCR1 2921 CXCL3 2203 FBP1 30817 ADGRE2 23601 CLEC5A 9582 APOBEC3B 6696 SPP1 55357 TBC1D2 2710 GK 29992 PILRA 942 CD86 728 C5AR1 6364 CCL20 55092 TMEM51 58191 CXCL16 64127 NOD2 53831 GPR84 1317 SLC31A1
TABLE-US-00011 TABLE 11 Pauciimmune-Fibroid-Specific Genes NCBI Gene ID Gene Name 11328 FKBP9 10810 WASF3 57498 KIDINS220 9627 SNCAIP 8555 CDC14B 23259 DDHD2 84255 SLC37A3 200734 SPRED2 121227 LRIG3 84632 AFAP1L2 57088 PLSCR4 4952 OCRL 23348 DOCK9 858 CAV2 23270 TSPYL4 10129 FRY 79188 TMEM43 256356 GK5 26224 FBXL3 9639 ARHGEF10 56256 SERTAD4 8853 ASAP2 10186 LHFPL6 7289 TULP3 90 ACVR1 10194 TSHZ1 51439 FAM8A1 57515 SERINC1 260425 MAGI3 57161 PELI2 342371 ATXN1L 3096 HIVEP1 7220 TRPC1 25959 KANK2 91404 SESTD1 55970 GNG12 51421 AMOTL2 8613 PLPP3 151011 SEPT10 5325 PLAGL1 54682 MANSC1 23328 SASH1 79901 CYBRD1 10427 SEC24B 2059 EPS8 5311 PKD2 79633 FAT4 5066 PAM 8476 CDC42BPA 5358 PLS3
TABLE-US-00012 TABLE 12 RA Biology-Associated Genes NCBI Gene ID Gene Name NCBI Gene ID Gene Name 115650 TNFRSF13C 952 CD38 7049 TGFBR3 6363 CCL19 4312 MMP1 1440 CSF3 4599 MX1 4314 MMP3 7494 XBP1 4050 LTB 11009 IL24 608 TNFRSF17 7078 TIMP3 4940 OAS3 640 BLK 4982 TNFRSF11B 2331 FMOD 3553 IL1B 929 CD14 7481 WNT11 3489 IGFBP6 439921 MXRA7 3543 IGLL1 5154 PDGFA 7852 CXCR4 55801 IL26 59067 IL21 959 CD40LG 50616 IL22 4318 MMP9 7124 TNF 6097 RORC 5175 PECAM1 8792 TNFRSF11A 1311 COMP 4322 MMP13 9235 IL32 9671 WSCD2 695 BTK 3512 JCHAIN 27177 IL36B 3429 IFI27 8600 TNFSF11 3440 IFNA2 3598 IL13RA2 53342 IL17D 29851 ICOS 11096 ADAMTS5 7076 TIMP1 27122 DKK3 3439 IFNA1 10563 CXCL13 3383 ICAM1 50604 IL20 57379 AICDA 930 CD19 6280 S100A9 79037 PVRIG 6382 SDC1 4939 OAS2 2247 FGF2 2919 CXCL1 3458 IFNG 5803 PTPRZ1 6279 S100A8 4319 MMP10 26585 GREM1 9241 NOG 9510 ADAMTS1 8483 CILP 29949 IL19 26525 IL36RN 1508 CTSB 5079 PAX5 91543 RSAD2 3624 INHBA 4049 LTA 3569 IL6 1435 CSF1 2254 FGF9 4938 OAS1 51561 IL23A 3662 IRF4 3456 IFNB1 1437 CSF2 60 ACTB 3557 IL1RN
Pathotype-Specific Eigengene Scores Predict Immunohistochemistry (IHC)-Determined Pathotypes
[0281] Eigengene scores were calculated as previously described (see, e.g., Bueno et al., *Nature Genetics.* 48(4):407-16, 2016, which is herein incorporated by reference in its entirety).
[0282] Gene expression values were collapsed to a single score per sample. A positive correlation was observed between synovial histological grade and the lymphoid or myeloid eigengene scores and, in contrast, a negative correlation between the pauciimune fibroid eigengene score and synovial histological grade (FIG. **2**D). A machine learning technique, k-nearest neighbors, was utilized to develop a predictor of IHC-determined pathotype using the pathotype-specific eigengenes. A five-fold cross-validation methodology predicted the IHC pathotype with 64% accuracy (kappa=0.44). In general, the lymphoid pathotype was more readily predicted than

myeloid or pauciimmune-fibroid, with most misclassifications occurring between pauciimmune-fibroid and myeloid. This suggests that while molecular phenotyping can approximate the IHC pathotype, each technique yields complementary information about the samples.

[0283] Unsupervised clustering of the NANOSTRING® expression data showed strong grouping of pathotype-defined genes in concordance with their initial pathotype assigned by histology (FIG. **3**A). Samples classified as either lymphoid or pauciimmune-fibroid showed highest expression of the lymphoid or pauciimmune-fibroid eigengene score, respectively (FIG. **3**B). Samples classified as myeloid had intermediate levels of the myeloid eigengene score, with higher expression than pauciimmune-fibroid samples, but lower expression than lymphoid samples (FIGS. **3**B and **3**C). Further, the use of a 1-way ANOVA to compare gene expression across the pathotypes revealed that almost every gene measured differed significantly across the three groups; all but 1 of 212 genes had an adjusted p-value<0.01 (FIG. **3**D).

[0284] Pauciimmune-fibroid eigengene scores were inversely correlated with lymphoid and myeloid eigengene scores, while lymphoid and myeloid eigengene scores were positively correlated with each other (FIG. **4**A). Comparing myeloid pathotype samples to either lymphoid or pauciimune-fibroid pathotype samples yielded fewer differentially expressed genes, with lower expression of lymphoid-specific genes in these samples relative to the combined lymphoid/pauciimmune-fibroid group (FIG. **4**B). This is consistent with the scoring algorithm used to determine the pathotypes: myeloid samples mainly differ from lymphoid samples by a relative lack of B cell staining.

[0285] Taken together, these data demonstrate that the pathotype-specific gene sets show strong association with the matched clinically and immunohistochemically determined pathotype.

Example 2. Association Between Pathotype, Gene Expression Levels, Clinical Covariates, Disease Activity, and Disease Progression of Individuals Having RA

[0286] Synovial pathotype and gene expression levels were evaluated for their association with clinical covariates to determine whether they were predictive of disease activity and/or progression in individuals having RA.

Synovial Pathotype and Gene Expression Predict Radiographic Progression

[0287] Baseline pathotypes or gene expression were assessed for association with ongoing structural damage as measured by the Sharp-van der Heijde radiographic progression scores (ShSS) at 12 months after initial biopsy. Although baseline pathotype was not predictive of 12-month erosion or joint space narrowing, lymphoid individuals showed a significantly higher increase in ShSS and had greater risk of progressive disease (>1 ΔShSS) relative to myeloid and pauciimmune-fibroid individuals (FIG. **5**A). Importantly, of the 16 individuals subsequently commenced on biologic therapy between 6 and 12 months of follow-up, a higher proportion fell within the lymphoid (22.8%, 8/35) compared to the myeloid/pauciimmune-fibroid (13.8%, 8/58) groups (Fishers exact test, p=0.27). Thus, despite more intensive treatment regimens (including higher rates of biologic therapeutic agent use), individuals with a lymphoid pathotype were significantly more likely to develop joint damage progression.

[0288] To examine the genes associated with subsequent radiographic progression, ShSS progressors and non-progressors were compared for pre-treatment gene expression (FIG. **5**B). 46 genes with a p-value<0.05 were identified, including the B cell-associated genes CD19, FCRL5, and BCMA. A complete list of these genes is found in Table 1. The pre-treatment lymphoid eigengene score was significantly elevated in individuals who had a 12-month elevation in ShSS score compared to those who did not (FIG. **5**C). In contrast, the myeloid and pauciimmune-fibroid eigengene scores were not elevated in progressors compared to non-progressors (FIG. **5**C). Further, a gene set previously defined as being expressed in osteoclasts (from a dataset in the Harmonizome database) were most highly expressed in lymphoid patients versus myeloid and fibroid patients. The overall osteoclast gene set was significantly enriched in lymphoid patients versus the others (FIG. **5**D), suggesting that the bone resorption mechanism and high B cell infiltration are

concurrent in these patients, perhaps as a result of very favorable conditions for osteoclastogenesis in the synovial lining. In contrast, non-progressors had elevated levels at baseline of pauciimmune-fibroid-associated genes including FGF family members (e.g., Noggin) and cartilage intermediate layer protein. Additionally, osteoprotegrin, a decoy receptor for receptor activator of nuclear factor kappa-B ligand (RANKL), and therefore a negative regulator of osteoclastogenesis, was also elevated in non-progressors.

[0289] To determine whether baseline clinical and gene expression data could be combined into a model for predicting radiographic progression, two complementary approaches were employed: (1) a logistic regression coupled with backward model selection to identify a minimal set of clinical predictors, and (2) a penalized method based on logistic regression, performed using the glm procedure from the stats package of R, with an L1 regularization penalty (LASSO) to identify genes that improve the clinical model.

[0290] Logistic regression, coupled with backward stepwise model selection was applied to baseline clinical parameters against a dependent variable of radiographic progression at 12 months to select which clinical covariate contributed the most to the prediction; 16 baseline clinical covariates were considered as candidates in the regression model. Baseline variables included gender, age, disease duration, ESR, CRP, RF titer, ACPA titer (as continuous variables), VAS, tender and swollen joint number, baseline DAS28-ESR, EULAR response at 6 months (categorical), baseline HAQ, 12 max USST and USPD scores, and baseline pathotype (two categories: Lymphoid v. Pauciimmune-fibroid/Myeloid). Stepwise variable selection yielded a model with 8 clinical variables: baseline RF titer, disease duration, VAS, swollen joint number, DAS28-ESR, baseline pathotype, and 12 max USST and USPD scores. Next, LASSO was applied on these eight clinical covariates and 46 genes identified as being significantly differentially expressed between progressors and non-progressors in order to determine the optimal sparse prediction model. The predictive performance of the models with the genes alone, with the clinical covariates alone, and with the combination of genes and clinical covariates was assessed by computing the area under the receiver operating characteristic curve (AUC). The AUC represents the probability that for any randomly selected pair of individuals with and without a radiographic progression, the individual who had a radiographic progression had a higher predicted risk. A value of 0.50 represents no discrimination, and 1 represents perfect discrimination. Both apparent and internal validations were assessed. The apparent predictive performance of the model evaluated by area under curve (AUC) was 0.85 (95% CI: 0.71-0.98) for clinical covariates alone and 0.91 (% CI CI: 0.79-1.0) for the genes alone (FIG. **5**E).

[0291] A model incorporating RF titer and the expression of seven genes (CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) included in Table 2, yielded a predictor of progression with a lambda ratio of 0.0631 (FIG. **6**A). This model resulted in an improved apparent prediction performance of the model (AUC 0.93, 95% CI: 0.86-1; FIGS. **5**E and **6**B). Bootstrap resampling was performed to correct the AUC for potential over-fitting. To perform the bootstrap, a random sample was drawn with replacement from the original sample. First, the model was trained on the bootstrap sample, validated on the original sample, and an AUC statistic was computed. Next, the model was trained and validated on the same bootstrap sample and another AUC statistic was computed. The difference between the two AUC was computed, and the procedure was repeated 500 times. The 500 differences were then averaged to give an estimate of the optimism. The optimism corrected estimate of the AUC was calculated as the apparent AUC minus the estimated optimism. The optimism-corrected AUC was 0.85 for the pure clinical model, 0.91 for the pure gene expression model, and 0.93 for the combined clinical and gene expression model, suggesting that while examining the expression of genes or clinical covariates alone results in a reliable prognostic model, including both clinical covariates and genes (e.g., CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A) in the model resulted in a prognostic model with improved discrimination between individuals with and without radiographic progression

compared to a prognostic model that only considered clinical covariates or gene expression alone.

Synovial Lymphoid Pathotype and Myeloid Gene Expression Score Correlate with Overall RA Disease Activity

[0292] Baseline disease activity characteristics across the synovial pathotypes were compared with no significant differences identified in age or disease duration across the groups (FIG. **7**A). However, the lymphoid pathotype had the highest levels of ESR, CRP, ACPA titer, swollen joint counts, and DAS28-ESR scores. Assessment of joint damage by X-ray indicated a trend for more severe joint space narrowing in the lymphoid group. Further, ultrasonographic assessment indicated that the lymphoid group had significantly higher levels of synovial thickening and Power Doppler (PD) scores (both within the biopsied joint and overall PD scores), indicating presence of synovitis and supporting the clinical association with active disease. In contrast, the pauciimmune-fibroid group had the lowest levels of acute phase reactants, RF and ACPA positivity, and PD ultrasound scores, despite the presence of active disease, as determined clinically by elevation of DAS28-ESR, swollen joint counts, HAQ and VAS scores, and synovitis determined by ultrasonography.

[0293] The expression of each eigengene score was compared to clinical covariates of disease activity, including DAS28-ESR score, ESR, CRP, swollen and tender joint counts, visual analog scale, HAQ DI score, and ultrasonography, both at the biopsied joint and overall, using Spearman's rank sum correlation method (FIG. **7**B). The myeloid eigengene score was highly associated with many aspects of disease severity, including ESR and CRP, joint counts, DAS28-ESR score, HAQ DI score, and overall PD ultrasonography score. The lymphoid eigengene score was also correlated with many of these, but at a lower level, and was more associated with ultrasonography (both Power Doppler and synovial thickening measures) at the biopsied joint. As expected based on gene expression correlations, the pauciimmune-fibroid eigengene score was negatively associated with many aspects of disease activity.

[0294] Together, these data show that the baseline pathotype or gene expression levels alone, or in combination with clinical covariates, can serve as a prognostic biomarker that is predictive of disease progression in an individual having RA. Consequently, evaluation of baseline pathotype and gene expression levels alone, or in combination with clinical covariates, can be used to identify individuals having RA who may benefit from a treatment including a therapeutic agent (e.g., a biologic therapeutic agent) other than, or in addition to, a DMARD. These results also suggest that the overall disease activity in an individual is most closely related to the expression of myeloid-associated genes in the joint, while elevation of pauciimmune-fibroid genes is associated with lower disease severity across a range of disease activity parameters.

Example 3. Association Between Myeloid and Lymphoid Eigengene Scores and Response to DMARD Therapy

[0295] As discussed in Example 2, pathotype-specific eigengene scores correlated with disease activity and progression. Pathotype-specific eigengene scores were further evaluated for their association with responsiveness of an individual to DMARD therapy, and to determine whether they could serve as biomarkers to predict and/or monitor responsiveness to DMARD therapy.

Pre-Treatment Pathotypes and Gene Expression Levels Association with Response to DMARD Treatment

[0296] Individuals in the three distinct histologically-determined pathotypes were compared for their response to DMARD therapy, as determined by a change in DAS28-ESR at six months and the EULAR response criteria. No significant association of baseline pathotype status with therapeutic outcome was observed, although it was notable that the myeloid and lymphoid pathotype individuals were treated more often with a combination of methotrexate and other DMARDs (FIG. **8**A), consistent with the treat-to-target approach used for individuals with more aggressive disease. Examination of gene expression differences between baseline and six months in individuals who achieved a EULAR good response indicated that multiple inflammatory pathways were reduced, including genes associated with lymphoid aggregates (e.g., CCL19, BTLA, IL21R,

CXCL13, LTA, and LTB) and inflammatory cytokines (e.g., IL6) (FIG. **8**B). In contrast, individuals who did not respond showed smaller decreases in inflammatory gene expression (FIG. **8**C), indicative of persistent synovitis.

[0297] Eigengene scores at baseline were then assessed for associations with therapeutic outcome following DMARD therapy. Higher myeloid and lymphoid eigengene expression (but not pauciimmune-fibroid) were associated with larger decreases in DAS28-ESR scores post-treatment (Spearman's rho=−0.3 (M), p=0.003; rho=−0.21 (L), p=0.044; FIG. **8**D).

[0298] To determine if pathotype eigengene scores were affected by DMARD therapy, baseline and six-month samples were compared separately for individuals that achieved a good response and those that did not respond to DMARD therapy. Individuals that showed a good response to DMARD therapy had very dynamic gene expression, with significant decreases in both lymphoid and myeloid eigengene scores, and a concomitant increase in pauciimmune-fibroid eigengene scores (FIG. **8**E). Non-responders exhibited a more muted change in gene expression, with significant decrease in lymphoid eigengene score, but highly variable changes in myeloid and pauciimmune-fibroid eigengene score (FIG. **8**E).

[0299] Together, these data show that a myeloid eigengene score can serve as a predictive biomarker that is predictive of therapeutic efficacy of a treatment including a DMARD, alone or in combination with a lymphoid eigengene score. Therefore, evaluation of eigengene scores can be used, for example, to identify individuals having RA who may benefit from a treatment including a DMARD, as well as in monitoring the response to a treatment including a DMARD. These results also indicate an ongoing presence of myeloid gene expression in individuals with continuing disease activity despite DMARD therapy.

Example 4. Association Between Serum Gene Expression Levels and Synovial Pathology

[0300] Circulating serum biomarkers, hypothesized from overexpression in synovial subsets of disease (Dennis et al. *Arthritis Research and Therapy.* 16(2): R90, 2014) were assessed for their potential to serve as reliable measures of disease activity in the PEAC cohort.

Serum Biomarkers Reflect Synovial Pathophysiology

[0301] Serum samples from 111 individuals at baseline were assessed for levels of soluble intercellular adhesion molecule 1 (sICAM1), C—X—C motif chemokine 13 (CXCL13), interleukin 8 (IL-8), and matrix metalloproteinase-3 (MMP3) using customized electrochemiluminescence assays incorporating sample diluent blocking reagents to minimize interference from heterophilic antibodies. Serum CXCL13 correlated with global disease metrics, including DAS28 score, serological and ultrasonographic measures of disease activity, and synovial histology (FIGS. **9**A and **9**B). Serum MMP-3 also showed modest yet significant correlation with acute phase reactants and DAS score, as well as synovial histology (FIGS. **9**A and **9**B). Notably, CXCL13 and MMP3 were both elevated in individuals with a lymphoid pathotype, compared to the other two pathotypes (FIGS. **9**C and **9**D). In contrast, sICAM1 and IL-8 exhibited modest and variable correlations with clinical indices, such as tender joint scores, acute phase reactants, auto-antibody titers, and tender joint scores (FIGS. **9**A and **9**B), despite previous reports of elevation in RA individuals (Dennis et al. *Arthritis Research and Therapy.* 16(2): R90, 2014; Cascao et al. *Arthritis Research & Therapy.* 12(5):R196, 2010.

[0302] In summary, these data demonstrate that elevation of some, but not all, inflammatory proteins in the serum of RA correlates with synovitis and clinical disease activity.

VI. Other Embodiments

[0303] Although the foregoing invention has been described in some detail by way of illustration and example for purposes of clarity of understanding, the descriptions and examples should not be construed as limiting the scope of the invention. The disclosures of all patent and scientific literature cited herein are expressly incorporated in their entirety by reference.

Claims

1. A method of predicting disease progression in an individual having rheumatoid arthritis (RA), the method comprising determining an expression level of one or more genes set forth in Table 1 in a sample from the individual, wherein a change in the expression level of the one or more genes relative to a reference expression level identifies the individual as one who is more likely to exhibit disease progression.
2. The method of claim 1, wherein the change is an increase, and the one or more genes set forth in Table 1 are selected from one or more genes set forth in Table 2.
3. The method of claim 2, wherein the one or more genes set forth in Table 2 comprise one or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
4. The method of claim 3, wherein the one or more genes set forth in Table 2 comprise two or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
5. The method of claim 4, wherein the one or more genes set forth in Table 2 comprise three or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
6. The method of claim 5, wherein the one or more genes set forth in Table 2 comprise four or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
7. The method of claim 6, wherein the one or more genes set forth in Table 2 comprise five or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
8. The method of claim 7, wherein the one or more genes set forth in Table 2 comprise six or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
9. The method of claim 8, wherein the one or more genes set forth in Table 2 comprise the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
10. The method of claim 9, wherein the one or more genes set forth in Table 2 consist of the seven following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
11. The method of any one of claims 2-10, wherein the expression level of the one or more genes set forth in Table 2 is increased in the sample relative to the reference expression level, and the method further comprises administering to the individual a therapeutic agent other than, or in addition to, a disease modifying anti-rheumatic drug (DMARD).
12. The method of claim 1, wherein the change is a decrease, and the one or more genes set forth in Table 1 are selected from one or more genes set forth in Table 3.
13. The method of claim 12, wherein the expression level of the one or more genes set forth in Table 3 is decreased in the sample relative to the reference expression level, and the method further comprises administering to the individual a therapeutic agent other than, or in addition to, a DMARD.
14. The method of any one of claims 1-13, wherein disease progression is radiographic progression.
15. The method of claim 14, wherein radiographic progression is characterized by an increase in ShSS.
16. A method of treating an individual having RA, the method comprising administering a therapeutic agent other than, or in addition to, a DMARD to the individual, wherein the individual has been identified as one who is more likely to exhibit disease progression by the method of any one of claims 1-10, 12, 14, and 15.
17. A method of treating an individual having RA, the individual being identified as having (i) an increased expression level of one or more genes set forth in Table 2 in a sample from the individual and/or (ii) a decreased expression level of one or more genes set forth in Table 3 in a sample from the individual relative to a reference expression level, the method comprising administering to the individual a therapeutic agent other than, or in addition to, a DMARD.

18. A method of treating an individual having RA, the method comprising: (a) determining an expression level of one or more genes set forth in Table 2 or Table 3 in a sample from the individual, wherein (i) the expression level of one or more genes set forth in Table 2 in the sample is determined to be increased and/or (ii) the expression level of one or more genes set forth in Table 3 is determined to be decreased relative to a reference expression level; and (b) administering to the individual a therapeutic agent other than, or in addition to, a DMARD based on the expression level of the one or more genes set forth in Table 2 or Table 3 determined in step (a).
19. The method of claim 17 or 18, wherein the one or more genes set forth in Table 2 comprise one or more of the following genes: CD180, CSF2, CXCL1, DENND1C, MMP10, SDC1, and UBASH3A.
20. The method of any one of claims 1-19, wherein the expression level of the one or more genes set forth in Table 1 is an average of the expression level of the one or more genes set forth in Table 1.
21. The method of claim 20, wherein the average of the expression level of the one or more genes set forth in Table 1 is an average of a normalized expression level of the one or more genes set forth in Table 1.
22. The method of any one of claims 1-19, wherein the expression level of the one or more genes set forth in Table 1 is a median of the expression level of the one or more genes set forth in Table 1.
23. The method of claim 22, wherein the median of the expression level of the one or more genes set forth in Table 1 is a median of a normalized expression level of the one or more genes set forth in Table 1.
24. The method of claim 21 or 23, wherein the normalized expression level of the one or more genes set forth in Table 1 is the expression level of the one or more genes set forth in Table 1 normalized to a reference gene.
25. The method of claim 24, wherein the reference gene is ACTB, GAPDH, GUSB, HPRT1, PGK1, RPL19, TUBB, TMEM55B, or a combination thereof.
26. The method of any one of claims 1-25, wherein the reference expression level is a pre-assigned expression level of the one or more genes set forth in Table 1.
27. The method of any one of claims 1-25, wherein the reference expression level is the expression level of the one or more genes set forth in Table 1 in a reference population of individuals having RA who have not previously been treated with a DMARD, the reference population of individuals consisting of a first subset of individuals who exhibited disease progression and a second subset of individuals who did not exhibit disease progression, wherein the reference expression level significantly separates the first and second subsets of individuals based on a significant difference between the expression level of the one or more genes set forth in Table 1 in the first subset of individuals compared to that of the second subset of individuals.
28. The method of claim 27, wherein the first subset of individuals exhibited disease progression and the second subset of individuals did not exhibit disease progression after about 12 months.
29. The method of any one of claims 1-15 and 18-28, further comprising determining one or more clinical covariates of the individual.
30. The method of any one of claims 16, 17, and 19-28, wherein one or more clinical covariates has been determined for the individual.
31. The method of claim 29 or 30, wherein the one or more clinical covariates are one or more of: disease activity score 28-erythrocyte sedimentation rate (DAS28-ESR), disease activity score 28-C reactive protein (DAS28-CRP), rheumatoid factor (RF) titer, disease duration, baseline pathotype, and 12 max ultrasound synovial thickening (USST) and ultrasound power Doppler (USPD) scores.
32. The method of claim 31, wherein the clinical covariate is DAS28-ESR.
33. The method of claim 31, wherein the clinical covariate is a RF titer.
34. The method of any one of claims 1-33, wherein the expression level is a nucleic acid expression level.

35. The method of claim 34, wherein the nucleic acid expression level is an mRNA expression level.
36. The method of claim 35, wherein the mRNA expression level is determined by direct digital counting of nucleic acids, RNA-seq, RT-qPCR, qPCR, multiplex qPCR or RT-qPCR, microarray analysis, or a combination thereof.
37. The method of any one of claims 1-33, wherein the expression level is a protein expression level.
38. The method of claim 37, wherein the protein expression level is determined by an immunoassay, liquid chromatography-mass spectrometry (LC-MS) technology, nephelometry, aptamer technology, or a combination thereof.
39. A method of identifying an individual having RA who may benefit from a treatment comprising a DMARD, the method comprising determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.
40. A method for selecting a therapy for an individual having RA, the method comprising determining a myeloid eigengene score from a sample from the individual, wherein a myeloid eigengene score from the sample that is at or above a reference myeloid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.
41. The method of claim 39 or 40, further comprising determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score that is at or above a reference lymphoid eigengene score identifies the individual as one who may benefit from a treatment comprising a DMARD.
42. The method of any one of claims 39-41, wherein the myeloid eigengene score from a sample is at or above a reference myeloid eigengene score, and the method further comprises administering to the individual a therapeutically effective amount of a DMARD.
43. A method of treating an individual having RA, the method comprising administering a DMARD to the individual, wherein the individual has been identified as one who is more likely to benefit from a treatment comprising a DMARD by the method of any one of claims 39-41.
44. A method of treating RA in an individual identified as having a myeloid eigengene score from a sample from the individual that is at or above a reference myeloid eigengene score, the method comprising administering to the individual a DMARD.
45. The method of claim 44, wherein prior to the administering, a lymphoid eigengene score from a sample from the individual has been determined to be at or above a reference lymphoid eigengene score.
46. A method of treating an individual having RA, the method comprising: (a) determining a myeloid eigengene score from a sample from the individual, wherein the myeloid eigengene score from the sample is determined to be at or above a reference myeloid eigengene score; and (b) administering to the individual a DMARD based on the myeloid eigengene score determined in step (a).
47. The method of claim 46, wherein prior to the administering, the method further comprises determining a lymphoid eigengene score from the sample from the individual, wherein a lymphoid eigengene score in the sample is determined to be at or above a reference lymphoid eigengene score.
48. The method of any one of claims 39-47, wherein the reference myeloid eigengene score is from a reference population of individuals having RA who have been treated with a DMARD therapy, the population of individuals consisting of a first subset of individuals who responded to the DMARD therapy and a second subset of individuals who did not respond to the DMARD therapy, wherein the reference myeloid eigengene score significantly separates the first and second subsets of individuals, based on a significant difference between the myeloid eigengene score in the first

subset of individuals compared to that of the second subset of individuals.
49. The method of claim 48, wherein the first subset of individuals responded to the DMARD therapy and the second subset did not respond to the DMARD therapy after about six months following the initiation of the DMARD therapy.
50. The method of any one of claims 41-43, 45, and 47-49, wherein the reference lymphoid eigengene score is from a reference population of individuals having RA, the population of individuals consisting of a first subset of individuals who responded to DMARD therapy and a second subset of individuals who did not respond to DMARD therapy, wherein the reference lymphoid eigengene score significantly separates the first and second subsets of individuals, based on a significant difference between the lymphoid eigengene score in the first subset of individuals compared to that of the second subset of individuals.
51. The method of any one of claims 39-50, wherein the individual has not been previously treated with a DMARD.
52. The method of any one of claims 39-50, wherein the individual has been previously treated with a DMARD.
53. A method for monitoring the response of an individual having RA to treatment with a DMARD, the method comprising: (a) determining a first myeloid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (b) determining a second myeloid eigengene score from a sample from the individual at second time point; and (c) comparing the first myeloid eigengene score with the second myeloid eigengene score, wherein a decrease in the second myeloid eigengene score relative to the first myeloid eigengene score is predictive of an individual who is likely to respond treatment with a DMARD.
54. The method of claim 53, further comprising: (a) determining a first lymphoid eigengene score from a sample from the individual at a first time point during or after administration of a DMARD; (b) determining a second lymphoid eigengene score from a sample from the individual at second time point; and (c) comparing the first lymphoid eigengene score with the second lymphoid eigengene score, wherein a decrease in the second lymphoid eigengene score relative to the first lymphoid eigengene score is predictive of an individual who is likely to respond treatment with a DMARD.
55. The method of claim 53 or 54, wherein the second myeloid eigengene score is decreased relative to the first myeloid eigengene score, and the method further comprises administering an additional dose of a DMARD to the individual.
56. The method of claim 55, wherein the second lymphoid eigengene score is decreased relative to the first lymphoid eigengene score.
57. The method of any of claims 53-56, wherein the individual has been previously treated with a DMARD.
58. The method of any one of claims 53-57, wherein the decrease is between about 1.25-fold to about 5-fold.
59. The method of claim 58, wherein the decrease is between about 1.25-fold to about 2-fold.
60. The method of claim 59, wherein the decrease is between about 1.25-fold to about 1.5-fold.
61. The method of any one of claims 53-60, wherein the decrease is at least about 1.25-fold.
62. The method of any one of claims 1-61, wherein the sample is a synovial sample.
63. The method of claim 62, wherein the synovial sample is a synovial tissue sample or a synovial fluid sample.
64. The method of any one of claims 11 and 13-63, wherein the DMARD is methotrexate, hydroxychloroquine, sulfasalazine, leflunomide, azathioprine, cyclophosphamide, cyclosporine, mycophenolate mofetil, or a combination thereof.
65. The method of any one of claims 11, 13-38, 62, and 63, wherein the therapeutic agent other than a DMARD is a B cell antagonist, a Janus kinase (JAK) antagonist, a tumor necrosis factor (TNF) antagonist, a decoy TNF receptor, a T cell costimulatory signal antagonist, an IL-1 receptor

antagonist, an IL-6 receptor antagonist, or a combination thereof.

66. The method of claim 65, wherein the JAK antagonist is tofacitinib.

67. The method of claim 65, wherein the IL-6 receptor antagonist is tocilizumab.

68. The method of claim 65, wherein the B cell antagonist is rituximab.