CLAIMS

1. A method for production of single stranded DNA amplicons via an asymmetric polymerase chain reaction (PCR), comprising the steps of:

(a) subjecting an initial mixture to a first phase of thermal cycling under amplifying PCR conditions, said initial mixture comprising:

(i) a nucleic acid sample;

(ii) an Asymmetric Primer Pair,

(iii) a thermostable DNA polymerase, and

(iv) PCR Reagents,

wherein the first phase of thermal cycling comprises cycling through at least three temperatures, said three temperatures comprising: (1) a first temperature above the T_m of the target nucleic acid for denaturation, (2) a second temperature below the T_m of the Unextended Primer for primer annealing, and (3) a third temperature suitable for extension by the thermostable DNA polymerase, thereby producing an intermediate mixture, said intermediate mixture comprising double stranded DNA amplicons extended from both the Unextended Primer and the Extended Primer if the target nucleic acid is present in the sample;

(b) subjecting the intermediate mixture to a second phase of thermal cycling, said second phase of thermal cycling comprising cycling through: (1) a fourth temperature above the T_m of the double stranded DNA amplicons for denaturation, (2) a fifth temperature that is

(i) below the T_m of the Extended Primer,

(ii) above the T_m of the Unextended Primer and

(iii) suitable for extension by the thermostable DNA polymerase,

thereby producing a final mixture, said final mixture comprising single stranded DNA amplicons if the target nucleic acid is present in the sample; wherein the asymmetric PCR is performed in the absence of a Generic Primer.

2. The method of claim 1, wherein step (a) is preceded by an initial denaturation period.

3. The method of claim 2, wherein the initial denaturation is carried out at the first temperature.

4. The method of claim 1, wherein step (b) is followed by an additional extension period.

5. The method of claim 4, wherein the additional extension is carried out at the fifth temperature.

6. The method of claim 1, wherein the first and fourth temperatures are the same and/or the third

and fifth temperatures are the same.

7. The method of claim 1, wherein the "B" region of the Extended Primer is Inverted Repeat of at least a portion of the "A" region.

8. The method of claim 1, wherein the Unextended Primer does not have a 5' tail extension.

9. The method of claim 1, wherein the Extended Primer comprises a "C" region.

10. The method of claim 9, wherein the "C" region alone forms a recognition site for a restriction endonuclease.

11. The method of claim 9, wherein the "C" region, in combination with the flanking "A" and/or "B" region nucleotides, forms a recognition site for a restriction endonuclease.

12. The method of claim 1, wherein the T_m of the Extended Primer is greater than the T_m of the Unextended Primer by at least 8^o C.

13. The method of claim 1, wherein the T_m of the "A" region of the Extended Primer and the T_m of the Unextended Primer vary by no more than 3^o C.

14. The method of claim 1, wherein (a) the first temperature is 95^o C.; (b) the second temperature is 58^o C.; (c) the third temperature is 72^o C.; (d) the fourth temperature is 95^o C.; (e) the fifth temperature is 72^o C.; or (f) any combination of (a)-(e).

15. The method of claim 1, wherein the nucleic acid sample is a test nucleic acid sample.

16. The method of claim 1, wherein the nucleic acid sample is a control nucleic acid sample.

17. The method of claim 1, which is a multiplex PCR reaction.

18. The method of claim 1, wherein the first phase results in exponential amplification of both strands of the target nucleic acid and/or the second phase results in linear amplification of a single strand of the target nucleic acid.

19. The method of claim 1, wherein the Asymmetric Primer Pair comprises sequences complementary to bacterial sequences or fungal DNA sequences.

20. The method of claim 1, wherein the sample is a biological sample.

21. The method of claim 20, wherein the biological sample is blood or a sample processed, extracted or fractionated therefrom.

22. The method of claim 20, wherein the biological sample is peritoneal dialysis fluid or a sample processed, extracted or fractionated therefrom.

23. The method of claim 20, wherein the biological sample is urine or a sample processed, extracted or fractionated therefrom.

24. The method of claim 20, wherein the biological sample is sputum or a sample processed, extracted or fractionated therefrom.

25. The method of claim 20, wherein the biological sample is a wound swab or a sample processed, extracted or fractionated therefrom.

26. The method of claim 1, wherein the sample is an environmental sample.

27. The method of claim 1, which further comprises detecting the single stranded PCR amplicon.

28. The method of claim 1, wherein the Unextended Primer has a 5' tail extension of 3 nucleotides or less.

29. The method of claim 1, wherein the "B" region of the Extended Primer is a Direct Repeat of at least a portion of the "A" region.