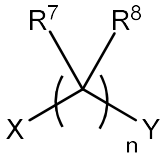
CLAIMS

1. An *in vitro* method for transducing a protein, a nucleic acid or a combination thereof into a cell, wherein the method comprises contacting said cell with the protein, a nucleic acid or a combination thereof and a transduction buffer, wherein the transduction buffer comprises (i) a transduction compound, (ii) a salt selected from sodium, lithium, potassium, caesium or rubidium salt; wherein the transduction compound

a) has the general formula:



(I)

wherein:

X is selected from NR^1R^2, $NR^1R^2R^3$+, OH and $COOR^4$;

Y is selected from SO_3H, SO_3^-, COO^-, $CONH_2$, $COOR^{12}$, $CONR^5R^6$, tetrazole, OH, $NR^{10}R^{11}$, and H;

n is 3, 4, 5 or 6;

R^1, R^2 and R^3, are each independently selected from H, C1-6 alkyl, C5-10 aryl, C6-15 aralkyl, COR^9; C1-6 alkyl, C5-10 aryl and C6-15 aralkyl may optionally be substituted with R^Y, OH or COOH;

or R^1 and R^2 may come together with the nitrogen to which they are attached to form heterocyclyl;

or when X is $NR^1R^2R^3$+, R^3 may be absent and R^1 and R^2 may come together with the nitrogen to which they are attached to form heteroaryl;

R^4, R^5, R^6, R^9, R^{10}, R^{11}, R^{12} are independently selected from H and C1-6 alkyl;

R^7 and R^8 are independently selected from H, C1-6 alkyl and OH;

heterocyclyl is a monocyclic ring which is saturated or partially unsaturated, containing where possible 1 or 2 ring members independently selected from N, NR^{13}, $NR^{13}R^{14}$+ and O, and 2 to 5 carbon atoms; heterocyclyl may optionally be substituted with C1-C6 alkyl, C1-C6 carboxylic acid or C1-C6 alkyl substituted with R^Y;

heteroaryl is a 5 or 6 membered aromatic ring containing, where possible, 1, 2 or 3 ring members independently selected from N, NR^{13}, $NR^{13}R^{14}$+ and O; heteroaryl may optionally be substituted with C1-C6 alkyl, C1-C6 carboxylic acid or C1-C6 alkyl substituted with R^Y;

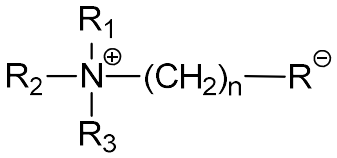
R^{13} and R^{14} are independently selected from H, C1-6 alkyl, C1-C6 carboxylic acid and C1-C6 alkyl substituted with R^Y;

alkyl is a linear or branched saturated hydrocarbon;

R^Y is selected from SO_3H, SO_3^-, COO^-, $CONH_2$, $COOR^{12}$, $CONR^5R^6$, tetrazole, OH and $NR^{10}R^{11}$;

C1-6 carboxylic acid means –COOH or a C1-5 alkyl chain substituted with COOH and tautomers, solvates, zwitterions and salts thereof; or

b) has the general formula:



wherein R_1 is methyl, ethyl, propyl, butyl, pentyl, hexyl;

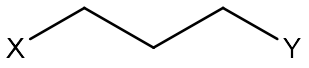
R_2 is methyl, ethyl, propyl, butyl, pentyl, hexyl;

R_3 is methyl, ethyl, propyl, butyl, pentyl, hexyl;

R^- is SO_3^- or COO^- or POO^- and

n is 3-6, *i.e.* 3, 4, 5 or 6.

2. The *in vitro* method of claim 1 wherein the transduction buffer wherein the transduction compound has the general formula:



(II)

wherein

X is selected from NR^1R^2 and $NR^1R^2R^3$+;

Y is selected from SO_3H, SO_3-, COO-, $CONH_2$, $COOR^{12}$ and $CONR^5R^6$;

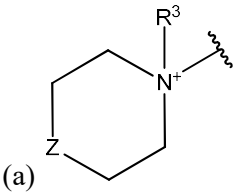
R^1, R^2 and R^3, are each independently selected from H and C1-6 alkyl which may optionally be substituted with OH or COOH; or R1 and R2 may come together with the nitrogen to which they are attached to form heterocyclyl, preferably piperidine, piperazine or morpholine, which may be optionally substituted; or when X is $NR^1R^2R^3$+, R^3 may be absent and R^1 and R^2 may come together with the nitrogen to which they are attached to form heteroaryl, preferably pyridyl, which may be optionally substituted;

and all other groups are as defined in formula I above.

3. The *in vitro* method of claim 1 or 2, wherein the transduction compound:

(a) contains a quaternary nitrogen group and wherein the quaternary nitrogen is part of an aliphatic or aromatic ring structure;

(b) is a compound of formula I or II wherein X is

(a)  ;

Z is selected from $C(R^{15})_2$, NR^{13}, $NR^{13}R^{14}$+ and O;

each R^{15} is independently selected from H, C1-6 alkyl, C1-C6 carboxylic acid and C1-C6 alkyl substituted with R^Y;

R^3 is selected from H, C1-6 alkyl, C5-10 aryl, C6-15 aralkyl, COR^9; C1-6 alkyl, C5-10 aryl and C6-15 aralkyl may optionally be substituted with R^Y, OH or COOH, preferably R_3 is $–CH_3$;

R^{13} and R^{14} are independently selected from H, C1-6 alkyl, C1-C6 carboxylic acid and C1-C6 alkyl substituted with R^Y and/or

(c) is selected from compounds 01-06, 08, 10-12, 14-17, 20-31 and 34-46 in Table 1.

4. The *in vitro* method of any one of the preceding claims, wherein the transduction buffer further comprises and osmoprotectant selected from glycine, histidine, alanine, isoleucine, arginine, asparagine, leucine, aspartic acid, lysine, glutami acid, cysteine, methionine, phenylalanine, glutamine, threonine, tryptophan, proline, valine, orninthine, selenocysteine, serine, tyrosine and proline.

5. The *in vitro* method of any one of the preceding claims, wherein:

the transduction buffer further comprises an osmoprotectant selected from glycine and/or glycerol;

the osmoprotectant is at a concentration of between about 5 and about 500 mM;

the transduction buffer further comprises an osmoprotectant at a concnetraiotn of between 5 and about 500mM;

the transduction buffer has an osmolality of at least 300 mOsm/kg;

the transduction compound is at a concentration of between about 0.1 mM and about 500 mM; and/or

wherein the salt is sodium chloride.

6. The *in vitro* method of any one of the proceeding claims, wherein the transduction buffer further comprises: a biological pH buffer such as PBS, TES or HEPES; a viscosity enhancer such as polyvinylpyrrolidone (PVP); an inhibitor of the interferon response pathway; and/or a growth factor, for example selected from EGF, FGF, HGF, BDNF, PDGF, VEGF or IGF, or is any combination thereof.

7. The *in vitro* method of any one of the preceding claims, wherein the transduction buffer comprises NDSB-201 as a transduction compound, and preferably further comprises GABA as a transduction compound.

8. The *in vitro* method of any one of the preceding claims, wherein the transduction buffer comprises: (i) NDSB-201 and GABA as transduction compounds; (ii) sodium chloride as the salt; and preferably (iii) glycine and/or glycerol as the osmoprotectant; and wherein the osmolality of the transduction buffer is at least 300 mOsm/kg.

9. The *in vitro* method of any one of the proceeding claims, wherein the transduction buffer further comprises an activator/enhancer of a sodium-hydrogen transporter, wherein said activator/enhancer is a growth factor.

10. A transduction buffer as recited in any one of claims 1 to 9, wherein the osmolality of the transduction buffer is at least 650mOsm/kg.

11. A pharmaceutical composition comprising the transduction buffer of claim 10.