*Acta virol.* **37**: 377-387, 1993

# PREPARATION AND CHARACTERIZATION OF MONOCLONAL ANTIBODIES DIRECTED AGAINST GLYCOPROTEINS OF BOVINE LEUKAEMIA VIRUS

O. ORLÍK, J. BÁN, E. GIECIOVÁ, V. ALTANEROVÁ, Č. ALTANER

Department of Molecular Virology, Cancer Research Institute, Špitálska 21, Bratislava 812 32, Slovak Republic

*Received December 11, 1992; revised February 10, 1993*

*Summary.* – Six monoclonal antibodies (MoAbs) directed against glycoproteins of bovine leukaemia virus (BLV) were prepared and characterized. Comparison of these MoAbs with anti-gp51 MoAbs of known epitope specificity by competition antibody binding assay allowed to distinguish two new conformational epitopes C1 and C2 on the molecule of gp51. The epitope C1 is involved in the process of inhibition of formation of syncytia but not in neutralization of VSV/BLV pseudotypes. Three newly prepared MoAbs were directed against known epitopes F, G and H, and their neutralizing activities of biological functions of gp51 were determined. MoAbs BLVgp30-94C11 which was directed against transmembrane glycoprotein gp30 was found not to be involved in neutralization of VSV/BLV pseudotypes and did not inhibit formation of syncytial cells as well.

*Key words: bovine leukaemia virus; monoclonal antibodies; epitope mapping; envelope proteins gp51, gp30*

## *Introduction*

BLV, an exogenous retrovirus distantly related to the human T-cell lymphotropic viruses is the etiological agent of enzootic bovine leukaemia of cattle (for review see Burny *et al.*, 1990). High antibody titers to viral proteins (glycoproteins gp51 and gp30 included) have been found in infected animals. The presence of virus-specific antibodies in infected animals is usually employed for immunodiagnosis of BLV infection. The envelope glycoproteins of retroviruses play a crucial role in the virion life cycle and are responsible for cellular tropism of the virus.

The BLV envelope glycoprotein is synthesized as a 72 K precursor which is processed into the 51 K (gp51) and 30 K (gp30) mature forms, gp51 is the external subunit containing the cell receptor-binding domain, and gp30 is the transmembrane subunit that anchors the gp51-gp30 complex in the viral

envelope. This BLV envelope glycoprotein complex is essential to infection and syncytium induction (Bruck *et al.*, 1982*a*; Voneche *et al.*, 1992). Several MoAbs against gp51 were already prepared and characterized (Bruck *et al.*, 1982*a, b*). The panel of MoAbs has allowed the definition of eight distinct antigenic sites (A to H) and three overlapping sites (B', D' and F'). The conformational epitopes (F, G and H) have been localized to the amino-terminal part of gp51 (Portetelle *et al.*, 1989*a*), and represent major determinants involved in the biological activities of the virus (infectivity and syncytium induction). Attempts to precisely localize these conformational epitopes have been only partially successful (Portetelle *et al.*, 1989*a*; Mamoun *et al.*, 1990). However, sequential epitopes A to E have been recently localized to the C-terminal half of native and recombinant gp51 as well (Callebaut *et al.*, 1991; Bán *et al.*, 1992).

In this report we describe the preparation of MoAbs with the intention of identifing naturally occurring immunodominant epitopes on the molecules of gp51 and gp30 of BLV. Six MoAbs were prepared and further characterized.

## *Materials and Methods*

*Cells and virus.* The cell clone derived from mouse myeloma cells Sp2/O-Ag14 was used as a fusion partner for hybridoma production (Schulman *et al.*, 1978). The lymph nodes were used as a source of antibody-producing cells. A single cell suspension of lymphocytes was obtained from lymph nodes by teasing them through a teflon-coated screen (filter holedr Millipore) using a rubber cell scraper. The suspension was washed three times with culture medium and collected by centrifugation (800 × g, 5 mins). The basal medium used was Dulbecco's modified Eagle's medium (DMEM) supplemented with glutamine (10 mmol/l), and selected horse serum (10–15 %).

For bovine leukaemia virus production, the cells of a highly virus-productive cell clone were used (Altaner *et al.*, 1985). The cell clone was originally derived from lamb kidney cell line FLK permanently infected with BLV (Van der Maaten and Miller, 1976). Tissue culture medium containing 1 % of foetal bovine serum harvested from 3-day old roller cultures at confluence was filtered through 0.45 $\mu$m Millipore membrane and used as a non-purified BLV preparation. Purification of the virus was performed by sucrose gradient centrifugation from tissue culture fluid harvested from roller cultures. The virus-containing medium was disintegrated by ultrasound (twice for 10 seconds at the highest output of 100 W MSE ultrasound generator) or by addition of Tween-20 to a final concentration of 0.5 %.

*Preparation of MoAbs.* Hybridoma cells producing anti-BLV MoAbs were prepared by the usual hybridoma technology. Some modifications described in details elsewhere (Orlík and Altaner, 1988) were used to simplify the procedure and increase the yield of antibody producing cells. For immunization the whole non-disrupted viral particles were used. Lymphocytes from lymph nodes were used for fusion with Sp2/O-Ag14 myeloma cells.

*Production and purification of MoAbs.* For mass production of MoAbs, about $1 \times 10^7$ hybridoma cells were injected intraperitoneally in Pristane-primed BALB/c mice, and ascitic fluid was collected from the mice 10–14 days later. The MoAbs from ascitic fluid were purified by affinity chromatography on Protein A-Sepharose or Protein G-Sepharose gel columns (Pharmacia) according to the manufacturer's recommendations. The concentration of proteins in solution of purified immunoglobulins was determined according to Lowry. Isotyping of monoclonal immunoglobulins was performed by double diffusion in 1 % agarose gel with monospecific anti-isotype goat antisera (Sigma).

*Enzyme-linked immunosorbent assay (ELISA).* The titer of purified MoAbs was detected by ELISA. The surface of wells of microtiter plates was coated with purified disrupted virus preparation.

Coating was performed overnight at 4 °C using the antigen dissolved in 0.2 mol/l bicarbonate buffer, pH 9.6. The adsorbed MoAbs tested were detected by the secondary antibody conjugated with peroxidase. Bound peroxidase was revealed by addition of the chromogenic substrate ortho-phenylenediamine (1.0 mg/ml in 0.05 mol/l citrate-phosphate buffer, pH 4.5) in the presence of 0.01 % hydrogen peroxide.

*Antigen capture assay* was performed in flat bottom high binding 96-well microtiter plates serving as the solid phase in ELISA. The wells were coated by overnight incubation with immunoglobulin of particular MoAb (10 μg/ml) in Tris-EDTA-saline (TEN), pH 7.5 at 4 °C. The polystyrene surface was then saturated with TEN containing 0.5 % bovine serum albumin (BSA) at 37 °C for 2 hrs. The wells were washed three times with TEN containing 0.05 % Tween-20 (washing buffer). Such plates were ready for use in the antigen capture assay. As a source of antigen, the medium from a virus-producing cell was used. The plates were incubated overnight at 4 °C with the virus containing medium and viral proteins were captured. Sera which were used for detection of captured viral proteins were diluted with TEN containing 0.01 % BSA, 0.1 % Tween-20, and 2 % glycerol (dilution buffer) and added to the plates. Incubation was performed at 37 °C for 1 hr. The plates were washed with washing buffer and peroxidase-labelled secondary antibodies of appropriate dilution were added. The incubation at 37 °C for 1 hr was followed by washings and immunoreaction was developed with chromogenic substrate (ortho-phenylene-diamine).

*Protein blot analysis.* The proteins were separated by sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE) according to Laemmli (1970). The proteins were transferred to nitrocellulose membranes using a semi-dry electroblotter (Kyhse-Andersen, 1984). After blocking nonspecific binding sites with 5 % skimmed milk in PBS containing 0.05 % Tween-20 for 20 mins at room temperature, the nitrocellulose strips were incubated for 60 mins with appropriate serum or MoAbs. This procedure was followed by another incubation at 37 °C for 1 hr with the second enzymatically-labelled antibody. Between incubations, the membrane was washed extensively with PBS containing Tween-20 (0.05 %). Immunoreactive bands were then visualized by reaction with chromogenic substrate (4-chloro-1-naphtol).

*Cell labelling and immunoprecipitation.* Subconfluent cultures of tested cells were labelled for 2 hrs at 37 °C in a methionine-free medium containing 10 % foetal calf serum and $^{35}$S-methionine (250 μCi/ml). Cells were washed twice with STE (100 mmol/l NaCl, 10 mmol/l Tris-HCl pH 8.0, 1 mmol/l EDTA pH 8.0). The labelled cells were lysed in RIPA buffer (150 mmol/l NaCl, 10 mmol/l Tris-HCl pH 8.0, 1 % Triton X-100, 0.5 % sodium deoxycholate, 0.1 % SDS). All buffers contained 1 mmol/l phenylmethylsulphonylfluoride (PMSF) as a protease inhibitor. After clarification by centrifugation (100 000 xg, 1 hr), the radioactivity of cell extracts was measured, and aliquots of cell lysates ($10^7$ cpm) were immunoprecipitated with appropriate antisera. After incubation (60 mins at 4 °C), 30 μl of 50 % suspension of Protein A-Sepharose (Pharmacia) was added and the mixture was mixed by rotation for another 1 hr at 4 °C. After two washings with NET-NON (650 mmol/l NaCl, 5 mmol/l EDTA, 50 mmol/l Tris-HCl pH 8.0, 0.5 % NP40, 0.1 % ovalbumine) and two washings with NET-N (NET-NON without ovalbumine and 0.5 mol/l NaCl), the Protein A beads were resuspended in a sample buffer (20 % glycerol, 10 % mercaptoethanol, 4 % SDS, 125 mmol/l Tris-HCl pH 6.8, 0.1 % bromophenol blue), heated at 100 °C for 3 mins and electrophoresed (SDS-PAGE, 12 %).

*Labelling of antibodies.* The immunoglobulins were labelled with $^{125}$I by means of 1,3,4,6-tetrachloro-3-alpha, 6-alpha-diphenyl glycoluril (IODO-GEN, Pierce) (Franker and Speck, 1978).

*Competition antibody binding assay.* Wells of microtiter plates were coated with purified disrupted BLV particles as described in the ELISA. Excess of unlabelled purified MoAbs in 100 μl of dilution buffer was added to each well. The plates were incubated overnight at 4 °C. The solution of $^{125}$I-labelled MoAb (100 μl; 150 000 cpm/well) was added into each well and plates were incubated at 37 °C for two hrs. The plates were than washed three times with washing buffer and the bound radioactive material was dissolved in 1 mol/l potassium hydroxide and measured by using a LKB Clini-gamma counter.

*Syncytia induction assay* was performed as described by Ferrer and Diglio (1976) and Ferrer and Cabradilla (1978). Briefly, CC81 indicator cells (murine sarcoma virus infected cat cells) were seeded in 60 mm Falcon dishes at a concentration of $10^5$ cells/dish. Twenty four hrs later, the medium was removed and 2 ml of the tested medium containing 5 μg Polybrene/ml were added to each dish. After 6–8 days the cells were washed, fixed with a mixture of acetone-methanol (1:1), stained with

Giemsa and syncytia containing five or more nuclei were counted.

*Virus neutralization test.* The vesicular stomatitis virus (VSV)-BLV pseudotype (VSV/BLV) neutralization test was performed using HeLa cells as described by Závada *et al.* (1979). Briefly, VSV/BLV pseudotypes were recovered from a culture of VSV- and BLV-infected foetal lamb spleen cells. Tissue culture fluid (0.5 ml) was mixed with 0.1 ml of sheep anti-VSV immunoglobulins. This mixture was diluted to 100 ml with PBS containing 1 % inactivated foetal calf serum and 30 mg/l DEAE-dextran. Examined samples were diluted with PBS and 0.1 ml aliquots were mixed with 1 ml of the pseudotype preparation, then incubated at 37 °C for 60 mins, and plated on a Petri dish with HeLa cells. The number of plaques was counted after 2–3 days.

## *Results*

### *Specificity of prepared MoAbs*

To prepare MoAbs directed against naturally dominant antigenic regions of the BLV envelope glycoproteins, the immunization of mice with a non-denatured form of the glycoproteins (whole viral particles) was performed. Five hybridomas permanently producing MoAbs directed against envelope glycoprotein gp51 and one hybridoma producing MoAb with anti-gp30 specificity were prepared. Some of their properties are shown in Table 1. The MoAbs reacted in radioimmunoprecipitation with lysates of BLV-producing cells. The anti-gp51 directed antibodies were able to detect env precursor Pr72 and processed gp51. The reactivity of anti-gp30 MoAb in radioimmunoprecipitation was weak; the antibody precipitated gp30 only, and no precursor was detected. Reaction of some of these MoAbs is shown in Fig. 1. None of these MoAbs were able to react with denatured forms of these antigens in Western blot analysis (data not shown). Therefore, the MoAbs were directed against conformational epitopes.

**Table 1. MoAbs directed against glycoproteins of BLV**

| MoAb | Isotype | Source of antigen for immunization | Reactivity | |
|---|---|---|---|---|
| | | | Radioimmuno-precipitation | Western blot analysis |
| BLVgp51-6A12 | IgG1 | virus | gp51 | – |
| BLVgp51-4E9/32A6 | IgG2b | virus | gp51 | – |
| BLVgp51-96H10B12 | IgG1 | virus | gp51 | – |
| BLVgp51-25E11/2 | IgG2b | virus | gp51 | – |
| BLVgp51-X30 | IgG1 | virus | gp51 | – |
| BLVgp51-B19[a] | IgG1 | gp51 | gp51 | gp51 |
| BLVgp30-94C11 | IgG1 | virus | gp30 | – |

[a]MoAb BLV-gp51-B19 was prepared by Dr. M. Merza, University of Upsalla, Sweden, and was kindly provided for further characterization.

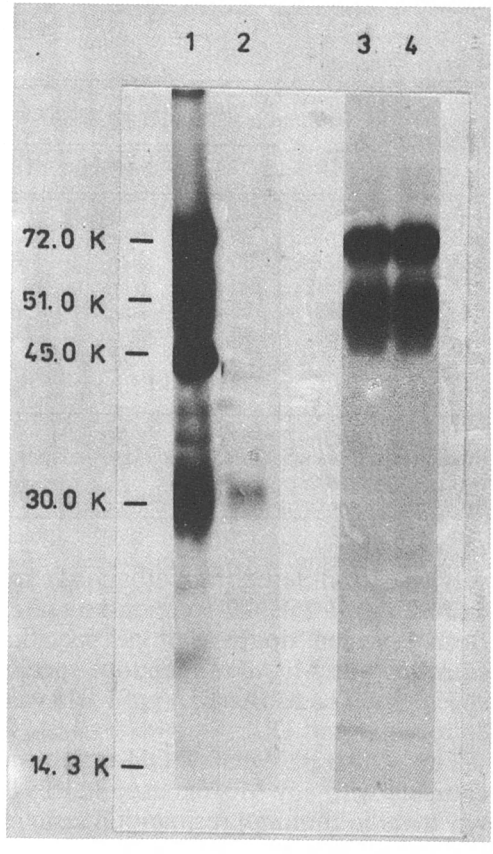


**Fig. 1**
Radioimmunoprecipitation of MoAbs with lysate from BLV-producing cells
Anti-whole BLV polyspecific serum (lane 1), BLVgp30-94C11 (lane 2), BLVgp51-6A12 (lane 3), BLVgp51-25E11/2 (lane 4).

*Epitope mapping of anti-gp51 BLV MoAbs*

Epitope specificity of the MoAbs was determined by competition antibody binding assay. All combinations among MoAbs prepared were tested (Table 2). It was found that MoAbs BLVgp51-4E9/32A6 and BLVgp51-25E11/2 were able to compete with each other and probably were directed against the same epitope. Any other substantial competition among other MoAbs was not found, suggesting that each MoAb recognized a different epitope on gp51. The MoAb BLVgp30-94C11 with anti-gp30 specificity did not compete with any of anti-gp51 MoAb tested, as was expected.

To classify epitope specificity of prepared MoAbs, their comparison with MoAbs of known epitope specificity was performed (Table 3). Competition experiments allowed to distinguish two new epitopes on gp51, designated C1 and C2 (defined by MoAbs BLVgp51-96H10B12 and BLVgp51-X30, respecti-

**Table 2. Relationship of antibodies against BLV glycoproteins among themselves detected by competition assay**

| Cold antibody | Radiolabelled MoAbs BLVgp51- (% of binding) | | | | | | |
|---|---|---|---|---|---|---|---|
| | -6A12 | -4E9/32A6 | -96H10B12 | -25E11/2 | -X30 | -B19 | BLVgp30-94C11 |
| BLVgp51-6A12 | 30 | 100 | 100 | 130 | 90 | 95 | 80 |
| BLVgp51-4E9/32A6 | 130 | 30 | 90 | **30** | 80 | 90 | 90 |
| BLVgp51-96H10B12 | 120 | 60 | 35 | 60 | 70 | 75 | 80 |
| BLVgp51-25E11/2 | 100 | **40** | 90 | 30 | 85 | 90 | 95 |
| BLVgp51-X30 | 90 | 80 | 80 | 100 | 30 | 85 | 100 |
| BLVgp51-B19 | 100 | 85 | 90 | 85 | 90 | 20 | 90 |
| BLVgp30-94C11 | 90 | 100 | 85 | 90 | 75 | 80 | 35 |

Binding of radiolabelled MoAbs to plastic-adsorbed BLV antigens in absence of competing antibody was taken as 100 %.
Bold values represent significant competition.

vely), which differed from all already known ones. MoAbs BLVgp51-4E9/32A6 and BLVgp51-25E11/2 were of the same epitope specificity as mentioned above. Both revealed double-epitope specificity F–H, because they were able to compete with MoAb of F epitope specificity as well as with MoAb of H epitope specificity. The MoAb BLVgp51-B19 was found to be directed against sequential epitopes D and D'.

The MoAb BLVgp30-94C11 which was found to be anti-gp30 specific did not compete with any MoAbs used in this test. Because of its weak immunoreactivity in radioimmunoprecipitation assay, two additional experiments were performed which indirectly confirmed its specificity. At first, this antibody did not bind to p24 and gp51, and it did not compete with eleven anti-p24 BLV MoAbs in binding to p24 (data not shown). At second, this MoAb was suitable to capture an antigen from the medium of virus-producing cells. This captured antigen could be detected by polyclonal anti-BLV serum (containing antibodies against gp30; see Fig. 1) but not with monospecific antisera directed against p24 or gp51 (Table 4).

*Biological activities of prepared MoAbs*

To further characterize the prepared MoAbs, their ability to neutralize the virus infectivity and to inhibit the formation of virus-induced syncytia was tested. Results of these experiments are shown in Table 5.

MoAbs BLVgp51-4E9/32A6 and BLVgp51-25E11/2 with epitope specificity F–H and MoAb BLVgp51-6A12 with G epitope specificity were found to posses virus-neutralizing activity. This concurs with the observation that epitopes F, G and H are involved in virus neutralization (Bruck *et al.*, 1982*a*).

**Table 3. Epitope specificity of anti-gp51 MoAbs detected by competition assay with MoAbs of known specificity**

| Cold antibody epitope | Radiolabelled MoAbs BLVgp51- (% of binding) | | | | | | |
|---|---|---|---|---|---|---|---|
| | -6A12 | -4E9/32A6 | -96H10B12 | -25E11/2 | -X30 | -B19 | BLVgp30-94C11 |
| Autocompetition | 30 | 30 | 35 | 25 | 30 | 20 | 30 |
| A[a] | 90 | 90 | 110 | 100 | 90 | 85 | 90 |
| B | 90 | 90 | 90 | 100 | 100 | 90 | 85 |
| B' | 100 | 85 | 95 | 100 | 85 | 110 | 100 |
| C | 140 | 65 | 100 | 90 | 65 | 85 | 90 |
| D | 100 | 80 | 100 | 120 | 90 | **35** | 85 |
| D' | 90 | 95 | 80 | 110 | 90 | **25** | 90 |
| E | 90 | 70 | 100 | 90 | 80 | 80 | 80 |
| F | 110 | **40** | 110 | **30** | 85 | 90 | 80 |
| G | **35** | 100 | 90 | 90 | 80 | 95 | 90 |
| H | 140 | **40** | 90 | **40** | 80 | 100 | 85 |

Binding of radiolabelled MoAbs to plastic-adsorbed BLV antigens in absence of competing antibody was taken as 100 %. Bold values represent significant competition.
[a]MoAbs of known anti-gp51 epitope specificity (A–H) were kindly provided by Dr. D. Portetelle.

**Table 4. Indirect evidence for anti-gp30 specificity of MoAb BLVgp30-94C11**

| Antisera | MoAbs adsorbed | | |
|---|---|---|---|
| | BLVgp30-94C11 | BLVgp51-6A12 | BLVgp24-X48 |
| anti-whole BLV | ++ | +++ | +++ |
| anti-p24 BLV | - | - | +++ |
| anti-gp51 BLV | - | +++ | - |

An antigen capture assay was performed as described in *Materials and Methods*. The wells of microtiter plates were coated with particular MoAb and disrupted viral particles were added. As a source of viral antigens tissue culture medium from BLV-producing cells was used.
(+++) absorbancy higher than 0.6
(++) absorbancy higher than 0.4
(-) absorbancy lower than 0.15

**Table 5. Neutralizing activities of prepared MoAbs**

| MoAbs | Epitope specificity | VSV/BLV pseudotype neutralization[a] | Syncytium induction assay[b] |
|---|---|---|---|
| BLVgp51-6A12 | G | 5000 | 0 (100 %) |
| BLVgp51-4E9/32A6 | F,H | 3000 | 94 (90 %) |
| BLVgp51-96H10B12 | C1 | 0 | 550 (44 %) |
| BLVgp51-25E11/2 | F,H | 2000 | 31 (97 %) |
| BLVgp51-X30 | C2 | 0 | 870 (12 %) |
| BLVgp51-B19 | D | 0 | 900 (10 %) |
| BLVgp30-94C11 | alpha | 0 | 858 (13 %) |
| BLVp24-X12 | BLVp24-IDR2 | 0 | 986 (0 %) |

[a]Values represent end point titer.
[b]Values represent number of syncytia per well and percentage (in parentheses), respectively. Non-neutralizing, unrelated MoAb BLV-p24-X12 served as control.

The MoAbs BLVgp51-96H10B12 and BLVgp51-X30 which defined two new epitopes C1 and C2 were not found to be virus-neutralizing in the pseudotype test, but MoAb BLVgp51-96H10B12 which recognized epitope C1 was able to inhibit partially the induction of syncytia formation. The MoAb BLVgp30-94C11 did not neutralize either the VSV/BLV pseudotypes or the formation of syncytia.

## *Discussion*

MoAbs and their use in epitope mapping is an experimental approach to look for immunodominant regions on native proteins. In preparation of MoAbs against viral proteins, the following propositions have been taken into consideration. Antibodies which are induced by natural virus infection could differ from antibodies obtained by immunization with purified viral proteins. The formers are directed preferentially against naturally immunodominant epitopes on the antigen, and usually possess virus-neutralizing activity. On the other hand, purification of protein antigens is often connected with changes in their three-dimensional structure, and the antibody response, instead of being directed against the conformational epitopes, is directed mainly against sequential and/or cryptogenic epitopes.

This was found true for BLV, where natural infection with BLV in cattle is leading to induction of virus-neutralizing antibodies which are directed against conformational epitopes F, G and H of viral envelope glycoprotein gp51 (Bruck *et al.*, 1984). When purified viral glycoprotein gp51 was used for immunization in

preparation of MoAbs, the identification of MoAbs against sequential epitopes of the glycoprotein was preferentially observed (Bruck *et al.*, 1982*a*) or exclusively (Platzer *et al.*, 1990).

Therefore, to prepare MoAbs directed against naturally dominant epitopes of viral glycoproteins, immunization with whole viral particles was performed. As expected, the form of antigen used for immunization substantially influenced the epitope specificity of obtained MoAbs. Six MoAbs were prepared against the envelope glycoproteins, and all of them displayed reactivity against conformational epitopes only.

MoAbs BLVgp51-96H10B12 and BLVgp51-X30 allowed to distinguish two new conformational epitopes C1 and C2 on gp51. Their localization on the molecule gp51 is not known, but from results of the VSV/BLV neutralization test it could be deduced that they are not localized in that part of the molecule, which is important for this biological activity.

MoAbs BLVgp51-4E9/32A6 and BLVgp51-25E11/2 are similar and display double-epitope specificity F–H. There are at least two possibilities to explain this finding: (a) the epitope recognized by these antibodies is localized between epitopes F and H and overlaps parts of both; (b) these MoAbs after binding to their epitope initiate conformational changes in the gp51 molecule which might result in alteration of F and H conformational epitopes. This can diminish following binding of anti-F and anti-H directed MoAbs. This explanation can be supported by the observation that epitopes F, G and H are localized at the $NH_2$ part of the gp51 polypeptide chain (Portetelle *et al.*, 1989*a*), and in the three-dimensional model of gp51 they are localized on the same side of the molecule gp51 (Mamoun *et al.*, 1990).

MoAbs BLVgp51-6A12, BLVgp51-4E9/32A6 and BLVgp51-25E11/2 which were directed against epitopes F–H and G were virus-neutralizing in the VSV/BLV pseudotype test and were also able to neutralize the formation of syncytia. This observation concurs with results obtained with MoAbs previously characterized (Bruck *et al.*, 1982*b*); only the neutralizing capacity of our MoAbs was found to be higher.

However, MoAb BLVgp51-96H10B12 defining epitope C1, despite its inability to neutralize VSV/BLV pseudotypes, was able to partially block the formation of syncytial cells. It seems that not only epitopes F, G and H, but also epitope C1 is involved in neutralization of syncytia formation. This is in contrast to the VSV/BLV pseudotype neutralization test, where the epitope C1 did not participate in neutralization of virus infectivity.

MoAbs BLVgp30-94C11 directed against gp30 was not found to be neutralizing in the VSV/BLV pseudotype test and did not inhibit formation of syncytia. These results have shown that an epitope recognized by this MoAb is not involved in biological functions of this molecule despite the fact that glycoprotein gp30 plays an important role in the process of formation of syncytia (Voneche *et al.*, 1992). A different effect was observed with MoAbs directed against transmembrane protein gp41 of HIV, where these MoAbs revealed

enhancing effects on HIV-1 infection (Niedrig *et al.*, 1992).

MoAbs which are directed against neutralizing epitopes F–H and G of gp51 are very suitable candidates for preparation of antiidiotypic antibodies, which should serve as an antiidiotypic vaccine against BLV infection.

*Acknowledgements.* The authors are indebted to Dr. D. Portetelle for providing anti-gp51 MoAbs of known epitope specificity, to Dr. M. Merza for MoAb B19, and Dr. J. Závada for VSV/BLV pseudotype. The authors would like to thank also Mrs. M. Zafková, Mrs. B. Puterová, and Mrs. H. Chorvátová for their excellent technical assistance.

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