CLAIMS

1. A composition comprising:

(a) a plurality of V-segment primers, and

(b) a plurality of J-segment primers,

each of said plurality of V-segment primers and said plurality of J-segment primers consisting of 15 to 50 nucleotides,

wherein said plurality of V-segment primers comprise T Cell Receptor Beta (TCRB) V-segment primers or immunoglobulin heavy chain (IGH) V-segment primers,

wherein said plurality of J-segment primers comprise TCRB J-segment primers or IGH J-segment primers,

wherein each of said plurality of V-segment primers comprises a first sequence and a second sequence, wherein said first sequence is complementary to a portion of a first region of a TCRB V-region segment or an IGH V-region segment V-segment, said first region located immediately 5' to a second region of said TCRB or IGH V-region segment where untemplated deletions occur during TCRB or IGH gene rearrangement, wherein said second region of said V-region segment is adjacent to and 5' to a V-recombination signal sequence (V-RSS) of said V-region segment, wherein said first sequence is located 3' to said second sequence on said V-segment primer,

wherein said second sequence of each of said TCRB V-segment primers is an adapter for a sequencing oligonucleotide and is not complementary to said first region of said TCRB V-region segment,

wherein said second sequence of each of said IGH V-segment primers is an adapter for a sequencing oligonucleotide and is not complementary to said first region of said IGH V-region segment,

wherein each of said plurality of J-segment primers has a first sequence and a second sequence, wherein said first sequence is complementary to a portion of a first region of a TCRB or IGH J-region segment, said first region located immediately 3' to a second region of said TCRB or IGH J-region segment where untemplated deletions occur during TCRB or IGH gene rearrangement, wherein said second region of said TCRB or IGH J-region segment is adjacent to and 3' to a J-recombination signal sequence (J-RSS) of said TCRB or IGH J-region segment, wherein said first sequence is located 3' to said second sequence on said J-segment primer,

wherein said second sequence of said TCRB J-segment primer is an adapter for a sequencing oligonucleotide and is not complementary to said first region of said TCRB J-region segment,

wherein said second sequence of said IGH J-segment primer is an adapter for a sequencing oligonucleotide and is not complementary to said first region of said IGH J-region segment, and

wherein a single multiplex polymerase chain reaction (PCR) amplification of rearranged nucleic acid molecules from a sample comprising lymphocytes obtained from a mammalian subject, using said plurality of V-segment and J-segment primers produces at least 10^6 distinct amplicons representing a diversity of rearranged TCRB or IGH CDR3 sequences present in said sample.

2. The composition of claim 1, wherein said plurality of V-segment primers have similar annealing strength.

3. The composition of claim 1, wherein all J segment primers anneal to a same conserved framework region motif.

4. The composition of claim 1, wherein the amplicon comprises a sequence that starts from a conserved motif, includes a CDR3 junction and extends into the V segment, and wherein the amplicon is capable of being used to diagnostically identify the J segment.

5. The composition of claim l, further comprising a set of sequencing adapter oligonucleotides, wherein the sequencing oligonucleotides are complementary to said second sequence of said V-segment primer or said second sequence of said J-segment primer.

6. The composition of claim 1, wherein each of the amplicons span a V-D-J junction.

7. The composition of claim 1, wherein said plurality of V-segment primers are each capable of hybridizing to a plurality of V-region segments and said plurality of J-segment primers are each capable of hybridizing to a plurality of J-region segments.

8. The composition of claim 1, further comprising a universal C segment primer for generating cDNA from mRNA.

9. The composition of claim 1, wherein the 3' end of each of said plurality of V-segment primers is anchored at position -43 in the Vβ segment relative to the recombination signal sequence (V-RSS).

10. The composition of claim 1, wherein the plurality of TCRB V-segment primers consist of 14 primers specific to 14 different TCRB V genes.

11. The composition of claim 1, wherein said first sequences of said plurality of TCRB V-segment primers comprise sequences that are selected from the group consisting of SEQ ID NOS:1-45.

12. The composition of claim 1, wherein said first sequences of said plurality of TCRB V-segment primers comprise sequences that are selected from the group consisting of SEQ ID NOS:58-102.

13. The composition of claim 1, wherein each V-segment is complementary to at least one V-segment primer.

14. The composition of claim 1, wherein said primers do not cross an intron/exon boundary.

15. The composition of claim 1, wherein said plurality of J-segment primers hybridize with a conserved element of said TCRB region segment and have similar annealing strength.

16. The composition of claim 1, wherein said plurality of J segment primers consist of at least five primers specific to five different TCRB J genes.

17. The composition of claim 1, wherein said plurality of J-segment primers have sequences that are selected from the group consisting of SEQ ID NOS:46-57 and 483.

18. The composition of claim 1, wherein said plurality of J-segment primers have sequences that are selected from the group consisting of SEQ ID NOS: 103-113, 468 and 484.

19. The composition of claim 1, wherein there is a J segment primer for each TCRB J-region segment.

20. The composition of claim 1, wherein the amplified TCRB J-region segments each have a unique four base tag at positions + 11 through + 14 downstream of the J-RSS site.

21. The composition of claim 20, further comprising a set of sequencing adapter oligonucleotides that hybridize adjacent to a four base tag within the amplified TCRB J-region segments at positions +11 through +14 downstream of the J-RSS site.

22. The composition of claim 21, wherein said set of sequencing adapter oligonucleotides are selected from the group consisting of SEG ID NOS:470-482.

23. A method comprising:

(a) contacting a sample comprising rearranged nucleic acid molecules of a TCRB or IGH CDR3 region from lymphocytes obtained from a mammalian subject with said composition of claim 1; and

(b) amplifying said rearranged nucleic acid molecules from said sample using said composition in a single multiplex PCR, thereby producing at least 10^6 distinct amplicons representing a diversity of TCRB or IGH genes.

24. The method of claim 23, further comprising sequencing said at least 10^6 distinct amplicons using a set of sequencing oligonucleotides that hybridize to a region of said at least 10^6 distinct amplicons.

25. The method of claim 23, further comprising calculating the total diversity of TCRβ CDR3 sequences among the distinct amplicons.

26. The method of claim 25, wherein said total diversity is greater than $1 \cdot 10^6$ sequences.

27. The method of claim 25, wherein said total diversity is greater than $2 \cdot 10^6$ sequences.

28. The method of claim 25, wherein said total diversity is greater than $3 \cdot 10^6$ sequences.

29. A method of diagnosing immunodeficiency in a human subject, said method comprising:

(i) measuring a diversity of at least 10^6 rearranged TCRB or IGH CDR3 sequences in a first sample from the human subject, wherein measuring the diversity of at least 10^6 rearranged TCR CDR3 sequences comprises the steps of:

(a) selecting a plurality of V-segment primers and a plurality of J- segment primers of the composition of claim 1;

(b) combining the plurality of V-segment and J-segment primers with a sample of genomic DNA comprising rearranged nucleic acid molecules of a TCRB or IGH CDR3 region from lymphocytes obtained from the subject;

(c) amplifying said rearranged nucleic acid molecules from said sample using said plurality of V-segment and J-segment prime, thereby producing at least 10^6 distinct amplicons representing a diversity of TCRB or IGH CDR3 genes in said sample;

(d) sequencing said at least 10^6 distinct amplicons; and

(e) calculating the diversity of TCRB or IGH CDR3 sequences among the at least 10^6 distinct amplicons for said subject; and

(ii) comparing the diversity of TCRB or IGH CDR3 sequences of said subject to a diversity of TCRB or IGH CDR3 sequences determined from a second sample.

30. The method of claim 29, wherein comparing the diversity is determined by calculating using the following equation:

$$\Delta(t) = \sum_{x} E(n_x)_{measurement1+2} - \sum_{x} E(n_x)_{measurement2} = S\int_{0}^{00} e^{-\lambda}(1-e^{-\lambda 1})dG(\lambda)$$

wherein $G(\lambda)$ is the empirical distribution function of the parameters $\lambda_1, ..., \lambda_s$, n_x is the number of clonotypes sequenced exactly x times, and

$$E(n_x) = S\int_0^{00}\left(\frac{e^{-\lambda}\lambda^x}{x!}\right)dG(\lambda).$$

31. The method of claim 29, wherein the first sample and the second sample comprises genomic DNA.

32. The method of claim 31, wherein the first sample A is from a patient and the second sample is from a normal subject.

33. The method of claim 31, wherein the first sample is from a patient before a therapeutic treatment and the second sample is from the patient after treatment.

34. The method of claim 29, wherein the first sample and the second sample of genomic DNA are from the same patient at different times during treatment.

35. The method of claim 29, further comprising diagnosing said subject based on the comparison of diversity among the samples of genomic DNA.

36. The method of claim 29, wherein the immunocompetence of a human patient is assessed by the comparison.

37. The composition of claim 1, wherein the sample comprises genomic DNA obtained from the mammalian subject.

38. The composition of claim 1, wherein the sample comprises cDNA transcribed from mRNA obtained from the mammalian subject.

39. The composition of claim 5, wherein the sequencing oligonucleotides are selected from the group consisting of SEG ID NOS:470-482.

40. The composition of claim 1, wherein said second sequence of each of said V-segment primers comprises a sequencing adapter oligonucleotide sequence.

41. The composition of claim 40, wherein said sequencing adapter oligonucleotide is selected from the group consisting of SEQ ID NOS:470-482.

42. The composition of claim 1, wherein said second sequence of each of said J-segment primers comprises a sequencing adapter oligonucleotide sequence.

43. The composition of claim 42, wherein said sequencing adapter oligonucleotide sequence is selected from the group consisting of SEQ ID NOS:470-482.

44. The composition of claim 1, wherein said first region of said V-segment is at least 40 bases from a consensus motif at a 5' end of the V-RSS.

45. The composition of claim 1, wherein each of said J-segment primers has a 3' end that is positioned at least 14 base pairs from a consensus motif at a 3' end of a J-recombination signal sequence (J-RSS).

46. The composition of claim 1, wherein an amplicon produced from said plurality of V-segment primers and said plurality J-segment primers is approximately 200 bases in length.

47. The composition of claim 1, wherein said first sequences of said plurality of V-segment primers consist of SEQ ID NOS:1-45.

48. The composition of claim 1, wherein said first sequences of said plurality of V-segment primers consist of SEQ ID NOS:58-102.