Nucleoside analogue-sparing antiretroviral combinations may be interesting as first-line therapies as they spare a complete class of drugs that will remain fully active for later use and prevent the risk of mitochondrial toxicity related to exposure to nucleoside reverse transcriptase inhibitors (NRTIs). This strategy is also used in patients failing NRTIs with cross-resistance to compounds in this class. Different combinations of antiretroviral drugs are theoretically available. Non-nucleoside reverse transcriptase inhibitors (NNRTIs) associated with protease inhibitor (PI) and boosted double-PI combinations have been studied through small, non-comparative clinical studies and preliminary results suggest that they are efficient and often well-tolerated. However, NNRTIs and PIs are extensively metabolized in the liver through cytochrome P450, leading to pharmacokinetic interactions; a good knowledge of the interactions between NNRTIs and PIs, or between PIs, is helpful in assisting physicians in clinical practice in choosing drugs and doses. Access to a therapeutic drug monitoring service to confirm that appropriate drug exposures are achieved is useful when using such regimens. Some negative kinetic interactions may lead to complicated combinations with a high pill burden that reduces their applicability. Gastrointestinal toxicity often remains a limiting factor in the use of boosted double-PI combinations. Non-comparative studies have allowed selection of NRTI-sparing options that now need to be compared with the current standard of care in comparative clinical trials before being considered as valuable options. Other NRTI-sparing therapeutic strategies are emerging: PI monotherapy with lopinavir/ritonavir has been evaluated in a small group of naive patients and appears promising. Drugs belonging to new classes currently under investigation, such as entry inhibitors, might be included early in the antiretroviral treatment of patients as soon as compounds with a convenient route of administration are available, increasing the number of therapeutic combinations without NRTIs.
compounds that is available within the class of entry inhibitors, has been studied in the context of virological failure to other classes of drugs; its value in first-line therapy has not been evaluated but may be limited by subcutaneous administration.

NRTI-sparing antiretroviral combinations may be useful as first-line therapies for the following reasons: i) sparing a complete class of drugs that will remain fully active for later use and ii) preventing the risk of mitochondrial toxicity related to NRTI exposure. In patients failing antiretroviral therapy with resistance to NRTIs and thus excluding this class of drugs from further therapeutic regimens, double PI combinations or NNRTI and PI combinations have been evaluated and may be effective. Entry inhibitors may be helpful in improving the virological response to treatment in these patients.

We will successively review the currently available data on NRTI-related toxicity, NRTI cross-resistance, the pharmacokinetic interactions, activity and tolerance of dual NNRTI–PI therapy and double-PI therapy in naive and pretreated patients, the limits of such combinations and the new perspectives in NRTI-sparing antiretroviral therapy due to the development of HIV entry inhibitors.

NRTI mitochondrial toxicity

NRTIs are incorporated into the elongated viral DNA molecules transcribed by HIV reverse transcriptase and thereby inhibit HIV replication. However, they also inhibit human DNA polymerase activity. Mitochondrial DNA (mtDNA) polymerase gamma appears particularly susceptible to inhibition. It has been proposed that NRTI-related mitochondrial dysfunction in various tissues is the cause of many of the side effects and toxicities associated with NRTIs, including lactic acidosis, hepatic steatosis, myopathy, pancreatitis, peripheral neuropathy and lipoatrophy.

Symptomatic hyperlactataemia refers to elevated lactate levels without acidosis but with associated symptoms, the most common being abdominal pain and distension, nausea, vomiting, diarrhoea, dyspnea and weight loss. Lactic acidosis syndrome refers to severe symptomatic decompensated hyperlactataemia with metabolic acidosis, hepatomegaly and steatosis. Fatalities have been reported in late pregnancy with a number of NRTI combinations, especially with stavudine and didanosine [3,4]. Antiretroviral treatment should be suspended in cases of clinical and laboratory manifestations of lactic acidosis. Small studies suggest that after resolution of symptoms, many patients can tolerate the administration of a revised NRTI-containing regimen, especially with abacavir or lamivudine [4,5], but insufficient data exist to recommend this strategy versus treatment with a NRTI-sparing regimen.

Certain features of lipodystrophy syndrome have been hypothesized as being tissue-specific mitochondrial toxicities caused by NRTI treatment [6–8]. A cross-sectional study found that NRTI therapy with zidovudine or stavudine was associated with mtDNA depletion in adipocytes, consistent with the hypothesis that NRTI-induced mtDNA depletion contributed to the pathogenesis of subcutaneous fat wasting [9]. Lipoatrophy in the face and extremities has been reported to increase with long-term NRTI exposure and different studies argue for a preponderant role of stavudine among NRTIs in the occurrence of lipoatrophy [8,10].

NRTI cross-resistance

Although numerous antiretroviral combinations are available to provide potent suppression of viral replication, therapeutic choices have to take into consideration the potential impact of viral resistance on future treatment options. Resistance and cross-resistance to NRTIs is becoming better understood [11]. Different mutations have been reported to be selected under NRTIs and, except for the M184V mutation, they all carry a risk of cross-resistance. The amino acid substitutions at codons 41, 67, 70, 210, 215 and 219 are selected by zidovudine or stavudine and both agents show reduced susceptibility in their presence. These mutations are termed thymidine analogue mutations (TAMs). The presence of multiple TAMs including 41L, 210W and 215Y causes class-wide resistance to NRTIs including zidovudine, stavudine, abacavir, didanosine, tenofovir and zalcitabine. The RT mutation K65R is selected by didanosine, tenofovir and abacavir and causes broad phenotypic cross-resistance among NRTIs. In contrast to the TAMs pathway, the occurrence of decreased susceptibility does not need the accumulation of multiple resistance mutations. Q151M, often reported after therapy with didanosine in combination with zidovudine or stavudine, confers resistance to zidovudine, didanosine, zalcitabine, stavudine and abacavir. The activity of tenofovir is also reduced. Insertions at position T69 are noted in a small number of heavily NRTI-experienced patients and confer high-level resistance to zidovudine, stavudine, abacavir, didanosine and zalcitabine when combined with TAMs [12].

Thus, resistance to multiple nucleoside analogues can result from several genetic pathways. Once failure has occurred under a nucleoside-containing regimen, therapeutic options within this class may be limited due to cross-resistance between compounds. For this reason, the use of a nucleoside-sparing regimen may be necessary. On the other hand, first-line antiretroviral therapy without NRTIs would leave this class entirely available for future treatment options.
NNRTI and PI combinations

Pharmacokinetic interactions
NNRTIs and PIs are extensively metabolized in the liver through cytochrome P450, leading to pharmacokinetic interactions. Nevirapine is an inducer of cytochrome P450 activity, efavirenz is a mixed inducer and inhibitor and delavirdine is an inhibitor of cytochrome P450. Thus, compared with nevirapine, delavirdine has opposite interactions to compounds utilizing the same metabolic pathway, particularly PIs, whose plasma concentrations are increased in the presence of delavirdine. Reciprocal kinetic interactions and recommended dosages of drugs are summarized in Table 1. Plasma levels of the three available NNRTIs are not significantly altered by available PIs, except delavirdine, whose AUC is decreased by 40% in the presence of nelfinavir and by 60% in the presence of amprenavir [13], and efavirenz, whose plasma AUC is increased by 21% in the presence of ritonavir. The decrease in PI plasma concentrations observed when they are combined with nevirapine or efavirenz is reduced when low doses of ritonavir, which strongly inhibits cytochrome P450, are associated with the combination of a PI and an NNRTI [14–16,19].

Clinical data
In naive patients, initiation of an antiretroviral treatment combining an NNRTI and a PI would allow an NRTI-sparing first-line therapy. Although guidelines recommend the use of a ‘triple-drug combination’, the intrinsic potency of both NNRTIs and PIs would allow the use of them in this unusual combination of only two drugs. Few trials have evaluated the activity of this type of antiretroviral therapy.

We will review the studies performed in i) truly naive patients, ii) patients previously exposed to nucleosides but naive for NNRTIs and PIs, for whom the residual antiviral activity of NRTIs associated with PIs and NNRTIs could be considered to be low due to the extensive prior exposure to this class of drugs, and iii) patients previously exposed to one or more PI.

In a large randomized, open-label trial, Staszewski et al. compared three drug regimens: i) efavirenz plus indinavir, ii) efavirenz plus zidovudine and lamivudine and iii) indinavir plus zidovudine and lamivudine [26]. The indinavir dosage was increased from 800 mg three times a day to 1000 mg three times a day in the efavirenz plus indinavir group to compensate for the increased metabolism of indinavir in the presence of efavirenz. Patients had not previously been treated with lamivudine, an NNRTI or a PI. Of the patients, 85% were naive to any antiretroviral therapy. Baseline mean CD4 cell count was 345/mm^3 and mean baseline plasma HIV RNA was 4.77 log_{10} copies/ml. A total number of 450 patients were randomized between the three arms. According to an intention-to-treat analysis, the percentages of patients with plasma HIV RNA levels of <400 copies/ml at week 48 were 70% in the group assigned to efavirenz plus NRTIs, 53% in the group assigned to indinavir and efavirenz, and 48% in the group assigned to indinavir plus NRTIs. At week 48, mean increases of 201, 185 and 180 CD4 cells/mm^3 were found in the group given efavirenz plus NRTIs, the group given indinavir plus NRTIs and the group given efavirenz plus indinavir, respectively. The rate of discontinuation as a result of adverse events was significantly higher in indinavir plus NRTIs group than in either of the efavirenz groups. These adverse events were largely gastrointestinal. The incidence of central nervous system symptoms was similar in the two arms containing efavirenz: 58% in the group given efavirenz and NRTIs and 53% in the group given efavirenz plus indinavir. In this study, 200 mg capsules of indinavir were used. Thus, patients assigned to receive efavirenz and NRTIs had to take far fewer pills than the other patients (four pills of indinavir taken three times daily without food vs three pills of efavirenz taken once daily). The superior results of the arm without indinavir could therefore be in part due to better adherence of patients to the regimen. However, this mode of administrating indinavir is no longer used and results may have been different with the combination of ritonavir at 100 mg twice daily and indinavir, which allows a twice-daily administration with a reduced daily number of pills, without restriction on food. It was possible to conclude from this study that an NNRTI/PI combination was as effective as a triple-drug therapy combining a PI with two NRTIs.

The BIKS study [27] was an open-label, non-comparative trial evaluating the activity and toxicity of lopinavir/ritonavir (533/133 mg twice daily) and efavirenz (600 mg once daily) in 86 patients. Sixty-five patients (76%) were antiretroviral therapy-naive and nine out of the 21 pretreated patients had been previously exposed to a PI. After a median follow-up of 36 weeks, treatment was discontinued in 14 patients, including six for toxicity. Grade 3 or 4 increases in cholesterol and triglycerides were observed in 29 and 13 patients, respectively. At week 24, 87% of patients had HIV RNA <400 copies/ml and the increase in CD4 cell count was 162/mm^3 in the intention-to-treat analysis.

The combination of lopinavir/ritonavir and nevirapine was studied in 31 antiretroviral-treated patients with plasma HIV RNA <80 copies/ml during at least 9 months. Patients were switched to either lopinavir/ritonavir 400/100 mg plus nevirapine (16 patients) or lopinavir/ritonavir 400/100 mg plus the two previous NRTIs (15 patients) [28]. At 48 weeks, viral suppression
<table>
<thead>
<tr>
<th>Protease inhibitor</th>
<th>NNRTI</th>
<th>NNRTI</th>
<th>NNRTI</th>
<th>NNRTI</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Nevirapine</td>
<td>Delavirdine</td>
<td>Efavirenz</td>
<td>References</td>
</tr>
<tr>
<td>Indinavir</td>
<td>Indinavir decreases 28%, nevirapine no effect; increase indinavir 1000 mg every 8 h or add ritonavir; nevirapine standard dose</td>
<td>Indinavir increases &gt;40%, delavirdine no effect; indinavir 600 mg every 8 h; delavirdine standard dose</td>
<td>Indinavir decreases 31%, efavirenz no effect; increase indinavir 1000 mg every 8 h or add ritonavir; efavirenz standard dose</td>
<td>[14,17–19]</td>
</tr>
<tr>
<td>Ritonavir</td>
<td>Ritonavir decreases 11%, nevirapine no effect; both standard dose</td>
<td>Ritonavir increases 70–100%; delavirdine no effect; monitor ritonavir levels and toxicity</td>
<td>Ritonavir increases 18%, efavirenz increases 21%</td>
<td></td>
</tr>
<tr>
<td>Saquinavir</td>
<td>Saquinavir decreases 25%, nevirapine no effect; co-administration not recommended without ritonavir boosting</td>
<td>Saquinavir increases fivefold, delavirdine no effect; allows administration of saquinavir hard gel without ritonavir boosting</td>
<td>Saquinavir decreases 62%, efavirenz decreases 12%; co-administration not recommended without ritonavir boosting</td>
<td>[15]</td>
</tr>
<tr>
<td>Nelfinavir</td>
<td>Nelfinavir increases 10%; nevirapine no effect; both standard dose</td>
<td>Nelfinavir increases twofold; delavirdine decreases 50%</td>
<td>Nelfinavir increases 20%; both standard dose</td>
<td>[20–22]</td>
</tr>
<tr>
<td>Amprenavir</td>
<td>Potential decreases in amprenavir level</td>
<td>Amprenavir increases twofold; delavirdine decreases 60%</td>
<td>Amprenavir decreases 36%; increase amprenavir dose or add ritonavir; efavirenz standard dose</td>
<td>[13,16]</td>
</tr>
<tr>
<td>Lopinavir/ritonavir</td>
<td>Lopinavir Cmin decreases 55%; consider 533/133 mg twice daily in PI-experienced patients; nevirapine standard dose</td>
<td>Lopinavir levels expected to increase</td>
<td>Lopinavir blood AUC decreases 40%, efavirenz decreases 19%; increase lopinavir/ritonavir to 433/133 mg twice daily; efavirenz standard dose</td>
<td>[23,24]</td>
</tr>
<tr>
<td>Atazanavir</td>
<td>Atazanavir decreases 74%; add ritonavir and increase atazanavir dosage to 400 mg; efavirenz standard dose</td>
<td></td>
<td></td>
<td>[25]</td>
</tr>
</tbody>
</table>
was maintained in both arms. Mean lopinavir C\text{min} levels were similar between both arms at steady state conditions. The mitochondrial DNA/nuclear DNA ratio increased significantly at week 24, compared with inclusion, in the group of patients who interrupted NRTIs only. Triglycerides and LDL cholesterol levels were significantly higher at week 24 than at baseline in patients receiving two NRTIs and lopinavir/ritonavir, whereas HDL-cholesterol increased significantly at week 24 compared with baseline in patients receiving nevirapine and lopinavir/ritonavir. In this study, lopinavir/ritonavir plus nevirapine seemed to be as safe and potent as lopinavir/ritonavir plus two NRTIs. Additionally, this combination could be used as an appropriate regimen in order to improve the lipid profile and to avoid or reverse the NRTI-related mitochondrial toxicity.

The HIV-NAT 009 study was a single-arm, open-label study switching patients failing NRTIs to indinavir/ritonavir (800/100 mg) twice daily and efavirenz 600 mg once daily [29]. A total of 61 patients were enrolled. Baseline mean HIV RNA was 4.19 log\text{10} copies/ml and median CD4 169 cells/mm\text{3}. At week 72, 47 patients (77\%) had HIV RNA <50 copies/ml and the median CD4 169 cells/mm\text{3}. At week 72, 28 patients (71\%) were still on treatment with the study drug-related serious adverse events occurred in 24 patients, mainly hypertriglyceridaemia (14/28 events, 50\%). Mean total cholesterol/HDL-cholesterol did not significantly change. Six patients (10\%) had nephrolithiasis of whom three interrupted therapy. Median ALT halved after switch. This study shows that this combination provided an adequate response in these patients failing NRTIs and was reasonably tolerated. In this study, the effect on lipodystrophy of ceasing combination NRTI therapy was also assessed by a standardized questionnaire, physical examination, fasted laboratory measurements, and DEXA and CT scans of mid-abdomen and thigh at weeks 0 and 48 [30]. Mid-thigh subcutaneous fat, abdominal visceral fat and subcutaneous fat increased significantly as well as fat in trunk, legs and arms, suggesting that ceasing NRTIs and switching to a PI and NNRTI regimen leads to an improvement of lipoatrophy and that PIs, through a separate mechanism, may lead to lipoaccumulation.

Lopez-Cortes et al. evaluated a once-daily regimen of saquinavir-sg and ritonavir (1200/100 mg) in combination with efavirenz (600 mg) in 42 patients treated with two NRTIs plus a PI or NNRTI and who had adverse effects attributable to NRTIs [31]. Viral load was <50 copies/ml in 22 patients. All patients were PI-experienced and 28 patients were NNRTI-experienced, including nine patients with a detectable viral load under NNRTI (six on efavirenz, three on nevirapine), when treatment was switched. After 52 weeks, 30 patients (71\%) were still on treatment with HIV RNA <50 copies/ml with an additional 5\% (two patients) with 71 and 460 copies/ml, respectively. Five patients dropped out during follow-up even though they had an undetectable viral load. Seven patients had a virological failure after 6 months of treatment. The median increase in CD4 cells was 97 cells/mm\text{3} after 6 months and 215 cells/mm\text{3} after 12 months. Patients with previous virological failure on ≥2 PIs or previous virological failure on NNRTIs had an increased risk of virological failure with the study treatment. This combination was well tolerated. The median value of fasting triglycerides increased from 169 mg/dl at inclusion to 208 mg/dl at 1 year.

In conclusion, NNRTI/PI combinations have been found to be effective in several non-comparative studies. It seems important to use a ritonavir-boosted PI in order to suppress the decrease in PI plasma levels induced by nevirapine or efavirenz. Regimens combining efavirenz and ritonavir might be more deleterious on lipid parameters. Further comparative studies comparing the respective activity and toxicity of NNRTI/PI combinations and standard antiretroviral regimens containing NRTI backbones are needed before such dual therapy can be recommended in routine clinical practice.

**Double-PI combinations**

**In vitro data**

Combining two drugs that target the same active site may not necessarily result in positive interactions. In vitro studies looking at the interaction between PIs have generally found that the effects tend to be only additive, but a favourable interaction seems to exist between saquinavir and lopinavir [32,33]. However, the degree to which these data predict clinical experience is currently unknown.

Combining two PIs would theoretically increase the genetic barrier to resistance, but this advantage is reduced by the overlap among PI resistance patterns. Since some PIs have specific key mutations or share resistance mutations with a limited number of PIs (such as the 150V mutation for amprenavir and 150L for atazanavir), they may be more suitable for combination with another PI.

**Pharmacokinetic interactions**

PI boosting with low doses of ritonavir (100 mg twice daily) has become routine in clinical practice. For drugs that have a first-pass metabolism, such as saquinavir and lopinavir, the effect of low dose ritonavir is to boost C_{\text{max}}, C_{\text{min}} AUC and to modestly prolong the half-life (t_{1/2}). For drugs with reasonable bioavailability but short t_{1/2}, such as indinavir and amprenavir, the effect of ritonavir is predominantly on t_{1/2}, C_{\text{trough}} and AUC.
Utilizing two PIs at therapeutic exposures is increasingly being considered. These regimens are most commonly used in patients who are intolerant to NRTIs or against virus with extensive NRTI or NNRTI resistance. A better knowledge of pharmacokinetic interactions between approved PIs is helpful in assisting physicians in clinical practice in choosing drugs and doses when using two PIs with a booster agent. Double-boosted PI combinations have shown three different types of interactions: i) ritonavir concomitantly boosts both PIs without producing simultaneous interactions between them (for example, a combination of lopinavir/ritonavir (lopinavir/r) and saquinavir); ii) ritonavir boosts PI₁ and PI₂ and additionally, PI₁ boosts PI₂ and ritonavir (for example, a combination of ritonavir with atazanavir and saquinavir) – this is particularly interesting in the case of PI-resistant virus but may lead to an increased risk of toxicity; and iii) ritonavir boosts both PIs, but PI₂ induces metabolism of PI₁ and PI₁ induces metabolism of PI₂, resulting in reduced plasma concentrations of both drugs (for example, a combination of lopinavir/r and amprenavir), which may favour virological failure.

Different double-PI combinations studied

**Saquinavir–lopinavir/r.** Data collected from 45 patients receiving saquinavir–lopinavir/r at the 1000 mg/400 mg/100 mg twice-daily dosage showed that effective plasma levels of saquinavir and lopinavir were achieved. Saquinavir pharmacokinetic parameters did not differ from those obtained in a control group of patients treated with saquinavir/r at the 1000 mg/100 mg twice-daily dosage and lopinavir levels were similar to historical controls [34]. In contrast, ritonavir levels were significantly lower in the saquinavir–lopinavir/r than in the saquinavir/r-treated patients. Other studies are in agreement with these findings [35–37]. A possible explanation for this finding could be a pharmaco-enhancing effect of lopinavir on saquinavir.

**Atazanavir–saquinavir.** A pharmacokinetic study investigating the co-administration of saquinavir/r 1600 mg/100 mg and atazanavir 300 mg once daily in 18 HIV-positive patients demonstrated that the addition of atazanavir significantly increased the C_{\text{trough}}, C_{\text{max}} and AUC_{0–24h} of saquinavir [38]. Atazanavir levels were consistent with historical control data indicating that saquinavir did not affect atazanavir pharmacokinetics. The pharmacokinetic interaction between atazanavir and saquinavir, without ritonavir, has also been evaluated [39]. Although saquinavir exposure is increased in the presence of atazanavir, a regimen of saquinavir–atazanavir 1200 mg/400 mg once daily was insufficient to achieve appropriate saquinavir plasma concentrations.

**Saquinavir–(fos)amprenavir.** The combination saquinavir–amprenavir has been found to reduce saquinavir exposure even in the presence of ritonavir, suggesting an increase in the saquinavir/r dosage to 1400 mg/200 mg twice daily [40]. The co-administration of fosamprenavir with saquinavir/r 1000 mg/100 mg resulted in a modest decrease in saquinavir pharmacokinetic parameters that was more than compensated for by the addition of another 100 mg ritonavir twice daily [41]. The optimal combination for saquinavir–fosamprenavir/r is 1000 mg/700 mg/200 mg twice daily.

**(fos)amprenavir–lopinavir/r.** A complex, unfavourable interaction is observed when amprenavir is combined with lopinavir/r. In addition to decreased lopinavir/r concentrations after induction of CYP450 by amprenavir, lopinavir also decreases amprenavir concentration [42,43]. An additional boost of ritonavir 100 mg twice daily improved the virological response to the amprenavir and lopinavir/r combination, suggesting that addition of more ritonavir to the regimen compensated for the negative pharmacokinetic interaction between lopinavir and amprenavir [44].

**Nelfinavir–lopinavir/r.** Lopinavir pharmacokinetic parameters are decreased by the co-administration of nelfinavir [45]. If this combination is used, it seems appropriate to increase the lopinavir/r dosage to 533 mg/133 mg twice daily.

**Indinavir–lopinavir/r.** Lopinavir shows synergistic anti-HIV activity when combined with indinavir in vitro. It has been shown that lopinavir/r and indinavir had no negative drug–drug interactions [46]. Adding lopinavir/r to an indinavir-containing regimen did not affect indinavir exposure at steady state. Therefore, dose adjustments are unnecessary for either drug.

**Tipranavir and other PIs.** Tipranavir is a recently available PI that is interesting due to its activity on PI-resistant virus. Unfortunately, tipranavir is a known inducer of CYP450 enzymes and was recently shown to negatively affect the pharmacokinetics of co-administered PIs, even in the presence of low-dose ritonavir. In study BI 1182.51, tipranavir was added to saquinavir/r, lopinavir/r or amprenavir/r and pharmacokinetic analysis was performed with and without tipranavir. When tipranavir was added to each regimen, saquinavir, lopinavir and amprenavir exposure was reduced by 70%, 49% and 45%, respectively [47].

In conclusion, when combining two PIs in the presence of ritonavir, dose modifications of all components may be required. Access to a therapeutic drug monitoring service to confirm that appropriate drug exposures are
achieved is useful when using such regimens. Beyond pharmacokinetic considerations, results of recent resistance tests and knowledge of adverse-effect profiles are important when selecting drugs. Table 2 summarizes the interactions between boosted double PIs. These are applicable to regimens that do not include NNRTIs.

Clinical data

Some descriptive non-comparative studies have been performed that provide insight into quantifying clinical responses with boosted double-PI regimens. Most studies have been performed in pretreated subjects failing antiretroviral therapy who initiated a boosted double-PI combination as salvage therapy. In some studies, nucleoside analogues are associated with PIs but residual activity is limited by cross-resistance in heavily pretreated patients. Although these studies do not include a nucleoside-sparing regimen, some of them will be reported here since they contribute some information about the activity of double-PI combinations.

Pretreated patients. Combining saquinavir or indinavir with lopinavir/rt had the advantage that ritonavir has a ‘double-boosting’ function for both lopinavir and the second PI (saquinavir or indinavir).

In the LOPSAQ study, Staszewski et al. [48] evaluated the activity of the boosted double-PI regimen combination of lopinavir/rt and saquinavir without an RTI in patients who had no RTI option due to resistance or systemic toxicity. The study includes 121 patients and the 24-week data have been presented for the first 64 patients. Median baseline characteristics were as follows: HIV RNA 5.2 \( \text{log}_{10} \) copies/ml, CD4 cell count 168 cells/mm\(^3\), 6.7 years of antiretroviral therapy with a previous exposure to 10 drugs. Before starting the boosted double regimen, 59% of patients underwent a structured treatment interruption. At week 24, 52 (81%) patients were still on therapy, median viral load \( \text{log}_{10} \) was 2.1 and median CD4 count was 299 cells/mm\(^3\). Plasma concentrations of lopinavir and saquinavir were lower in non-responders. Higher CD4 count at baseline and fewer PI mutations in the last failing regimen were also associated with response to therapy.

In the CrixiLop study, patients with limited RTI options due to resistance or toxicity were switched to a lopinavir/rt 400 mg/100 mg plus indinavir 800 mg twice daily, with or without previous structured treatment interruption. Twenty-eight patients were studied. Median baseline characteristics were as follows: HIV RNA 5.2 \( \text{log}_{10} \) copies/ml, CD4 cell count 116 cells/mm\(^3\) and 7.1 years of antiretroviral therapy. Eighteen patients (64%) had a structured treatment interruption before starting the study treatment. The addition of lopinavir did not significantly affect the indinavir AUC under steady state conditions despite significantly lower ritonavir plasma levels. Eleven patients discontinued therapy due to intolerance (eight patients) or virological failure (three patients). At week 24, 17 patients (61%) remained on therapy, the median CD4 count was 186 cells/mm\(^3\) and the median viral load was 1.9 \( \text{log}_{10} \) copies/ml [49].

In the Puzzle (ANRS 104) study, patients who had failed multiple antiretrovirals were randomized to receive lopinavir/rt (400 mg/100 mg) and amprenavir (600 mg), with or without an additional boost of 200 mg ritonavir/day. Forty patients were randomized and 37 started treatment. Median baseline characteristics were as follows: HIV RNA 4.7 \( \text{log}_{10} \) copies/ml, CD4 cell count 207 cells/mm\(^3\) and seven PI resistance mutations. The average number of antiretrovirals taken prior to randomization was 7.7. At week 52, patients with an additional boost of ritonavir 200 mg/day had a larger median decrease in plasma viral load than patients without additional ritonavir (–2 vs –1.1 \( \text{log}_{10} \) copies/ml, respectively) and a higher incidence of undetectable viral load (39% vs 11% reaching less than 50 copies/ml). Mean increase in CD4 cell count was 156 versus 100 cells/mm\(^3\), respectively. Toxicity was common: 44% of patients with additional ritonavir and 33% of those without discontinued at least one PI. This study shows that, despite a negative pharmacokinetic interaction between lopinavir and amprenavir, combination of these two drugs in pretreated patients leads to

### Table 2. Summary of interactions between boosted double-Pis

<table>
<thead>
<tr>
<th>Regimen</th>
<th>Interactions</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>LPV/RTV + SQV</td>
<td>LPV ↔ SQV</td>
<td>[34–36]</td>
</tr>
<tr>
<td>SQV/RTV + ATV</td>
<td>SQV ↑ ATV</td>
<td>[38]</td>
</tr>
<tr>
<td>SQV/RTV + APV or FosAPV</td>
<td>APV or FosAPV ↓ SQV</td>
<td>[40, 41]</td>
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<tr>
<td>LPV/RTV + APV</td>
<td>LPV ↓ APV</td>
<td>[42–44]</td>
</tr>
<tr>
<td>LPV/RTV + NFV</td>
<td>NFV ↔ LPV</td>
<td>[45]</td>
</tr>
<tr>
<td>LPV/RTV + IDV</td>
<td>LPV ↔ IDV</td>
<td>[46]</td>
</tr>
</tbody>
</table>

LPV, lopinavir; RTV, ritonavir; SQV, saquinavir; APV, amprenavir; FosAPV, fosamprenavir; NFV, nelfinavir; IDV, indinavir; ATV, atazanavir.
a subsequent virological response when 200 mg ritonavir/day is added [44]. Some degree of divergence is seen between resistance patterns of lopinavir, amprenavir and saquinavir. Ritonavir boosting of these agents has been commonly employed and tolerated in clinical practice. Combinations of these agents may therefore represent a possibility for consideration in double-PI boosting.

**Naive patients.** Few studies are available on naive patients. The first study is rather old since it was initiated in April 1996, when the PI class became available [50]. This study evaluated the combination of full-dose ritonavir and saquinavir at different dosages in 141 patients: ritonavir 400 mg and saquinavir 400 mg, twice daily (arm A), ritonavir 600 mg and saquinavir 400 mg twice daily (arm B), ritonavir 400 mg and saquinavir 400 mg three-times daily (arm C) and ritonavir 600 mg and saquinavir 600 mg twice daily (arm D). Investigators were allowed to add up to two RTIs to the patient’s regimen at week 12 in case of failure to achieve or maintain HIV RNA ≤200 copies/ml. Median baseline characteristics were as follows: HIV RNA 4.6 log10 copies/ml and CD4 cell count 273 cells/mm3. Of the patients, 78% had been treated with RTIs but all were naive for PI. Twenty-two patients withdrew due to toxicity, the most common risk of long-term metabolic toxicity and the necessity of drug–drug interactions leading to a high pill burden, the most common cause of discontinuation being related to gastrointestinal symptoms. In the intention-to-treat analysis, the proportion of patients with plasma HIV RNA below 200 copies/ml at week 48 was 55% to 74% according to the arm of randomization. Overall, a median increase of 128 CD4 cells/mm3 was observed at week 48. Fourteen patients (10%) developed grade 3 or 4 liver toxicity, but the majority of these patients were in arm D. Sixteen patients (11%) developed hypertriglyceridaemia >15 g/l but no patient interrupted treatment due to hypertriglyceridaemia and no pancreatitis was observed. The durability of the antiviral activity of this combination was also reported [51]. Up to year 5, 56/66 patients (82%) in the study had a viral response <200 copies/ml with a median CD4 cell count increase of 381 cells/mm3 from baseline. These 5-year results showed that significantly fewer patients in the PI-only arm experienced symptoms of lipoatrophy [52]. These findings were confirmed by the Prometheus study, in which PI- and stavudine-naive patients were randomized between ritonavir/saquinavir (400 mg/400 mg) twice daily, with or without stavudine [53]. Lipodystrophy was reported in 29/175 (17%) patients during 96 weeks of follow-up, and was more frequent in patients randomized to ritonavir/saquinavir/stavudine (22/88; 25%) than in patients randomized to stavudine-sparing double-PI regimen (7/187 patients; 8%). Although the dosage of ritonavir of 400 mg twice daily is no longer used, these studies illustrate the value of double-PI treatment.

Hellinger et al. [36] evaluated the boosted double regimen using saquinavir 1000 mg plus lopinavir/r 400 mg/100 mg twice daily in 20 PI-naive patients. Median baseline characteristics were as follows: HIV RNA 4.4 log10 copies/ml, CD4 cell count 274 cells/mm3 and lack of PI resistance mutations. Gastrointestinal intolerance in six patients (30%) led to dose reduction. Four patients discontinued the study due to adverse events or difficulty in adherence and two required tenofovir intensification after week 12. Intention-to-treat analysis at week 48 showed that 70% of patients had fewer than 400 copies/ml and 65% fewer than 50 copies/ml. The median increase in CD4 cell count was 194 cells/mm3. Lipid levels increased in most subjects. Lipoatrophy was detected in only one patient but fat accumulation was observed in 67% of subjects, central fat accumulation being the most frequent. Trough levels of saquinavir and lopinavir were appropriate. Table 3 summarizes studies of RTI-sparing boosted double-PI regimens.

In conclusion, the use of boosted double-PIs is a means of avoiding NRTI-containing regimens and seems effective in view of preliminary studies. However, the use of these combinations may be limited by drug–drug interactions leading to a high pill burden, the risk of long-term metabolic toxicity and the necessity for food restriction due to the poor digestive absorption of PIs compared with RTIs. Efforts have been made to improve some PI formulations, such as the development of fosamprenavir, the prodrug of amprenavir that reduces the number of pills fourfold. In any case, double-PI boosting has to be compared with the current standard of care in clinical trials before being considered as a valuable option.

**PI monotherapy**

In theory, single-agent therapy ought to reduce the cost and minimize adverse drug reactions of treatment. Although this type of treatment was refuted in the initial era of AIDS therapy due to its lack of efficacy, the availability of potent drugs with a high genetic barrier has led care providers to reconsider this concept. Lopinavir/r appeared as an appropriate candidate to test this strategy. In a proof-of-concept study, Gathe et al. [54] reported an open-label study of lopinavir/r monotherapy in treatment-naive subjects. Thirty treatment-naive patients with a mean CD4 cell count of 170 cells/mm3 and a mean plasma HIV RNA of 5.42 log10 copies/ml were enrolled. Subjects received lopinavir/r on an open-label basis for up to 48 weeks. In cases of virological failure, therapy could be intensified with the addition of tenofovir, lamivudine and saquinavir. After 48 weeks, 20 subjects (60%) remained on the study, all of whom had their viral load...
### Table 3. Summary of studies of NRTI-sparing boosted double-PI regimens

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Regimen</th>
<th>Study type and regimen</th>
<th>Virological outcome</th>
<th>CD4 cell count response</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cameron [50]</td>
<td>141</td>
<td>Ritonavir 400 mg/saquinavir 400 mg twice daily</td>
<td>Randomized study</td>
<td>55–78% of patients &lt;200 copies/ml at week 48 (ITT)</td>
<td>Median increase of 128 CD4 cells/mm³ at week 48</td>
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<tr>
<td></td>
<td></td>
<td>Ritonavir 600 mg/saquinavir 400 mg twice daily</td>
<td>Pl-naive patients</td>
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<tr>
<td></td>
<td></td>
<td>Ritonavir 400 mg /saquinavir 400 mg three-times daily</td>
<td>RTI intensification allowed at week 12</td>
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<tr>
<td></td>
<td></td>
<td>Ritonavir 600 mg/saquinavir 600 mg twice daily</td>
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<tr>
<td>Hellinger [36]</td>
<td>20</td>
<td>Saquinavir1000 mg/lopinavir/r 400 mg/100 mg twice daily</td>
<td>Non-randomized observational cohort</td>
<td>70% of patients &lt;400 copies/ml and 65% of patients &lt;50 copies/ml at week 48 (ITT)</td>
<td>Median increase of 194 CD4 cells/mm³ at week 48</td>
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<tr>
<td></td>
<td></td>
<td>Pl-naive patients</td>
<td></td>
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<tr>
<td>LOPSAQ</td>
<td>121</td>
<td>Lopinavir/r/saquinavir</td>
<td>Non-randomized observational cohort</td>
<td>Median 3.1 log₁₀ viral load reduction at week 24</td>
<td>Median increase of 131 CD4 cells/mm³ at week 24</td>
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<tr>
<td>Staszewski [48]</td>
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<td>Pl-heavily experienced patients (median: three PIs)</td>
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<tr>
<td></td>
<td></td>
<td>Previous STI in 59% of patients</td>
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<tr>
<td>CRIXILOP</td>
<td>28</td>
<td>Lopinavir/r/indinavir</td>
<td>Non-randomized observational cohort</td>
<td>Median 3.3 log₁₀ viral load reduction at week 24</td>
<td>Median increase of 70 CD4 cells/mm³ at week 24</td>
</tr>
<tr>
<td>Staszewski [49]</td>
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<td>Pl-heavily experienced patients (median three PIs)</td>
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<tr>
<td></td>
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<td>Previous STI in 64% of patients</td>
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<tr>
<td>Puzzle</td>
<td>37</td>
<td>Lopinavir/r/amprenavir ± additional ritonavir</td>
<td>Randomized study</td>
<td>Median 2.0 and 1.1 log₁₀ viral load reduction at week 52 and 39% and 11% of patients &lt;50 copies/ml in additional ritonavir and non-additional ritonavir arms, respectively</td>
<td>Mean increase of 156 and 100 CD4 cells/mm³ at week 52 in additional ritonavir and non-additional ritonavir arms, respectively</td>
</tr>
</tbody>
</table>

ITT, intention-to-treat; STI, structured treatment interruption.
suppressed to levels below 50 copies/ml. The remaining 10 subjects were excluded from data analysis due to loss to follow-up, poor adherence, gastrointestinal intolerance or other issues. There were only two cases of virological failure, neither of which exhibited resistance to lopinavir. These results are promising but further studies are necessary to determine the potential of such a strategy. Although potent, lopinavir/r monotherapy may be insufficient to fully suppress patients with a high viral load. Other studies with lopinavir/r monotherapy, in particular in the context of maintenance therapy, are ongoing.

New classes of drugs

The new class of entry inhibitors includes drugs that target one of the following steps: CD4 receptor attachment, chemokine receptor attachment (using either the CXCRe4 or CCR5 cell surface receptor) and viral–cell membrane fusion. The first entry inhibitor currently available in the treatment of HIV-1 infection is enfuvirtide, a fusion inhibitor. Its cost and the need for subcutaneous administration have contributed to the delayed use of this drug, which proved to be effective in two large Phase III trials in pretreated patients failing antiretroviral therapy [55,56]. It is unlikely that enfuvirtide will be used as part of a first-line therapy instead of orally available classes of drugs. However, some compounds that are orally available, such as some CCR5 inhibitors, are under investigation and may be part of initial therapy if ongoing trials confirm that they are efficient and well tolerated.

In conclusion, NRTI-sparing regimens may be considered because of NRTI resistance and NRTI toxicity. The two main strategies, NNRTI/PI combination and boosted double-PI regimen, remain relatively complex due to the presence of pharmacokinetic interactions requiring often complicated dose adjustments. Preliminary studies suggest that these combinations are efficient and well-tolerated. However, the lack of comparative studies evaluating these combinations versus standard recommended regimens precludes using them as first-line therapies in clinical practice until further information is available. Single PI treatment is another alternative, but again further studies are necessary to validate this strategy. Finally, compounds from new classes, in particular orally available drugs such as some CCR5 inhibitors, may enhance the panel of therapeutic options in the next few years.

References

Nucleoside-sparing regimens


