CLAIMS

1. A method of providing one or more instances of a target double-stranded nucleic acid from a plurality of nucleic acid fragments, comprising:

a plurality of initial hybridisation steps (H_1-H_4), each initial hybridisation step comprising hybridising respective pairs of partially overlapping nucleic acid fragments to form a plurality of hybridised fragments; and

one or more further hybridisation steps (H_5-H_7), each further hybridisation step comprising hybridising respective pairs of partially overlapping hybridised fragments which are the direct product of a corresponding pair of earlier hybridisation steps to form longer hybridised fragments, where each of the pair of earlier hybridisation steps comprises one of the initial hybridisation steps or one of the further hybridisation steps;

wherein said one or more further hybridisation steps comprise at least one further hybridisation step (H_F) for which both of the corresponding pair of earlier hybridisation steps comprise an error-detecting type of hybridisation step (H_E);

the error-detecting type of hybridisation step (H_E) comprising:

performing an error detecting operation to detect whether the hybridised fragments formed in the error-detecting type of hybridisation step comprise at least one erroneous hybridised fragment comprising at least one mismatching base pair in an overlap region hybridised in the error-detecting type of hybridisation step; and

discarding at least part of said at least one erroneous fragment to exclude the at least one erroneous fragment from a subsequent further hybridisation step;

wherein the target double-stranded nucleic acid comprises a first strand of single-stranded nucleic acid hybridised to a second strand of single-stranded nucleic acid; and

in each hybridisation step, the hybridised fragment of nucleic acid formed in that hybridisation step is bound to a surface of a reaction site via the first strand or the second strand.

2. The method of claim 1, wherein one of said at least one further hybridisation step performed at a given reaction site comprises hybridising:

first hybridised fragments bound to the surface of the given reaction site via one of the first strand and the second strand; and

second double-stranded fragments formed at an earlier reaction site in an earlier hybridisation step, when bound to a surface of the earlier reaction site via the other of the first strand and the second strand.

3. The method of any of claims 1 and 2, wherein the initial hybridisation steps and the at least one further hybridisation step form a sequence of hybridisation steps in which for any pair of hybridisation steps in which the second hybridisation step of the pair hybridises a hybridised

fragment formed in the first hybridisation step of the pair with a further fragment, the hybridised fragments formed in the pair of hybridisation steps are bound to a surface of a corresponding reaction site via opposite ones of the first strand and the second strand respectively.

4. The method of any preceding claim, wherein the method is performed using an apparatus (2) comprising at least one lane (9) of reaction sites (6) aligned in a predetermined direction and a fluid control element to direct a flowing fluid over each reaction site in the predetermined direction.

5. The method of claim 4, the apparatus further comprising temperature control circuitry to independently control a temperature at each reaction site.

6. The method of claim 4, wherein the reaction sites comprise one of:

portions of a surface without a physical barrier between adjacent reaction sites, and

portions of a surface with a selectively removable physical barrier between adjacent reaction sites.

7. The method of any preceding claim, wherein said error detecting operation comprises at least one of:

weakening a bond between the partially overlapping fragments forming each detected erroneous hybridised fragment, and providing fluid to wash away said at least part of said at least one erroneous hybridised fragment; and

exposing said hybridised fragments to a mismatching base pair detecting enzyme.

8. The method of any preceding claim, wherein said error detecting operation comprises adjusting a temperature of a reaction site on which the hybridised fragments are formed to a target temperature corresponding to a margin below an expected melting temperature of the overlap region formed in that hybridisation step for an error-free hybridised fragment.

9. The method of claim 8, wherein partitioning of the target double-stranded nucleic acid into the nucleic acid fragments is selected such that, at each overlap region, a difference between the expected melting temperature of the overlap region in an error-free hybridised fragment and an expected melting temperature of the overlap region in an erroneous hybridised fragment with at least one base error within that overlap region is greater than a predetermined threshold.

10. The method of claim 9, wherein said predetermined threshold is at least 0.1 °C.

11. The method of any preceding claim, wherein hybridised fragments are transported in a flowing fluid between reaction sites on which respective hybridisation steps are performed.

12. The method of any preceding claim, wherein in at least one of said error-detecting type of hybridisation step, remaining hybridised fragments following the error detection operation are selectively detached from a surface of a reaction site.

13. The method of claim 12, wherein the selective detaching of the remaining hybridised fragments is temperature-controlled.

14. The method of any preceding claim, wherein each hybridisation step, other than any hybridisation step performed on a pair of single-stranded fragments, comprises a ligation operation performed on the hybridised fragments;

wherein for an error-detecting type of hybridisation step, the ligation operation is performed on the remaining double-stranded fragments excluding the at least one erroneous hybridised fragment detected in the error detection operation.

15. The method of any preceding claim, wherein each of the plurality of nucleic acid fragments comprises at least one overlap region for overlapping with a corresponding overlap region of another of the nucleic acid fragments; and

each base of the target double-stranded nucleic acid is within one of the overlap regions of one of the plurality of nucleic acid fragments.

16. A method of providing one or more instances of a target double-stranded nucleic acid from a plurality of nucleic acid fragments, comprising:

a plurality of initial hybridisation steps (H_1-H_4), each initial hybridisation step comprising hybridising respective pairs of partially overlapping nucleic acid fragments to form a plurality of hybridised fragments; and

one or more further hybridisation steps (H_5-H_7), each further hybridisation step comprising hybridising respective pairs of partially overlapping hybridised fragments which are the direct product of a pair of earlier hybridisation steps to form longer hybridised fragments, where each of the pair of earlier hybridisation steps comprises one of the initial hybridisation steps or one of the further hybridisation steps;

wherein the target double-stranded nucleic acid comprises a first strand of single-stranded nucleic acid hybridised to a second strand of single-stranded nucleic acid;

in each hybridisation step, the hybridised fragment of nucleic acid formed in that hybridisation step is bound to a surface of a reaction site via the first strand or the second strand; and

the method comprises controlling an order and timing of a sequence in which the plurality of initial hybridisation steps and one or more further hybridisation steps is performed;

wherein at least one of said one or more further hybridisation steps comprises an error-detecting type of hybridisation step;

wherein the error-detecting type of hybridisation step comprises:

performing an error detecting operation comprising adjusting a temperature of a reaction site on which the hybridised fragments are formed to a target temperature corresponding to a margin below an expected melting temperature of an overlap region hybridised in that hybridisation step for an error-free hybridised fragment; and

following the error detection operation, selectively detaching remaining hybridised fragments from a surface of the reaction site, wherein the selective detaching of the remaining hybridised fragments is temperature-controlled.